SCIENTIFIC OPINION

Welfare of domestic birds and rabbits transported in containers

EFSA Panel on Animal Health and Welfare (AHAW),
Søren Saxmose Nielsen, Julio Alvarez, Dominique Joseph Bicout, Paolo Calistri,
Elisabetta Canali, Julian Ashley Drewe, Bruno Garin-Bastuji, Jose Luis Gonzales Rojas,
Christian Gortázar Schmidt, Mette Herskin, Virginie Michel, Miguel Ángel Miranda Chueca,
Barbara Padalino, Helen Clare Roberts, Hans Spoolder, Karl Stahl, Arvo Viltrop,
Christoph Winckler, Malcolm Mitchell, Leonardo James Vinco, Eva Voslarova, Denise Candiani,
Olaf Mosbach-Schulz, Yves Van der Stede and Antonio Velarde

Abstract

This opinion, produced upon a request from the European Commission, focuses on transport of domestic birds and rabbits in containers (e.g. any crate, box, receptacle or other rigid structure used for the transport of animals, but not the means of transport itself). It describes and assesses current transport practices in the EU, based on data from literature, Member States and expert opinion. The species and categories of domestic birds assessed were mainly chickens for meat (broilers), end-of-lay hens and day-old chicks. They included to a lesser extent pullets, turkeys, ducks, geese, quails and game birds, due to limited scientific evidence. The opinion focuses on road transport to slaughterhouses or to production sites. For day-old chicks, air transport is also addressed. The relevant stages of transport considered are preparation, loading, journey, arrival and uncrating. Welfare consequences associated with current transport practices were identified for each stage. For loading and uncrating, the highly relevant welfare consequences identified are handling stress, injuries, restriction of movement and sensory overstimulation. For the journey and arrival, injuries, restriction of movement, sensory overstimulation, motion stress, heat stress, cold stress, prolonged hunger and prolonged thirst are identified as highly relevant. For each welfare consequence, animal-based measures (ABMs) and hazards were identified and assessed, and both preventive and corrective or mitigative measures proposed. Recommendations on quantitative criteria to prevent or mitigate welfare consequences are provided for microclimatic conditions, space allowances and journey times for all categories of animals, where scientific evidence and expert opinion support such outcomes.

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Summary

In the framework of its Farm to Fork Strategy, the European Commission is undertaking a comprehensive evaluation of the animal welfare legislation, including Council Regulation (EC) No 1/2005. The current EU legislation on the protection of animals during transport is based on a scientific opinion adopted in 2002. Against this background, the Commission requested EFSA to give an independent view on the welfare of animals during transport for different groups and categories of farmed animals. It also requested EFSA to propose detailed measures to prevent and/or mitigate the welfare consequences for seven specific scenarios. This Opinion deals with the welfare of domestic birds and rabbits transported in containers. The Commission requested that welfare issues relating to the transport of laying hens (end-of-career animals) to the slaughterhouse would be given specific consideration.

Three transport scenarios are addressed in the Opinion:

- Scenario 1 – Road transport of domestic birds, excluding day-old chicks, to slaughterhouses and production sites. There is a specific emphasis on the welfare issues relating to the transport of chickens for meat (broilers) and end-of-lay hens. Welfare issues in relation to the transport of pullets, turkeys, ducks, geese, quail and game birds were assessed to a limited extent due to scarce scientific evidence.
- Scenario 2 – Road transport and air transport of day-old chicks (recently hatched chickens of less than 72 h) to production sites.
- Scenario 3 – Road transport of rabbits to production sites or slaughterhouses.

The scientific assessment was carried out by breaking down the transport of domestic birds and rabbits in containers into five distinct stages, namely preparation, loading, journey, arrival and uncrating. Each stage was described in terms of current practices. The loading, journey, arrival and uncrating stages were also assessed in terms of welfare consequences. For each welfare consequence, animal-based measures (ABMs) and hazards were identified and assessed, and both preventive and corrective or mitigative measures proposed. Recommendations on quantitative criteria to prevent or mitigate welfare consequences were provided for microclimatic conditions and space allowances during the journey, along with journey times for the categories of animals where scientific evidence and expert opinion supported such outcomes.

Information contained in the scientific articles and reports identified as relevant during a literature search was used as a basis for the text of this Opinion. A public consultation was also carried out. The data obtained from the literature and public consultation were complemented by EFSA expert opinion. This Scientific Opinion follows the guidance protocol that was developed by the Animal Health and Welfare Panel to deal with all the mandates in the context of the Farm to Fork strategy revision (EFSA AHAW Panel, 2022). An uncertainty analysis was also performed.

Data on the transport of live animals are difficult to obtain. In particular, there are no data about movements within the individual Member States, such as transporting animals between farms for growing, breeding or to the slaughterhouse. However, based on production numbers, it is evident that several billion domestic birds are transported in the EU annually. According to TRACES, more than 1.4 billion poultry were moved annually, across Member States (MSs) in 2018 and 2019. Poultry constitutes about 97% of the total intra-EU trade of live animals. Road transport accounted for 99% of total transports of poultry between MSs in 2018 and 2019. On occasions, day-old chicks are transported by air. Extractions from TRACES show that about half of the journeys of poultry reported had a duration of less than 4 h. Poultry, usually day-old chicks, are exported to a variety of countries from the EU, including Albania, Belarus, Egypt, Ghana, Morocco and Ukraine. For rabbits, a recent report from the European Commission highlighted that there were around 180 million farmed rabbits reared for meat consumption in the EU in 2016. Around 119 million (66%) were kept in commercial farms and transported for slaughter. According to TRACES, road transport constituted 99% of total transports of rabbits between MS reported in 2018.

In total, nine welfare consequences were selected as being highly relevant for the welfare of domestic birds during transport based on severity, duration and frequency of occurrence. These were, in order of possible occurrence, (i) handling stress, (ii) injuries, (iii) restriction of movement, (iv) sensory overstimulation, (v) motion stress, (vi) heat stress, (vii) cold stress, (viii) prolonged hunger and (ix) prolonged thirst. Domestic birds may experience one or more negative affective states when exposed to these welfare consequences, including fear, pain, discomfort, distress and frustration. The type of welfare consequence varied depending on the stage and means of transport. For example, cold stress is a particular problem for end-of-lay hens due to the fact that they have minimal body fat and often, they

are poorly feathered owing to wear and tear feather loss in cage systems and to injurious pecking in all systems. The lack of insulation provided by feathers (including the opportunity to fluff the feathers) makes them more sensitive to cold. Specific ABMs were identified for each of the relevant welfare consequences. These include escape behaviours, piling up and vocalisations (alarm calls) for handling stress, wounds for injuries, sitting posture and head posture for restriction of movement, escape attempts and distress calls for sensory overstimulation, panting for heat stress, huddling, fluffing up of feathers and shivering for cold stress. No feasible ABMs for assessing prolonged hunger, prolonged thirst and motion stress were identified. A definition and description of the measure were provided in the Opinion for each ABM. Some ABMs are relevant to more than one welfare consequence. A wide variety of hazards were identified for the different welfare consequences and different journey stages. These were related to factors such as rough handling by operators, acute or sudden auditory or visual stimuli, structural deficiencies of vehicles and facilities, poor driving conditions, unfavourable climatic conditions and poor handling practices. Preventive, corrective and mitigative measures were also provided for each welfare consequence.

Throughout the scientific literature, it is agreed that making sure that animals are fit for transport before departure is of utmost importance. Depending on the duration and quality of a journey, transport can represent significant challenges for domestic birds. The main conditions rendering domestic birds unfit for transport are evident signs of illness, cachexia, severe lameness (unable to stand or walk more than a few steps), open wounds and prolapse, fractures (legs, wings, etc.), dislocations, and wet plumage and poor feather cover in low effective temperature. Wet plumage is not a risk for ducks and geese. End-of-lay hens with poor feather cover are unfit to travel if they are to be transported in cold weather without the application of preventive and corrective measures.

In total, seven welfare consequences were selected as being highly relevant for the welfare of day-old chicks during transport. These were (i) handling stress, (ii) sensory overstimulation, (iii) motion stress, (iv) heat stress, (v) cold stress, (vi) prolonged hunger and (vii) prolonged thirst. As in the case of domestic birds, the type of welfare consequence varied depending on the stage and means of transport. Specific ABMs were identified for each of the relevant welfare consequences. They include posture and orientation on the conveyor belt, falling on the floor, distress calls and escape attempts for handling stress; respiration frequency, mean surface body temperature and cloacal temperature for heat stress; and huddling, distress calls, mean surface body temperature, cloacal temperature, lethargy and respiration frequency for cold stress. A wide variety of hazards were identified for the different welfare consequences and different journey stages. These were related to factors such as rough handling by operators during catching, changes in velocity, drop height, acceleration and speed of conveyor belts, exposure to unexpected loud sounds/noises, exposure to certain visual stimuli such as bright lights, structural deficiencies of vehicles and facilities, poor driving practices or conditions, unfavourable climatic and environmental conditions and poor husbandry practices. Day-old chicks of high genetic value (grandparents, great-grandparents) are transported very long distances and therefore by plane. Air transport contributes to additional hazards that can adversely affect chick welfare before, during and after flights. Possibly due to the lack of environmental control (mainly climatic environment) during the holding stages preceding and following flights (loading/unloading/waiting), these periods can be even more detrimental to the chicks' welfare than the flights themselves.

The welfare consequences selected as being highly relevant for the welfare of rabbits during transport were (i) handling stress, (ii) injuries, (iii) restriction of movement, (iv) sensory overstimulation, (v) motion stress (vi) heat stress, (vii) cold stress, (viii) prolonged hunger and (ix) prolonged thirst. Specific ABMs were identified for each of the relevant welfare consequences. They include escape attempts, piling up and vocalisations for handling stress, wounds and bruises for injuries, sitting posture, lying posture and ear position when sitting for restriction of movement, panting for heat stress and piloerection, shivering and core body temperature for cold stress. No feasible ABMs were identified to assess sensory overstimulation, prolonged hunger, prolonged thirst and motion stress. A wide variety of hazards were identified for the different welfare consequences and different journey stages. These were related to factors such as acute or sudden auditory or visual stimuli, structural deficiencies of vehicles and facilities, poor driving conditions, unfavourable climatic and environmental conditions and poor handling practices. The main conditions making rabbits unfit for transport are evident signs of illness, cachexia, severe lameness (unable to stand or walk more than a few steps), female rabbits in the last third of gestation, female rabbits after parturition, unweaned rabbits, open wounds, prolapses, abscesses, fractures, dislocations, wet fur in low effective temperature. While preventive measures are available for many of the welfare consequences, handling stress cannot be prevented or corrected, only mitigated.

The Opinion contains conclusions and recommendations relating to the highly relevant welfare consequences associated with the transport of domestic birds, day-old chicks and rabbits. For other animal categories such as quail, geese and game birds, available data and evidence in literature are scarce, and therefore, the AHAW Panel considered to which extent the conclusions and recommendations of the most studied species were applicable.

The level of certainty associated with the conclusion is reported, for those conclusions, typically involving quantitative thresholds, which could be the subject of risk management decisions.

In relation to domestic birds:

- Inversion and carrying birds by the legs will increase the severity of handling stress with 90–100% certainty and the risk of injuries (dislocated joints, fractures in legs or wings and bruises) with 66–100% certainty compared to handling birds in an upright position.

- The generic allometric equation 'space allowance (cm^2) = 290 × live weight ($kg^{2/3}$)' can be used to calculate the minimum required floor space during transport for most types of birds to adopt a sitting position and have the possibility to shuffle around. For pullets and end-of-lay hens up to 2 kg, especially when well feathered, more floor space is needed than predicted by using the allometric equation, and therefore, the space allowance derived from planimetric measurements is preferable. Recommended minimum space allowances for the different animal categories are reported in Table 11 (Section 3.7.3.4).

- Several indices based on dry-bulb temperature and relative humidity have been developed to measure effective temperature inside the transport containers. Two of these indices have been validated for poultry, which are the apparent equivalent temperature (AET) and the enthalpy comfort index (ECI) that also combine dry-bulb temperature and relative humidity.

- As regards heat stress, if the AET value is below 40, domestic birds, including end-of-lay hens, will not experience heat stress during transport (safe zone) with 90–100% certainty. Between AET values of 40 and 65, there will be an increasing risk of heat stress (alert zone) and above AET of 65, the birds' mechanisms to cope with heat stress will become less effective and birds are likely to experience heat stress (danger zone). When the ECI index is used, if the ECI remains below 48.0 kJ/kg, with 90–100% certainty, poultry will not experience heat stress during transport (comfort zone). If ECI exceeds this threshold, there will be an increasing risk of heat stress (warning zone). Above 57.6 kJ/kg, the birds' mechanisms to cope with heat stress will become less effective and the birds will experience heat stress (critical zone). For end-of-lay hens, similar ranges of temperature and humidity to those recommended for domestic birds can be used with 66–100% certainty.

- As regards cold stress, if the effective temperature in the containers remains above 10°C, domestic birds will not experience cold stress with 66–100% certainty. For end-of-lay hens, the lower limit of the comfort zone is estimated to be at 18°C with 66–100% certainty. If the temperature falls below this threshold, the hens' mechanisms to cope with cold stress will become less effective and the hens will experience cold stress.

- As regards prolonged hunger, the total feed withdrawal starts when feed is removed on farm and ends when all animals are removed from containers following unloading from the vehicle and fed or slaughtered. Domestic birds, excluding end-of-lay hens, subjected to feed withdrawal periods longer than 6 h will exhaust liver glycogen reserves, and therefore, the AHAW Panel concluded with > 50–100% certainty that they will experience prolonged hunger. Furthermore, the AHAW Panel concluded with 90–100% certainty that domestic birds subjected to feed withdrawal periods longer than 12 h will experience prolonged hunger as well as intestinal cell breakdown, which is detrimental to their welfare.

- End-of-lay hens are at particular risk of experiencing prolonged hunger as a consequence of being metabolically exhausted with few body reserves at the end of economic production. The AHAW Panel concluded with 66–100% certainty that, under thermoneutral conditions, end-of-lay hens will experience prolonged hunger after feed withdrawal of 10 h. As regards prolonged thirst, the AHAW Panel concluded with 66–100% certainty that domestic birds subject to water deprivation periods longer than 6 h will experience prolonged thirst. Domestic birds subject to water deprivation periods longer than 12 h may show an increase in plasma creatinine; the AHAW Panel therefore concluded with 90–100% certainty that they will experience prolonged thirst, which is detrimental to their welfare. End-of-lay hens are at particular risk of experiencing prolonged thirst as they are still laying eggs of high-water content following 12 h of water deprivation (90–100% certainty).

- For ducks, geese, quails and game birds, there is limited scientific evidence of the consequences of water withdrawal; the AHAW panel, therefore, concluded with 66–100% certainty that their thirst experience will be similar to other domestic poultry.
- Not much evidence exists on the maximum journey duration. However, as domestic birds cannot be fed in the containers, the maximum journey duration including on-farm feed withdrawal should not exceed 12 h (10 h in case of end-of-lay hens).
- DOA is a relevant iceberg indicator for the assessment of welfare during transport. The cause of DOA should be investigated when it exceeds 0.1% in all domestic birds.

In relation to day-old chicks:

- Changes in velocity that exceed 0.4 m/s, a drop height above 280 mm and speed of belts over 27 m/min will generate handling stress (with 66–100% certainty), but there are no published data about specifications to completely avoid handling stress.
- Transport of fertilised eggs and on-farm hatching will avoid handling stress as well as the other welfare consequences associated with transport.
- The upper limit of the comfort zone of day-old chicks is estimated to be 35°C (near the chicks). Below this threshold, day-old chicks will not experience heat stress during transport (comfort zone) (with 66–100% certainty). The lower limit of the comfort zone of day-old chicks is estimated to be at 30°C. If the effective temperature is below this threshold, the chicks will experience cold stress.
- Day-old chicks subject to feed and water withdrawal periods longer than 48 h will be at risk of experiencing severe prolonged hunger (with 90–100% certainty) and thirst, which is detrimental to their welfare.
- Provision of feed and water during transport (in the boxes) is also a way to prevent or mitigate prolonged hunger and thirst.
- DOA is a relevant iceberg indicator for the assessment of welfare during transport. The cause of DOA should be investigated when it exceeds 0.1% in day-old chicks.

In relation to rabbits:

- The planimetric measures when the rabbits are in ventral recumbency, front legs extended and hind legs bent to the body and the allometric equation 'space allowance (cm^2) = 270 × live weight (kg$^{2/3}$)' provide the minimum required floor space for rabbits to adopt a ventral recumbency and sitting–resting position and with the possibility to change position.
- The height of transport containers should allow the rabbits to keep their ears erect in a natural position while sitting. The AHAW Panel concluded with 66–100% certainty that crate heights of 35 and 40 cm will, respectively, ensure slaughter rabbits (up to 3 kg) and breeding rabbits (between 4.5 kg and 6 kg) can sit with their ears erect.
- As regards heat stress, if the temperature–humidity index (THI) remains below 27.8, rabbits will not experience heat stress during transport (safe zone). Between THI values of 27.8 and 28.9, there will be an increasing risk of heat stress (alert zone) and above THI of 28.9, the rabbit's mechanisms to cope with heat stress will become less effective and the rabbits will experience heat stress (danger zone) with a 66–100% certainty.
- As regards cold stress, below 10°C, the rabbit's mechanisms to cope with cold stress will become less effective and the rabbits will experience cold stress with a 66–100% certainty.
- As regards prolonged hunger, the total feed withdrawal starts when feed is removed on farm and ends when all animals are removed from containers following unloading from the vehicle and fed or slaughtered. The AHAW Panel concluded with > 50–100% certainty that rabbits subjected to feed withdrawal periods longer than 6 h will experience prolonged hunger, and those subjected to feed withdrawal periods longer than 12 h will lose weight and will experience prolonged hunger with 66–100% certainty.
- As regards prolonged thirst, the AHAW Panel concluded with 66–100% certainty that rabbits subjected to water deprivation periods longer than 12 h will experience prolonged thirst.
- Not much evidence exists on the maximum journey duration. However, as rabbits cannot be fed in the containers, the maximum journey duration including on-farm feed withdrawal should not exceed 12 h.
- DOA is a relevant iceberg indicator for the assessment of welfare during transport. The cause of DOA should be investigated when it exceeds 0.1% in rabbits.

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1. Introduction

1.1. Background and Terms of Reference as provided by the requestor

In the framework of its Farm to Fork strategy, the Commission will start a comprehensive evaluation of the animal welfare legislation. This will include the following acts:

1) Council Directive 98/58/EC of 20 July 1998 concerning the protection of animals kept for farming purposes;
2) Council Directive 1999/74/EC of 19 July 1999 laying down minimum standards for the protection of laying hens;
3) Council Directive 2008/119/EC of 18 December 2008 laying down minimum standards for the protection of calves;
4) Council Directive 2008/120/EC of 18 December 2008 laying down minimum standards for the protection of pigs;
5) Council Directive 2007/43/EC of 28 June 2007 laying down minimum rules for the protection of chickens kept for meat production;
6) Council Regulation (EC) No 1/2005 of 22 December 2004 on the protection of animals during transport and related operations and amending Directives 64/432/EEC and 93/119/EC and Regulation (EC) No 1255/97;
7) Council Regulation (EC) No 1099/2009 of 24 September 2009 on the protection of animals at the time of killing.These acts are based on scientific opinions that are outdated. The current EU legislation on the protection of animals during transport is based on a scientific opinion adopted in 2002. Since then the EFSA adopted opinions in 2004 (two opinions) and 2011. In the context of possible drafting of legislative proposals, the Commission needs new opinions that reflect the most recent scientific knowledge.

Against this background, the Commission would like to request the EFSA to review the available scientific publications and possibly other sources to provide a sound scientific basis for future legislative proposals.

This request is about the protection of terrestrial animals during transport.

The Commission therefore considers opportune to request EFSA to give an independent view on the protection of animals during transport for the following groups and categories of farmed animals:

Free-moving animals (group 1):

1) Equids (horses, donkeys and their crossings),
2) Bovine animals (cattle and calves),
3) Small ruminants (sheep and goats),
4) Pigs;

Animals in containers (group 2):

5) Domestic birds (chickens for meat, laying hens, turkeys, ducks, geese, quails, etc.).
6) Rabbits.The request refers to any journey, i.e. journeys of less than 8 h ('short journeys'), journeys of more than 8 h ('long journeys') and long journeys that need unloading and/or feeding ('very long journeys').

For each category of animals (1–6), the EFSA will describe, based on existing literature and reports, the current practices regarding:

a) the preparation for transport (including catching and crating of poultry and rabbits), loading, unloading and handling of animals at all stages of the journey, including at destination;
b) the means of transport by road, roll-on-roll-off vessels, livestock vessels, the means of transport by rail and by air;
c) the conditions within the means of transport: space, microclimatic conditions, watering and feeding;
d) the journey duration and its circumstances as well as the resting of animals in the vehicle being stationary or being unloaded;
e) the conditions for areas where animals are unloaded and/or grouped as part of the journey (assembly centres, livestock markets, control posts, EU ports);

Additionally, for each of the above practices, the EFSA will:

- Describe the relevant welfare consequences for each category of animals during each step of the process. Relevance will not need to be based on a comprehensive risk assessment, but on EFSA's expert opinion regarding the severity, duration and occurrence of each welfare consequence.
- define qualitative or quantitative measures to assess the welfare consequences during transport (animal-based measures);
- identify the hazards leading to these welfare consequences;
- provide recommendations to prevent, mitigate or correct the welfare consequences (resource and management-based measures).

For the following scenarios, the Commission has identified practical difficulties or insufficient information in ensuring the welfare of animals. At least for them, the EFSA is asked to propose detailed animal-based measures and preventive and corrective measures with, where possible, either qualitative (yes/no question) or quantitative (minimum/maximum) criteria (i.e. requirements to prevent and/or mitigate the welfare consequences):

1) 'Export by livestock vessels' – Transport of adult cattle, weaned calves and sheep over long journeys involving the combination road/livestock vessels;
2) 'Export by road' – Transport of adult cattle, weaned calves and sheep over long journeys by road involving the use of facilities where animals are unloaded and reloaded (control posts, livestock markets) or when animals are kept in stationary vehicles for hours (exit points) including in third countries;
3) 'Roll-on–roll off' – Transport of adult cattle, calves and sheep over long journeys involving the combination road/roll-on-roll-off vessels;
4) 'End-of-career animals' – Transport of end of career animals to slaughterhouses of dairy cows, breeding sows and laying hens;
5) 'Un-weaned calves' – Transport of un-weaned calves over long journeys; this scenario will particularly consider the risks regarding fitness for transport, watering, feeding and thermal comfort under Section c of the current practices associated with inappropriate drinkers and liquid feed for un-weaned calves;
6) 'Horses' – Transport of horses on long journeys to slaughterhouses;
7) 'Special health status animals' – Transport of ruminants and pigs where unloading them before the final destination might jeopardise their health status.

For all scenarios, the EFSA will consider the risks regarding microclimatic conditions under Section c of the current practices associated with extremely high or low temperatures including the difficulty of measuring of temperature, humidity and gas concentration within animals' compartment.

1.2. Interpretation of the Terms of Reference

In the framework of its Farm to Fork strategy, the European Commission has started a comprehensive evaluation of the animal welfare legislation and has asked EFSA to review the available scientific publications and possibly other sources to provide a sound scientific basis for future legislative proposals on the protection of animals during transport.

This scientific opinion covers the animals transported in containers (group 2) of the term of reference (ToR), namely domestic birds and rabbits kept for farming purposes. A 'container' is defined as any crate, box, receptacle or other rigid structure used for the transport of animals, which is not a means of transport. The crates, cages, container drawers and cardboard boxes used for transport as described in this opinion are all considered to be containers.

For the animals covered by the mandate, the keeping of these animals on farm, killing and slaughter is not part of the request and so these practices will not be covered. The domestic birds mentioned in the mandate refer to 'chickens for meat, laying hens, turkeys, ducks, geese and quails, etc.', meaning that the list of species is not exhaustive. In addition to the categories of domestic birds specifically mentioned, when information is available, EFSA will consider also farmed game birds, such as pheasants and partridges due to the fact that within the EU, these birds are farmed and transported, mainly to be released for hunting purposes. Both 'domestic birds' and 'poultry' are used in this scientific opinion, referring to the same animals. Newly hatched chicks of the species covered will be considered as one animal category, referred to as day-old chicks.

The European Commission requested EFSA to assess the current practices of transporting domestic birds and rabbits by road, roll-on–roll-off vessels, livestock vessels, rail and air in the EU at all stages of the journey, including at destination.

During their lives, domestic birds and rabbits are usually transported to other production sites or to slaughterhouses. Based on available knowledge of production of domestic birds and rabbits in the EU (summarised in Section 3.1 and 5.1, respectively), it is evident that transport of these animals is mainly by road and within a Member State. Air transport of day-old chicks to many parts of the world is also common as it constitutes the majority of EU exports of domestic birds to third countries. A small fraction of the transports reported in TRACES is by sea (0.58%), mainly by roll-on–roll-off vessels as part of road transport. Transport by rail also occurs, but compared to road and air, the extent of this mode of transport is very limited and poorly documented.

The following **transport scenarios** (combination of category of animal and type of transport) were identified by EFSA as relevant current practices to be addressed in this scientific opinion:

Scenario 1:

- Road transport of animals kept for meat production to slaughterhouses

    o chickens for meat (broilers, capons, cockerels)
    o turkeys
    o ducks
    o geese
    o quail
    o game birds

- Road transport of animals kept for breeding or egg production, to production sites or slaughterhouses

    o breeders of the species mentioned above;
    o pullets of laying hens and quail kept for egg production;
    o end-of-lay hens (in the mandate referred to as end of career laying hens); and end-of-lay quail.

- Road transport of game birds such as pheasants and partridges to be released for hunting purposes

Scenario 2:

- Road transport and air transport of day-old chicks (recently hatched chickens of less than 72 h) of the domestic birds mentioned above to production sites.

Scenario 3:

- Road transport of rabbits to production sites or slaughterhouses

For some scenarios, the European Commission requested EFSA to propose detailed animal-based measures and preventive and corrective measures with, where possible, either qualitative (yes/no question) or quantitative (minimum/maximum) criteria (i.e. requirements to prevent and/or mitigate the welfare consequences). Of the specific scenarios listed by the requestor, only number 4 describing 'end-of-career animals' is considered relevant for this scientific opinion and concerns the end-of-lay hens. However, due to the number of animals affected and the current state of transport legislation, EFSA will provide, when possible, recommendations on quantitative criteria also for other species covered by the mandate, such as broilers, rabbits and day-old chicks.

Scientific publications on the domestic birds covered by this scientific opinion refer mainly to broilers, and to a less extent to turkeys, end-of-lay hens, ducks and day-old chicks. For other animal categories such as quail, geese, capons, cockerels and game birds, available data and evidence in literature are scarce and do not provide sufficient scientific support for species-specific conclusions and recommendations. For these species, the AHAW Panel will consider to which extent the conclusions and recommendations of the most studied species are applicable.

Scientific evidence and reports on the welfare of rabbits during transport relate only to the transport of animals for slaughter. No data are available on the impact of transport practices on rabbits transported for other purposes. However, extrapolation was made to transport of farmed rabbits to production sites.

Transport is the movement of animals effected by one or more means of transport and the related operations, including loading, unloading, transfer and rest, until the unloading of the animals at the place of destination is completed. However, as animals transported in containers in most cases will experience the welfare consequences of transport until they are removed from the containers, in this scientific opinion, transport is considered to end when all animals have been removed from the containers. Thus, the total time in the containers will be taken into consideration in the assessment of welfare consequences.

The mandate refers to journeys of less than 8 h as 'short journeys', journeys of more than 8 h as 'long journeys' and journeys that need unloading and/or feeding as 'very long journeys'. For the animals of group 1 of this mandate, transport practices differ between these journeys, e.g. related to vehicle equipment and need for control posts. These practices do, however, not apply to domestic birds and rabbits as vehicles are not equipped with drinking devices and do not stop in control posts. For this reason, the categorisation of journeys as 'short', 'long' and 'very long' as described in the mandate will not be applied in this scientific opinion.

The scientific opinion focuses on the highly relevant **welfare consequences** that were identified for each transport scenario (combination of category of animal and type of transport). Species and categories of domestic birds were merged as appropriate when the same welfare consequences were identified as highly relevant. As a result, three chapters (scenarios) have been developed, for domestic birds, day-old chicks and rabbits.

In each chapter, current practices relevant to the different stages are described. The relevance of each welfare consequence is described, and for each of these welfare consequences, animal-based measures (ABMs) considered feasible for inspection purposes identified and defined, and the measurement, sensitivity and specificity qualitatively assessed. Relevant measures to prevent and correct the hazards or to mitigate the welfare consequences are also identified. The identification of preventive measures is of particular importance in the transport of animals, as corrective measures are often not possible to implement during the course of a journey.

In some circumstances, valid and reliable ABMs may not exist, or are not feasible in the context of transport (e.g. if all or some of the animals in a vehicle cannot be properly inspected). If welfare consequences are identified for the visible animals, it is plausible that other animals on the vehicle are also affected to a lesser or larger degree, depending on the position of the container in the vehicle. In this type of situation, particular emphasis is given to the presence of hazards. If certain hazards are present, it can be assumed that the animals experience the related welfare consequences.

In this opinion, journey duration is considered as the period the vehicle is in transit until destination is reached. Transport duration considers the time the animals are in the containers and the feed withdrawal period on farm. Therefore, the opinion refers to the maximum transport duration instead of journey duration.

Dead on arrival (DOA) is the most widely used ABM, scientifically and in practice, when assessing the overall consequence of transport on the welfare of poultry and rabbits. DOA is an iceberg indicator linked to several welfare consequences and stages of transport, but it can only be assessed when uncrating animals. It will be covered in a dedicated section of each chapter.

EFSA's scientific opinion will ascertain the welfare consequences for animals for a given practice, but not whether current practices are acceptable, as this is the function of the risk manager.

2. Data and methodologies

2.1. Data

2.1.1. Data from literature

Information from the papers selected as relevant from the literature search described in Section 2.2.1 and from additional literature identified by the EFSA experts was used for a narrative description and assessment to address the ToRs (see relevant Sections 3, 4 and 5).

2.1.2. Information from the public, Member States and expert opinion

To consult interested parties and gain feedback on EFSA's interpretation of the transport mandate (free-moving animals and animals transported in containers), a public consultation was launched in the period from 15 April 2021 to 10 June 2021. In particular, EFSA called for interested parties to:

- identify current transport practices of particular concern not already identified by EFSA in the interpretation of mandate;
- describe the practical difficulties or insufficient information in ensuring the welfare of animals, for the specific transport practices listed in the request from the European Commission and for any other additional practices of concern that might be identified;
- provide any available recorded data from road or sea transport, e.g. from a data logger, related to the microclimatic environment (temperature, humidity and ammonia levels). The data should demonstrate a link between the microclimatic conditions and any adverse welfare consequences that are experienced by the animals during transport.

The information received in the public consultation were considered by the EFSA experts as part of their work on this scientific opinion (See Annex A: Report of the Public Consultation on the Protection of Animals during Transport, published under 'Supporting Information' in the Opinion on transport of small ruminants).

Information on the extent and nature of transport of end-of-lay hens in the EU Member States (MSs), Norway and Switzerland were requested by EFSA to the National Contact Points (NCPs) for Council Regulation (EC) No 1099/2009 Network representatives during October–December 2020. In total, eight countries replied to the request. This material was not sufficient to provide a comprehensive overview of current practices, although the information provided has been reported when considered useful to describe current practices.

Information on production, the extent and nature of transport of ducks, geese and quail was requested in collaboration with the European Commission in an EU Survey to the MSs, Norway and Switzerland. The survey was open online from August to December 2021. All 27 MSs, Norway and Switzerland replied to the survey. The results were analysed and the result provided some relevant information on transport of ducks, geese and quail in some MSs. Due to the variability in the data provided, it was not possible to generate a comprehensive overview of current practices. However, information from this survey has been used to describe current transport practices for these species.

2.2. Methodologies

2.2.1. Protocol for the Farm to Fork mandates

This scientific opinion follows the protocol detailed in the methodological guidance that was developed by the AHAW Panel to deal with all the mandates in the context of the Farm to Fork strategy revision (EFSA AHAW Panel, 2022).

According to the protocol, EFSA translated the assessment questions into more specific subquestions. These are interrelated, meaning that the outcome of each subquestion is necessary to proceed to the next subquestion. The approach to develop the subquestions is based on using both evidence from the scientific literature and expert opinion. The translation of the assessment questions into subquestions is mapped in Table 1.

Table 1: Overview of the translation of the assessment questions into subquestions

Assessment questions	Subquestions	
i. **Describe the current transport practices**	**1.** *Identify and select all relevant transport scenarios (animal transport practices per species and animal category)*	**2.** *Describe the transport practices*
	Aim: Animal transport practices to be considered in the assessment are identified and selected to be representative of the current practice in the EU. Approach: expert opinion via group discussion. Relationship with assessment question: This subquestion is necessary for the overall assessment question requiring the description of the practices.	Aim: All the animal transport practices per animal category identified and selected from subquestion 1 are described narratively. Approach: literature review. Relationship with assessment question: this corresponds to the assessment question and is necessary for the next assessment question.
ii. **Describe the relevant welfare consequences that may occur due to the practices**	**3.** *Identify the welfare consequences common for all mandates and provide their definitions*	**4.** *Select the highly relevant welfare consequences for each animal transport practice*
	Aim: To identify the welfare consequences that may impair the welfare of animals, and to provide a definition for them. EFSA generates a list of welfare consequences common for all mandates. Approach: expert opinion via group discussion. Relationship with assessment question: The list of all possible welfare consequences is necessary for the next assessment question asking to identify the highly relevant ones per each system.	Aim: To select the highly relevant welfare consequences for each of the previously defined animal transport scenarios per species and animal category. Approach: expert opinion via EKE. Relationship with assessment question: This corresponds to the assessment question, is related to subquestion 1 in which relevant welfare consequences are identified only for current transport scenarios.
iii. **Define qualitative or quantitative animal-based measures (ABMs) to assess these welfare consequences**	**5.** *Identify feasible ABMs for the assessment of the highly relevant welfare consequences*	**6.** *Describe feasible ABMs for the assessment of the highly relevant welfare consequences*
	Aim: The ABMs for the assessment of the welfare consequences previously identified as relevant are selected (only for feasible ABMs). Approach: expert opinion via group discussion. Relationship with assessment question: this corresponds to the assessment question and is related to subquestion 4 in which ABMs are identified only for the highly relevant welfare consequences.	Aim: The ABMs for the assessment of the welfare consequences previously identified as the highly relevant are described. Approach: literature review. Relationship with assessment question: related to subquestion 5.

Assessment questions		Subquestions	
iv.	Identify the hazards leading to these welfare consequences	*7. Identify the hazards leading to the highly relevant welfare consequences*	*8. Describe the hazards leading to the highly relevant welfare consequences*
		Aim: The hazards leading to the highly relevant welfare consequences are identified.	Aim: The hazards are described.
		Approach: expert opinion via group discussion.	Approach: literature review.
		Relationship with assessment question: this corresponds to the assessment question and is related to subquestion 4 in which hazards are identified only for the highly relevant welfare consequences.	Relationship with assessment question: related to subquestion 6.
v.	Provide recommendations to prevent, mitigate or correct the hazards	*9. Identify the preventive and corrective measures for the highly relevant welfare consequences*	*10. Describe the preventive and corrective measures for the highly relevant welfare consequences*
		Aim: Preventive and corrective measures for the highly relevant welfare consequences for the previously defined transport scenarios per animal category are identified.	Aim: Preventive and corrective measures are described.
		Approach: expert opinion via group discussion.	Approach: literature review.
		Relationship with assessment question: This corresponds to the assessment question and is related to subquestion 4 in which preventive and corrective measures are identified only for the highly relevant welfare consequences.	Relationship with assessment question: related to subquestion 8.

The specific protocol for the assessment of each of the subquestions listed above is presented in Annex 1. Evidence needs and methods used for answering each of the subquestions are presented separately depending on whether they are based on expert opinion or data extracted from literature reviews.

2.2.2. Experts' opinion

As described in the above Table 1, expert opinion was used for the subquestions requiring the identification of transport scenarios, welfare consequences, ABMs, hazards, preventive and corrective or mitigative measures.

Expert opinion was mainly elicited via EFSA expert group discussion. Only for the identification of highly relevant welfare consequences, a structured Expert Knowledge Elicitation (EKE) was carried out.

The mandates request the identification of the highly relevant welfare consequences for each of the defined animal transport scenarios (Subquestion 4). This identification of the highly relevant welfare consequences is executed via expert opinion. Hereto, the opinion of the EFSA experts is elicited through an exercise of individual classification of welfare consequences in terms of relevance followed by group discussion to identify the highly relevant ones by consensus.

The starting point is the list of 33 specific welfare consequences identified under Subquestion 3 (for details, see Section 3.1.1.3 of the methodological guidance, EFSA AHAW Panel, 2022). The exercise is carried out separately for each of the animal transport stages per transport scenario resulting from Subquestion 1.

The exercise consists in selecting the highly relevant welfare consequences out of these 33 for each of these combinations.

For each combination, the EFSA experts classify, based on an estimate of their magnitude, the 33 welfare consequences into four categories of relevance: (i) non-applicable, e.g. the welfare consequence 'Inability to perform suckling behaviour' is not considered relevant for the animal categories covered by the mandate, (ii) slightly relevant, e.g. the welfare consequence 'inability to perform play behaviour' might apply, but its effect on welfare (in terms of prevalence and/or severity) was considered to be minimal compared to other welfare consequences of the transport scenarios, (iii) moderately relevant and (iv) highly relevant. The magnitude of a welfare consequence is defined as the product of three parameters (severity, duration and frequency of occurrence) (EFSA AHAW Panel, 2022). Severity refers to the intensity of the welfare consequence. Duration refers to the time an animal spends within a transport scenario while the occurrence refers to the prevalence of animals experiencing the welfare consequence in that transport scenario.

Owing to the lack of published data on these three parameters, the EFSA experts express their qualitative expert opinion on the magnitude of welfare consequences.

Expert opinion is elicited in three phases:

1) First phase: The experts go individually through the list of welfare consequences and identify those that would fall in the 'non-applicable' or 'slightly relevant' categories. Their individual judgements are then collated, and those welfare consequences unanimously identified as belonging to these two categories are removed and not considered for further assessment. Those welfare consequences for which there is no consensus whether they are considered 'non-applicable' or 'less relevant' remain for further assessment and require an open group discussion to find a consensus.

2) Second phase: The experts go individually through the list of remaining welfare consequences and identify those that would fall in the category of 'highly relevant' in order to identify only the highly relevant welfare consequences that are kept for further assessment procedure (Subquestion 5 Section 2.2.1). Similarly, as during the first phase in case discrepant opinions emerge, consensus is sought through group discussion.

3) Third phase: The experts are asked to rank individually all the remaining welfare consequences in the list that are not already identified as highly relevant (and thus kept) or non-applicable or slightly relevant (and thus removed) from the highest to the least relevant. Their individual rankings are then discussed again in an open group discussion with the aim to assign the remaining welfare consequences into the category 'highly relevant' or in the category 'moderately relevant'.

The scientific opinions only report, for each of the defined animal transport scenario(s), those welfare consequences that are selected to be highly relevant from this exercise.

2.2.3. Literature searches

As described in the above Table 1, literature searches were carried out for the subquestions requiring the description of transport scenarios, welfare consequences, ABMs, hazards, preventive and corrective or mitigative measures.

First broad searches for literature providing information on current practices on transport for domestic birds (all categories) and rabbits were carried out. Restrictions were applied in relation to the date of publication, considering only those records published after a previous EFSA opinion on the topic (EFSA AHAW Panel, 2011).

Following the broad searches, more specific searches were carried out focusing on welfare consequences, ABMs, hazards, preventive and corrective or mitigative measures.

For broilers, the search results (broad + specific) yielded a total of 815 (169 + 646) records that were exported to an EndNote library together with the relevant metadata (e.g. title, authors, abstract). Titles and abstracts were firstly screened to remove irrelevant publications (e.g. related to species, processes and research purposes that were out of scope of this scientific opinion) and duplicates, and successively to identify their relevance to the topic. The screening led to 258 relevant records. Experts screened these papers and decided upon 75 references for further assessment. The full text of these papers was retrieved and made available to the experts.

For laying hens, the search results (broad + specific) yielded a total of 256 (76 + 180) records that were reduced to 119 after screening for relevance and further to 33 for further assessment.

For ducks, geese, turkeys and quail, a common search resulted in a total of 353 (48 + 305) records that were reduced to 51 after screening for relevance and further to 29 for further assessment.

For game birds, the general and specific search was combined, yielding a total of 336 records that were reduced to 43 after screening for relevance and further to 28 for further assessment.

For day-old chicks, the search results (broad + specific) yielded a total of 588 (136 + 452) records that were reduced to 18 after screening for relevance and further to 14 for further assessment.

For rabbits, the search results (broad + specific) yielded a total of 923 (22 + 901) records that were reduced to 47 after screening for relevance and further to 44 for further assessment.

Full details of the literature search protocol are provided in Appendix A.

The searches were saved in Web of Science and relevant results (records) appearing at a later stage were screened and added to the pool of papers available to the experts. In addition, the experts selected relevant references starting from scientific papers, including review papers, books chapters, non-peer-reviewed papers known by the experts themselves or retrieved through non-systematic searches, until the information of the subject was considered sufficient to undertake the assessment by the EFSA experts. If needed, relevant publications before 2011 were considered.

2.2.4. Uncertainty analysis

The uncertainty in the assessment performed for this Scientific Opinion was investigated in a qualitative manner following the procedure detailed in the EFSA guidance on uncertainty analysis in scientific assessments (EFSA Scientific Committee, 2018a,b). The outcome of this Scientific Opinion is the identification and description of the highly relevant welfare consequences, the related animal-based measures (ABM) – which may be measured in a qualitative or quantitative way – and hazards causing these welfare consequences per each stage of transport and per each animal category. Based on the identification of welfare consequences and ABMs, conclusions and recommendations are formulated allowing different mitigation and preventing measures for the identified welfare consequences.

The Panel agreed to tackle the uncertainty related to the methodology employed to identify welfare consequences, ABMs and related hazards by describing the potential sources. A table describing the sources of uncertainty is presented in Annex B.

Regarding the overall impact of uncertainties on the developed conclusions of the opinion, it was agreed to perform an assessment only for a subset of key conclusions, typically involving quantitative thresholds, which could be the subject of risk management decisions.

These conclusions were rephrased in order to refer to well-defined questions of interest related to an animal category (e.g. day-old chicks), a welfare consequence (e.g. cold stress) and a hazard (e.g. too low effective temperature), so that they adopted the general form 'how likely is that 90% or more of all animals belonging to a specific animal category traveling in the next year will experience a specific welfare consequence when a specific hazard is present'. In some cases, the inverse formulation (i.e. how likely is that 90% or more of all animals belonging to a specific animal category traveling in

the next year will **not experience** a specific welfare consequence when a specific hazard **is not present**) was preferred. The 90% threshold was selected to ensure that conclusions were referring to situations in which a large proportion (≥ 90%) of the animals for each target animal category would experience/not experience the welfare consequence as the presence/absence of the hazard. Experts were then asked to provide their individual judgement on the certainty for each question of interest according to three predefined agreed certainty ranges (Table 2 here below), which are derived from the approximate probability scale from the guidance on uncertainty (EFSA, 2019).

Table 2: Three ranges used to express agreed (consensus) certainty around conclusions

Quantitative assessment	Certainty range		
	> 50–100%	66–100%	90–100%
Qualitative translation	More likely than not	From likely to almost certain	From very likely to almost certain

Experts were asked to individually identify the certainty range better reflecting their degree of certainty for each conclusion, and then, a group discussion was held during which they had the chance to explain the rationale behind their judgement; finally, a consensus on which range better reflected the overall certainty was reached and, if no consensus was achieved, the wider range encompassing all individual judgements was selected.

3. Assessment of scenario 1: Road transport of domestic birds

3.1. Introduction

Domestic birds as described in this chapter include broilers, turkeys, pullets, laying hens, ducks, geese, quail and game birds.

Information on the production of poultry in the EU is mainly taken from the DG Agri official website on this topic (EC Poultry Meat Dashboard, 2022). Here, the EU production of poultry is estimated at 13,741,000 t of poultry meat in 2021. Of this, broilers constituted 82%, turkeys 14%, ducks 3% and others 1%. Poland is the largest poultry producer, followed by France, Spain, Germany, the Netherlands and Italy. Together, these six MSs produce 74% of the total poultry meat in the EU.

More than 6.3 billion broilers were slaughtered in the EU in 2020. The highest producing Member State was Poland (~ 1.2 billion), followed by France (~ 770 million), Spain (~ 696 million) and Germany (~ 623 million), the Netherlands (~ 598 million) and Italy (~ 574 million).

For turkeys, the total number slaughtered in 2020 was ~ 188 million. The highest producing Member State was Poland (~ 41 million), followed by France (~ 39 million), Germany (~ 35 million), Italy (~ 29 million) and Spain (~ 27 million).

For ducks, the number of birds slaughtered was reported to be ~ 134 million, with the highest production in France (~ 61 million), Poland (~ 27 million) and Hungary (~ 24 million).

Numbers on production of geese were only retrieved up to 2008, where approx. of 12.5 million geese were reported slaughtered, with almost all production situated in the two MSs Poland (~ 6.8 million) and Hungary (~ 5.1 million). 481,000 geese were reported slaughtered in France.

In 2020, a total of 372.4 million laying hens were reported to be kept in the EU (European Commission official website on Food, Farming and Fisheries[1]). Germany, Poland, France, Spain, Italy and the Netherlands were the biggest producers, and together these five MSs kept 63% of laying hens in the EU. At the end of production, between 52 and 100 weeks of age depending on breed and production system, laying hens are either killed on farm or transported to the slaughterhouse. Information on the extent and nature of transport of end-of-lay hens in the EU MSs, Norway and Switzerland was requested by EFSA to the National Contact Points (NCPs) for Council Regulation (EC) No 1099/2009 Network representatives during October–December 2020. Eight countries replied to the request. Of these, Italy, Portugal, Belgium, Czech Republic and Finland informed that all or nearly all end-of-lay hens were transported to slaughterhouses. Two countries reported that a proportion of the end-of-lay hens were killed on farm, Cyprus (26.4% killed on farm) and Switzerland (23% killed on farm). Slovenia reported lack of data on this topic.

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[1] https://ec.europa.eu/info/food-farming-fisheries/animals-and-animal-products/animal-products/en (Accessed: 4 February 2022).

Eurostat categorises quail as 'small poultry' together with several other species, making estimates on production difficult. In the survey to the EU MSs, Norway and Switzerland, 23 countries reported to have the production of quail. The total number of animals reported was ~ 20 million birds. Of these, Italy produced 13.7 mill, France 4 mill and Portugal 1.7 mill birds, meaning that for the remaining countries, production was very low.

The release of captive-reared individuals into hunting grounds is one of the most widely used gamebird management techniques. Although it is not possible to estimate with certainty the extent of this practice, it is likely that at least several million gamebirds are introduced each year in the European Union (Arroyo and Beja, 2002). In France, 15–20 million pheasants are released into nature every year. Releases may have two different objectives, with a different timescale: reinforcement of breeding numbers (long-term objectives) or increase of bird numbers during the hunting season (short-term objectives) (Arroyo and Beja, 2002).

A recent briefing report from the European Parliament (EPRS, 2021) concluded that data on the transport of live animals are difficult to obtain. In particular, there are no data about movements within the individual MSs, such as transporting animals between farms for fattening, breeding or to the slaughterhouse, although domestic transport [within a Member State] accounts for the majority of the transport of live animals in the EU. The majority of domestic birds kept for the production of meat and/or eggs are transported twice in their life; as newly hatched chicks from the hatchery to the rearing farm and later to the slaughterhouse (Mitchell, 2006a,b; Mitchell and Kettlewell, 2009). Pullets of laying hens and breeder poultry are also transported from the rearing house to the egg production site (layer farm). Based on production numbers, it is evident that several billion domestic birds are transported in the EU annually.

Transport of animals between MSs is registered in TRACES. According to TRACES (data extracted in 2020), more than 1.4 billion poultry were traded, and thereby transported, between MSs in 2018 and 2019 (see Table 3). According to EPRS, poultry constitute about 97% of the total intra-EU trade of live animals. The numbers of domestic birds reported in TRACES in 2018 and 2019 is summarised in Table 4.

Table 3: Numbers of domestic birds transported between EU MSs in 2018 and 2019 (EFSA extraction from TRACES)

Poultry category	Year	
	2018	**2019**
Poultry Weighing Less than 185 g (i.e. chicks)	891,770,887	887,808,427
Chicken (*Gallus gallus domesticus*)	516,271,515	519,937,815
Turkeys	13,598,262	13,354,465
Ducks	4,226,243	4,870,497
Geese	493,208	615,247
Total	1,426,360,115	1,426,586,451

3.1.1. Means of transport reported in TRACES

Extractions from TRACES (Table 4) show that road transport constituted 99% of total transports of poultry between MS reported in 2018 and 2019. Transport within MSs is likely similarly by road. Roll-on–roll-off vessel is the second most reported means of transport, and these vessels normally constitute part of road transport. Transport by air (aeroplane), mainly of day-old chicks, has some prevalence, while transport by ship, rail and 'other' constitutes a minor fraction of the total number of transported birds.

Table 4: Number of domestic birds transported between EU MSs by different means of transport in 2018 and 2019 (EFSA extraction from TRACES)

Means of transport	Year			
	2018		**2019**	
Aeroplane	3,481,675	0.24%	4,927,270	0.35%
Other	198,057	0.01%	239,062	0.02%
Railway wagon	24,724	0.00%	29,434	0.00%
Road vehicle	1,414,389,031	99.16%	1,413,489,336	99.08%

Means of transport	Year			
	2018		**2019**	
Ship	29,600	0.00%	27,680	0.00%
Ship/Railway wagon	19,111	0.00%	14,920	0.00%
Roll-on–roll-off	8,230,364	0.58%	7,887,370	0.55%
Total	1,426,372,562	100%	1,426,615,072	100%

3.1.2. Journey times reported in TRACES

Extractions from TRACES (Figure 1) show that about half of the journeys of poultry reported in 2018 and 2019 had a duration of less than 4 h. This is based on estimated journey times (excluding loading and unloading) as reported in journey plans in which journeys start from the moment the animals leave their place of origin.
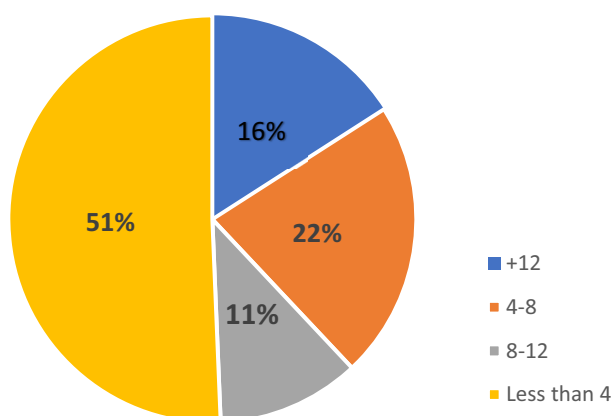
Figure 1: Distribution of journey times (hours) for domestic birds transported between MSs in 2018 and 2019 (EFSA extraction from TRACES)

3.1.3. Export of poultry from the EU

As is the case for intra-EU trade, in extra-EU trade, poultry is the most traded farmed animal species (98% of total exports). Poland (61.9 million heads), Hungary (35.6 million heads) and the Netherlands (29.8 million heads) are the biggest exporters of live poultry. Ukraine, Belarus, Ghana, Egypt, Morocco and Albania are major importers of poultry from the EU (EPRS, 2021; Eurogroup for Animals, 2021). About 70% of poultry exported from EU to third countries are day-old chicks of layers and broilers, but other poultry species (turkeys, ducks, guinea fowls and geese) are also exported (DG Agri, pers. com.)

EU trade in live animals with non-EU countries represents less than 10% of all movements of animals within the EU. Poultry are transported to third countries mainly by road (66% of animals, measured in tonnes live weight), but also by air (26%) and sea (8%). Less than 1.7 million live poultry were imported to the EU in 2019.

3.1.4. Stages of transport

Transport of domestic birds as described in this scientific opinion consists of five stages:

Stage 1: Preparation includes planning of the journey and preparation of the birds by the removal of feed and assessment of fitness for transport.

Stage 2: Loading includes catching the birds, placing them in containers (crating) and loading of containers onto the vehicle.

Stage 3: Journey includes the movement of birds by vehicle and eventually intermediate stops (e.g. change drivers) along the way until the place of destination is reached.

Stage 4: Arrival includes the period from arrival of the vehicle, unloading of the containers from the vehicle and waiting period up to the start of the uncrating. In the slaughterhouse, this stage includes the lairage period (containers on vehicle or stacked in the lairage area).

Stage 5: Uncrating includes the removal of the birds from the containers.

3.2. Stage 1: Preparation

3.2.1. Description

In this section, the practices relevant for this stage, namely the planning of the transport (loading, journey, arrival and uncrating), the removal of feed and assessment of fitness, are considered. Preparation is an important stage as it can prevent the exposure to the hazards leading to the welfare consequences that birds might experience during the later stages of transport, such as heat and cold stress, injuries, prolonged thirst and hunger. If birds are not correctly prepared for transport, this will increase the risk of them experiencing welfare consequences during the journey. Several of the preventive measures suggested for welfare consequences appearing in later stages of transport should be applied during the preparation for the transport phase.

3.2.2. Planning of the transport

Bird transports are usually planned in advance from the stage of loading to arrival and uncrating. This includes the coordination of the different stages of the transport (itinerary for loading and unloading, location of any driver resting places/stops during the journey), estimation of their duration and time of arrival. Effective planning includes communication with live bird catching teams and hauliers and with the slaughterhouse or destination farm (European Commission, 2017a, 2018) for planning and coordinating the arrival of live birds. Identification of the potential welfare consequences and their possible hazards (such as analysis of weather forecast), as well as implementation of preventive and mitigating measures are not always carried out in a structured way. Some companies have developed contingency plans with the purpose of helping the driver and the transport company to ensure the security and the welfare of the animals in case of emergency (European Commission, 2017b).

3.2.3. On-farm feed withdrawal

Feed withdrawal is carried out before transport of birds to slaughterhouses to reduce the risk of contamination of carcasses by contents of the gastrointestinal tract during slaughter. Fasting prior to transport is not required for birds transported to production sites. In broilers and turkeys, on-farm fasting is commonly carried out 4–8 h before catching the birds. For broilers, industry guidelines recommend a total feed withdrawal time of minimum 8 h and maximum 12 h before slaughter (Aviagen, 2018; Cobb-Vantress Inc., 2018). In meat duck production, feed is usually removed at least 8 h before catching. Owing to their low value, feed may be withdrawn early or reduced in quantity and nutrition in end-of-lay hens before depopulation. This issue is compounded in very large houses, which take several days to depopulate the flock. Timing of feed withdrawal in pullets depends on catching time and birds are often transported during the overnight dark period when they would not be eating anyway, thus effectively reducing sensations of hunger. In quails, total feed withdrawal is between 6 and 8 h before expected time of slaughter to avoid carcass contamination and feather spoilage during transport. However, feed is not always withdrawn on farm and this practice is less common in cage-housed quails. Water is generally offered until catching commences.

3.2.4. Fitness for transport

3.2.4.1. Description

Throughout the scientific literature, it is agreed that – in terms of animal welfare – making sure that animals are fit for transport before departure is of utmost importance (Grandin, 2001; Cockram, 2019). However, currently, no scientific definition of the concept of fitness for transport exists (Herskin et al., 2021).

Animals sent for slaughter with pre-existing conditions of poor health are more likely to die in transit, increase the severity of the welfare consequences they are exposed to during the journey or be condemned as unfit for human consumption upon arrival at the slaughterhouse (Cockram, 2019). Fitness should relate not just to individual pathological conditions but also to the suitability of a flock of birds to undertake the journey planned, and in the conditions prevailing at the time.

It is very difficult to observe the fitness for transport of pre-existing conditions such as illness, cachexia, lameness and wounds during catching and once the birds are in transport containers, so only animals fit for transport should be presented and animals injured during the catching process should not be loaded. Prior to catching and/or at the latest during catching and loading, the fitness for

transport of each animal should be assessed, and animals that are not fit to be transported should receive appropriate treatment or be immediately culled (EFSA AHAW Panel, 2018). Additionally, animals which would be rejected as unfit for human consumption should not be transported to slaughter to avoid exposing them unnecessarily to transport.

To minimise the risk of animals becoming unfit for transport between the last inspection and the time of catching, the final inspection should be as close as possible to the time of catching (e.g. within 12 h prior to catching based on expert opinion). At all times before a person trained in killing methods must be available to dispatch injured and unfit animals. In addition, suitable equipment such as captive-bolt stunners should be available. In automated catching and crating processes, one or more people should be present at the point of loading of the containers to remove any injured animals.

The advantages for animal welfare of training stock people and handlers on how to identify unfit animals is increasingly recognised (Hester, 2005), with specific benefits of training leading to improved handling and transport (Broom, 2005) and resulting from altered attitudes towards animals (Hemsworth, 2003). An example of existing training is the Poultry Handling and Transportation Manual and Education Program developed by the Canadian Poultry Service Association (PSA, 2017). The implementation of both incentive payment and auditing by restaurant company customers reduced the prevalence of broken wings to 1% or less in lightweight birds and to less than 3% in heavy broilers (Grandin, pers comm).

3.2.4.2. Conditions leading to domestic birds unfit for transport

Key conditions identified by the Consortium of the Animal Transport Guides Project (2017) are birds with broken bones (legs, wings), with 'severe difficulties to move' and wet birds. A principal UK welfare assurance scheme specifies for end-of-lay hens that birds which are visibly unfit (including those that are lame, fatigued, injured or ill) should not be transported (RSPCA, 2017). The Canadian Food Inspections Agency has a longer list of conditions rendering animals unfit for transport, including those which appear to be dehydrated, and views wet birds as compromised (HAR, 2020[2]).

The principal conditions, identified by the EFSA experts based on literature and expert opinion, which will make domestic birds unfit for transport are:

- Evident signs of illness.
- Emaciation and cachexia.
- Severe lameness: Unable to stand or walk more than a few steps.
- Open wounds and prolapse.
- Poor feather cover in low effective temperature (end-of-lay hens only).
- Broken bones (legs, wings) and dislocations.
- Wet plumage in low effective temperature (except for ducks and geese).

The list is not exhaustive but is based on the limited published evidence and expert opinion. Further research is needed to identify other conditions and thresholds that make the animals unfit for transport.

Evident signs of illness

Ill animals are at a high risk of compromised welfare during transport. High levels of mortality on farm, which is often indicative of ill health of the flock, have been shown to be positively correlated to levels of DOA or other physiological indices after transport in both broilers (Chauvin et al., 2011; Kittelsen et al., 2015a,b; Jacobs et al., 2017a,b) and end-of-lay hens (Weeks et al., 2012). Evident signs of illness include distressed breathing (gasping), diarrhoea (freshly soiled plumage beneath the tail), neurological symptoms (e.g. head withdrawn towards body), hunched posture, ascites in broilers (dilated abdomen/oedema, distressed breathing). Further indicators are being moribund i.e. unable to or struggling to move when approached; having swollen, puffy heads and sinuses; dark red, purple or black combs or wattles.

Emaciation and cachexia

Emaciation refers to poultry that are underweight compared with other animals in the group, which are thin and emaciated. Cachexia is the term used to describe the end stage of emaciation. Gregory and Austin (1992) reported that 24% of DoA broilers that weighed less than 1 kg were severely dehydrated and speculated that runts would probably become unable to reach drinkers which were raised with the growth of the rest of the flock. Cachexia was a risk factor for DOA in broilers in

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[2] https://inspection.canada.ca/animal-health/humane-transport/health-of-animals-regulations-part-xii/eng/1582126008181/1582126616914#a7

Belgium (Jacobs et al., 2017a,b) and a major reason for condemnation of broilers and meat ducks in a French survey (Salines et al., 2017). In the UK, low body weight was associated with an increased risk of DOA in end-of-lay hens (Weeks et al., 2012).

Severe lameness

Any bird unable to stand or walk more than a few steps is unfit to be transported. The ability of broilers to stand and to walk and the quality of their walking ability has been scientifically evaluated using various mobility tests (e.g. Caplen et al., 2014). However, scoring systems that provide a description relating to each numerical score remain the most practical and quick for commercial assessment of lameness. The original 6-point gait scoring system for broilers was developed by Kestin et al. (1992), where 0 represents a sound bird. The following descriptions apply to severely lame birds at the other end of the scale: Gait Score (GS) 4 'The bird had a severe gait defect. It was still capable of walking but only when driven or strongly motivated. Otherwise it squatted down at the first available opportunity. Its acceleration, manoeuvrability and speed were all severely affected.' and GS 5 'The bird was incapable of sustained walking on its feet. Although it may have been able to stand locomotion could only be achieved by the assistance of the wings or by crawling on the shanks.' Welfare standards (e.g. RSPCA, 2017) require that broilers of GS4 and GS5 are routinely culled on farm and thus they would be considered unfit for transport. However, there is evidence that broilers with intermediate walking ability (GS2.5 and GS3) experience pain which is relieved by the administration of analgesics which then improve their mobility (e.g. Danbury et al., 2000; Caplen et al., 2013; Hothersall et al., 2016). The welfare consequences of transporting moderately lame birds, most of which are likely to be experiencing pain, have not been formally examined but it is probable that handling and motion stressors would increase levels of pain.

For turkeys, a 4-point score (from GS0 to GS3) was used by Olschewsky et al. (2021) where the description of GS 3 is 'Bird sits down again as soon as possible or can only move with great effort (e.g. flapping of wings)'. Turkeys assessed to have GS3 are to be considered unfit.

Open wounds and prolapses

The opinion of the EFSA experts is that wounds that penetrate all skin layers, or which also damage the underlying tissue, are a cause for concern (Figure 2). Minor abrasions, scratches or wounds that are healed do not constitute a reason for declaring birds unfit for transport. More severe skin tears and lacerations involving all skin layers that are unhealed or such wounds that are infected and inflamed (e.g. abscesses) will result in pain and discomfort and should be regarded as reasons for exclusion from transport.

Figure 2: An example of a wounded hen unfit for transport. Source: laywel.eu.

In domestic birds, any cloacal prolapse is essentially similar to wounds comprising unprotected, protruding flesh that is potentially painful and susceptible to further injury and infection (Figure 3), thus rendering the animal unfit for transport.

Figure 3: Prolapse in laying hen. (Photo credit: Gaynor Davies)

Fractures and dislocations

A large survey of broilers at a Canadian slaughter plant identified heavier weight (2.29 kg compared with those ≤ 2.14 kg) as a risk factor for DoA and the authors speculated a likely reason for this was femoral hip dislocation when carried inverted during loading (Caffrey et al., 2017), confirming earlier work by Gregory and Wilkins (1990), who found 27% of DoA broilers from six UK plants had dislocated femurs. Several studies have associated fractures with DoA and rejects (Warriss et al., 1992; Nijdam et al., 2004; Mönch et al., 2020). These conditions make a bird unfit for transport.

Poor feather cover (applies to end-of-lay hens only)

The impact of cold stress is exacerbated when end-of-lay hens have insufficient insulation from plumage. A survey of 13.3 million hens transported in the UK found that the risk factors for DoA for end-of-lay hens relating to on-farm fitness included poor feather cover (Weeks et al., 2012) which is associated with increased mortality in low effective temperatures. Thus, end-of-lay hens with poor feather cover (Figure 4) have a high risk of DoA and may be unfit for transport if conditions during catching and when in the containers result in low effective temperature (below their thermoneutral zone – see Section 3.7.8 on cold stress). Several scoring systems are used to assess feather cover (for instance, Roehe et al., 2020). The LayWel system[3] provides photographs (e.g. Figure 4).

Figure 4: An example of poor feather cover on the back. Source: laywel.eu.

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[3] https://www.laywel.eu

<u>Wet plumage in low effective temperatures (except for ducks and geese)</u>

Poultry should not be allowed to become wet before and during transport if the effective temperatures are low (see Section 3.7.8 on cold stress) and there is a risk of cold stress during transport. Wet plumage is unlikely to be a risk for DoA for ducks and geese. Thus, wet integument only makes birds unfit to travel if they will be transported in cold weather without the application of preventive and corrective measures discussed in Section 3.9.1.

3.3. Stage 2: Loading of domestic birds for transport by road

3.3.1. Description

The loading stage includes catching the birds, placing them in containers or crates and loading of containers onto the vehicle.

3.3.2. Types of containers used for transport of domestic birds

Domestic birds are transported in containers, of which three different systems are established:

a) Loose crate systems with openings on top or on the front (a1 and a2 of Figures 5, 8, 9). This system is the most common used for ducks, end-of-lay hens and quails, although it may be used also for broilers and turkeys. In the case of pullets, end-of-lay hens, ducks and quail, theses crates may be placed onto dollies and wheeled for loading (Figures 7, 9). The dimensions vary between types of crates, e.g. for broilers and quails, they can be 75–90 × 50–55 × 23 cm.

b) Fixed cages on the vehicle, whereby the birds are carried to the vehicle (b on Figure 5).

c) Modular systems (c1 and c2 on Figure 5), which are compatible with slaughterhouse automation, are common in broiler and turkey transport and are also used for ducks and end-of-lay hens. The module consists of a framework supporting a stack of containers. A widely used module consists in a framework supporting sliding drawers. In this case, loading starts by placing the birds (manual or mechanically) into the opened bottom drawer of the column. The loaded drawer is then closed by pulling back the empty drawer immediately above, to prevent birds escaping. Loading continues up the column of drawers. Modular systems with side access (Figure 6) are also used. The whole module is handled by forklift for transfer onto and off the vehicle. The typical systems have between four and five levels, the dimensions might vary between companies and the height is 23 cm in broilers and around 34 cm in turkeys.

Löhren (2012) reported a rough estimate of the types of container systems used in the EU for the different poultry species.

Broilers: 70% modular systems and 30% crates.

Turkeys: 40% liners (fixed cage on the vehicle), 40% crates (special crates for turkeys) (Figure 10) and 20% modular systems (Figure 11).

End-of-lay hens: 75% crates and 25% closed containers,

Ducks: 33% crates 33% modular systems (mainly drawer type modules) and 33% liners (different compared to turkeys).

Quails: 100% crates.

Recent information on the use of various systems has not been retrieved. However, the AHAW Panel believes that the proportion of birds for slaughter transported in modular systems is higher today than in 2011, especially for birds exposed to controlled atmosphere stunning for which only modular systems can be used.
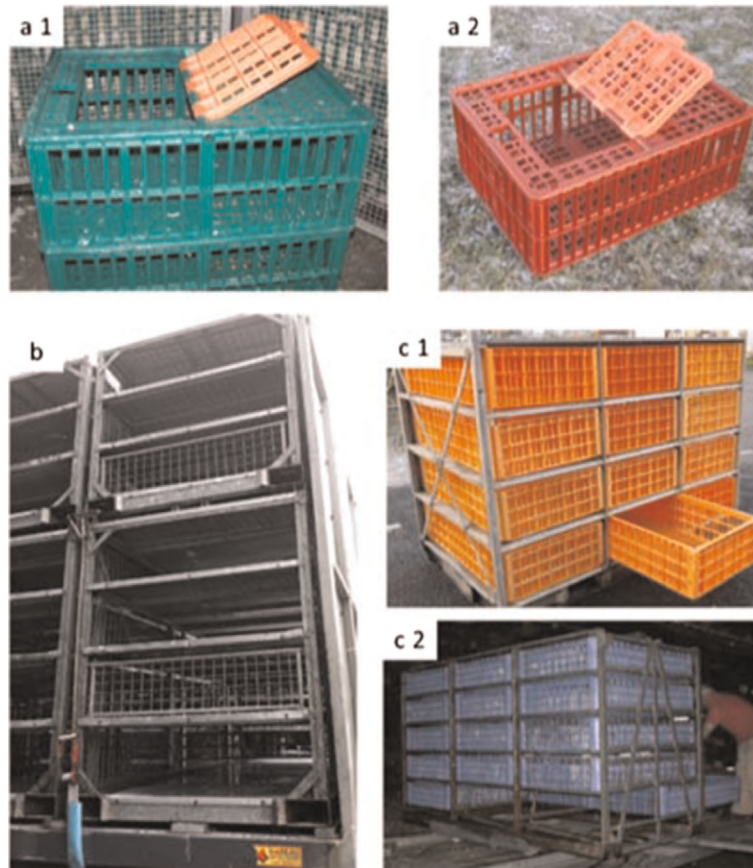
Figure 5: Most common types of containers used for the transport of poultry are (a) plastic crates with top opening or top and side opening, depending on model; (b) fixed containers with front and side access; c1 and c2: Modules with plastic drawers that slide (Source: Avip le Formation)

Figure 6: Modular system with side loading (courtesy of Malcolm Mitchell)

Figure 7: Dolly modules used for transporting pullets (courtesy of Jean-Paul Michalski)
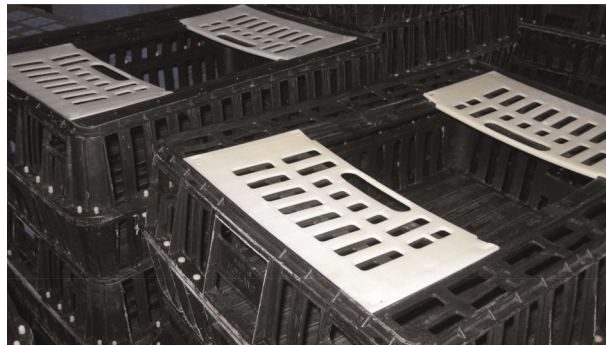
Figure 8: Crate type frequently used for transport of laying hens and broilers. The size of the crate is 85 × 66 × 30 cm, with an opening of 30 × 35 cm (courtesy of Thea van Niekerk)

Figure 9: Crates moved on a wheeled dolly (courtesy of Thea van Niekerk)

Figure 10: Loose crate for turkey transport. The size is 97 × 58 × 42 cm. (Source: Giordano Poultry Plast SpA)

Figure 11: Turkey transport module. The size is 119.5 × 113.5 × 39 cm (Courtesy of Leonardo J. Vinco)

Adult and juvenile pheasants and partridges are usually transported in disposable cardboard boxes (Figure 12).
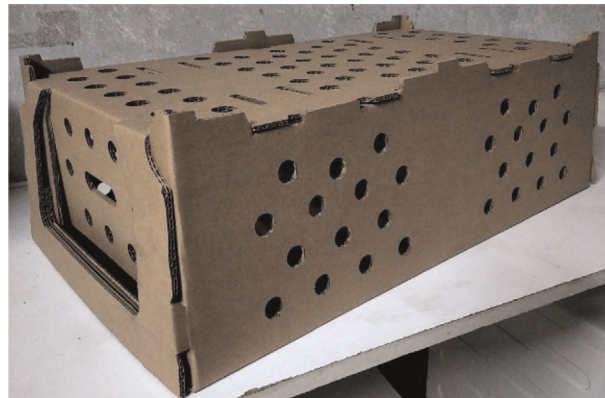
Figure 12: Cardboard box used for transport of pheasants and partridges. The dimensions are 64 × 37 cm with 18.8 cm height for pheasants and 14.5 cm height for partridges (Courtesy of Virginie Michel)

The space allowance for poultry during transport is defined in Table 5 according to the live weight in the current EU transport regulation (EC Reg 1/2005) as follows:

Table 5: Current requirements for space allowance for poultry in the EU transport regulation

Category	Area in cm²
Poultry other than day-old chicks: weight in kg	*Area in cm² per kg*
< 1.6	180–200
1.6 to < 3	160
3 to < 5	115
> 5	105

3.3.3. Catching and crating

For catching and crating poultry, two different methods are practiced: (1) manual and (2) mechanical.

1) Manual catching is the most common method and is carried out by farm staff, purposely recruited staff or professional catching teams. Birds may be manually caught and loaded into any of the different types of containers described above. Where possible, the containers are placed into the shed close to the birds by hand, wheeled on dollies or by using a forklift. Bird handling may differ in relation to species and category. In the case of broilers and layers, catchers grasp the birds by one leg and carry them in an inverted position with two to four birds per hand to the container which may be inside or outside the house or sometimes fixed on the vehicle. As a better practice, birds might be carried by both legs with support of the body. Carrying birds by two legs will limit the number of birds to be carried to maximum of two to three birds per hand. Operators could use their own legs as breast support to calm the birds after inversion and minimise wing flapping. A less common practice, applied mainly to end-of-lay hens, is to catch the birds in an upright position, around the wing and chest and a maximum of two animals at a time. After catching, operators carry the birds up to the containers and load these.

In the case of manual catching (Figure 13), turkeys are grasped with one hand by the legs, from behind and lowered onto their breast. With the other hand, the shoulder of the wing furthest away is grasped to lift and carry the bird. Manual catching of turkeys has been largely abandoned by the turkey industry mainly due to the weight and size of the turkeys at slaughter age (hens around 9 kg at 15 weeks of age, toms around 21 kg at 20 weeks of age). Instead, they may be herded and guided towards the containers and pushed in.

Figure 13: Manual catching and loading by forklift of turkeys. (Courtesy of Leonardo J. Vinco)

Pullets kept in loose-housed rearing systems are often driven into portable pens to aid capture. Loose crates are commonly used. Narrow dolly modules (Figure 14) can be wheeled into the laying house upon arrival (including down aisles of caged housing systems), thus facilitating pullet placement. For biosecurity reasons, they are generally loaded outside the pullet rearing house. Due to reduced labour availability, dollies are increasing in popularity in Europe.

A labour intensive but thermally more comfortable system comprises crates built as permanent fixtures on both sides of the bed of the vehicle with either an open-top central ventilation channel running the length of the vehicle (liners) or with fan ventilation. Hinged openings to the outside are used to load and unload the birds, which therefore must be carried out of the containers and into their housing. They may also be passed in handfuls from person to person.

Independently of the housing system, catching, crating and loading end-of-lay hens is very labour intensive, with bunches of inverted hens commonly being passed along a human chain before they are placed in the transport container, which is frequently placed near the entrance to the house rather than next to the birds, although containers on wheels are increasingly used (Figures 9 and 14). Both loose crates and modular systems are commonly used for end-of-lay hens, although vehicles with liners fixed to the vehicle are also in use.

In cages, end-of-lay hens are caught individually or in groups of two or three by pulling them out by one leg. During removal from the cage, they may be struck against the structures that are inside (e.g. food trough, perches and nest boxes). End-of-lay hens may also hit cages or roof supports, as they are carried down the narrow aisles of a battery house (Knowles, 1991). Most catching teams have people at the far side (back) of the cage to help drive the hens towards the grasp of the catchers. Catchers have to climb to the highest tiers and also to walk the full length of very large houses carrying the birds, making the process hazardous and time-consuming. In aviary systems, end-of-lay hens might be difficult to catch, as they tend to crowd and pile up at the end of aisles. This might cause suffocation or attempts to flap and fly with the risk of injury to the catchers. Containers may be placed onto dollies and wheeled into the aviary house for loading, avoiding inversion (Figure 14). However, litter needs to be moved from the floor or tracks laid down. That preparation would also be needed for trolleys carrying module drawers as there is seldom sufficient space in layer housing to take a whole module into the house. Otherwise, hens are carried inverted in bunches of 3 per hand to be loaded outside into their transport container.

Figure 14: Narrow, modular systems (dollies) used for laying hens that can be loaded and unloaded directly into cages or aviaries. (Photo credit: Niek Samyn)

Ducks are caught and carried by one wing and carried one bird per hand, or by the bases of both wings but may also be driven towards the containers.

Quails are housed in cages or floor systems. In caged systems, the catching and crating is similar as for end-of-lay hens. In floor systems, a current practice for catching these quails is to gather them in buckets or bags of 20–30 birds and transferring them to the crates that can be allocated either in the barn or on the vehicle.

Game birds are kept outside and catching takes place during daylight. In warm weather, it usually takes place in the morning to take the advantage of cooler temperatures. Animals are directed to restricted pens, where they are caught by hand or with landing nets and put into boxes.

2) Mechanical catching systems are increasingly applied since the 1990s in loose housing systems to replace manual catching as they have potential to reduce injuries and stress for birds and humans alike (Mitchell and Kettlewell, 2004a,b). The most common commercial systems use sweeping mechanisms provided with soft rubber 'fingers', mounted on vertically rotating rotors (FAWC, 2019). The fingers sweep the birds onto a conveyor belt system that transfers them into the containers. Mechanical catching systems more commonly used as they become more reliable and the difficulty of recruiting human bird catchers increases in some countries (Weeks et al., 2019). Nevertheless, catching poultry mechanically has to be overseen by humans trained in the operation of the machines and with knowledge of bird behaviour. With automated harvesting systems, container systems or liners are exclusively employed. There are automated systems for loading liners based on rotating floors and conveyors, but these are uncommon in the EU. Crates cannot be used because of the small openings.

In turkeys, automatic collecting and crating systems have been developed since the seventies and refined over the years with clear improvement of worker's ergonomics and animal welfare thanks to reduced handling of the birds. Unlike automated collecting and crating systems developed for broilers, systems developed for turkeys require that the birds are manually herded towards the conveyor belt of the loader. They are then automatically driven up towards the transport crates. Insertion of the birds into the crates will differ according to the different technology of the loader. In less recent models, the birds are collected manually at the top of the loader's conveyor belt and inserted manually into the crates ('balcony' system). Birds are grasped by the base of the wings, with one hand and the contralateral leg (or both legs) with the other hand and pushed into the crate sliding the keel bone on the floor of the crate with the bird's head facing forward. In more recent models, the tongue of the conveyor belt is inserted into the crates, so that the birds are directly conveyed into the crate with no handling at all. This later system requires the use of crates with a higher head space to allow smooth entrance of the birds and to avoid that the birds may flip over by colliding with the top entrance of the crate, as the insertion of the tongue into the cage causes a reduction in head space.

A specific housing system (Patio system) developed for broilers places chicks just before hatching on belts within a multi-tier system. This avoids the need to transport day-old chicks, which jump down after hatching into the system where feed and water are available. Each encaged tier of the system runs the full length of the house and is fitted with a conveyor belt covered with litter. At the end of the growing period, the conveyor is started up and moves the birds slowly to the end of the house where the litter drops down and the broilers move onto a system of conveyors that takes them directly to the transport container. The containers are loaded automatically in the same way as with mechanical catching.

3.3.4. Loading of containers onto the vehicle

Where loose crates or liners are used, loading of the vehicles is commonly manual. Where the loose crates are stacked onto pallets or dollies and modular systems are employed, they are loaded onto the vehicles by means of a forklift. Some mechanical catching systems also load the vehicle automatically, often via a system of conveyors.

3.4. Stage 3: The journey by road of domestic birds

3.4.1. Description

This stage includes the movement of animals by vehicle and intermediate stops along the way until place of destination is reached. During the journey stage, the assessment of welfare of the birds using animal-based measures (ABMs) is difficult. Poultry are subjected to mass transportation within crates, containers or chick boxes, which fill the load space, so inspection of each individual animal is not feasible.

Road transport

The vast majority of broiler, turkeys and end-of-lay hen transport vehicles are roofed, open-sided and naturally ventilated. Several vehicle types are used from flatbed to articulated lorries and either can pull an extra trailer. To protect the birds from adverse weather conditions, most vehicles are fitted with side curtains that can be open or closed depending on the wind and weather conditions. Many

have vents in the headboard and tailgate which can be adjusted to modify airflow according to weather conditions. A few vehicles may have fans fitted to aid ventilation during stops (e.g. for some turkeys, breeders, pullets). Most of transport of gamebirds is done by vans. For mature birds, the ventilation system can be passive or active and the capacity of the vehicle varies from up to 200 birds to 5,000 birds.

Legislation governing driver rest times (Reg. (EC) 561/2006) requires the driver to have a 45-min break after 4.5 h of driving. This 45-min stop can be avoided by using two drivers. Maximum journey times for poultry are not defined in the EU transport regulation (EC 1/2005), but it requires that poultry are given water and if necessary, feed after 12 h of journey, excluding time spent loading and unloading. Some vehicles have a watering system installed to provide water inside the containers. Feed is not usually offered to birds during transport as it is not practical to distribute feed to containers.

The majority of journeys for poultry in the EU (as reported in TRACES) are less than 4 h, but the average duration varies between categories of birds. Broilers and turkeys normally have short journeys to slaughter due to the large production volume of these species and consequent high availability of slaughterhouses. Ducks kept for foie gras production are generally raised close to the slaughterhouse, resulting in short journeys. Journey duration from the farm to the slaughterhouse was reported in the survey to the MSs to be 0–3 h for foie gras ducks and 3 h for meat ducks. In Spain, most of transport to the slaughterhouse lasts less than 2 h. Very few slaughterhouses for poultry process end-of-lay hens (generally only 1–3 per MS), and the reduced availability increases journey durations for these birds.

Gamebirds are routinely transported over varying distances including international transports first shortly after hatching from the hatchery to the rearing facility and subsequently from the rearing facility to the place where they are released. Pheasants and partridges might be transported up to 12–15 h in where the vehicles can stop in several farms before ending the delivery. Therefore, animals might stay on the vehicle for duration reaching up to 20 h. Usually, no feed or water is provided.

Travel distance and journey duration are expected to be strongly associated (Di Martino et al., 2017a). However, longer durations than expected by the spatial distance can occur e.g. due to multiple stops during the journey or unpredictable delays e.g. traffic obstructions, accidents and related hazards (Warriss et al., 1990). Unpredictable as well as predictable factors require a contingency plan (Figure 15).

Figure 15: Vehicle for adult pheasant and partridge transport (courtesy of Virginie Michel)

3.5. Stage 4: Arrival of domestic birds

3.5.1. Description

This stage includes the period from arrival of the vehicle, unloading of the containers from the vehicle and lairage period (on vehicle or in lairage area) up to the start of the uncrating.

Following arrival of the vehicle at the slaughterhouse, poultry are kept either in lairage on the vehicle or unloaded and moved to designated areas. The containers (or crates) are removed from the vehicle mechanically (forklift) or manually and placed in a lairage area where they can stay until the time of

slaughter. The containers are arranged in spaced rows allowing human passage and observation of the birds. This arrangement also allows air to circulate between the containers. Commercial practices vary from unloading of the birds and moving them straight to the point of stunning without lairage, to holding them in lairage for some hours.

When reaching farms or releasing sites, vehicles are usually unloaded immediately, with an unavoidable delay between first and last animal unloaded.

The time that animals spend in the containers during lairage can be very variable. In Italy, Grilli et al. (2015) estimated this time for 233 different batches of broilers as ranging between 0.2 and 9.4 h with a mean of 4 h. A Belgian study (Jacobs et al., 2016) undertaken in six slaughter plants, found lairage durations from 15 to 555 min (9.3 h), with a mean at 275 min (4 h 35 min) for broiler chickens.

3.6. Stage 5: Uncrating of domestic birds

3.6.1. Description

During this process, birds are removed from the containers and released into new housing or in nature (game birds) or prepared for stunning at the slaughterhouse. In slaughterhouses with water bath stunning systems, the birds are manually taken out of the containers and shackled. When gas stunning systems are used, birds are stunned while still in the containers so they are not exposed to the process of uncrating.

Uncrating is the stage where dead birds (DoAs) can be assessed, and visibly sick or injured birds can be identified.

3.7. The highly relevant welfare consequences identified for transport of domestic birds

As explained in Section 2.2.2, an exercise based on expert knowledge elicitation was performed to identify the highly relevant welfare consequences for transport of domestic birds. Welfare consequences were not identified for the preparation stage, as they will mainly appear at later stages of transport. An overview of the results is presented in Table 6 below. The description and assessment of the welfare consequences are provided in this chapter.

Table 6: Welfare consequences identified as most relevant for road transport of domestic birds to slaughterhouses or production sites. (n.a. = non-applicable, x = highly relevant, − = not relevant)

Transport scenario 1: Road transport of domestic birds to slaughterhouses or production sites					
Welfare consequence	*Preparation*	*Loading*	*Journey*	*Arrival*	*Uncrating*
Handling stress	*n.a.*	x	−	−	x
Bone lesions	*n.a.*	x	x	x	x
Soft tissue lesions and integument damage	*n.a.*	x	x	x	x
Restriction of movement	*n.a.*	x	x	x	x
Sensory overstimulation	*n.a.*	x	x	x	x
Motion stress	*n.a.*	−	x	x	−
Heat stress	*n.a.*	−	x	x	−
Cold stress	*n.a.*	−	x	x	−
Prolonged hunger	*n.a.*	−	x	x	−
Prolonged thirst	*n.a.*	−	x	x	−

It was noted that the two welfare consequences i. bone lesions and ii. soft tissue lesions and integument damage apply to all stages and will have same preventive and corrective measures. For this reason, to avoid repetition of text, they will be presented in the same section under the common term 'Injuries' which will therefore referred to as from here onwards to indicate these two welfare consequences.

3.7.1. Handling stress

3.7.1.1. Description

Domestic birds are predominantly caught in their housing and crated in containers, then moved and loaded onto the vehicle. Although, a few designs of vehicles have fixed containers, so birds are carried outside to the vehicle and loaded into the containers. Handling stress, meaning that the animals experience stress and/or negative affective states such as fear resulting from human or mechanical handling, is regarded a highly relevant welfare consequence for all animals transported in containers. The duration of the handling is limited to the stages of loading, arrival and uncrating, but the welfare consequences can prevail throughout the entire transport operation. The severity of handling stress is considered as moderate, meaning that the welfare of the animals is clearly affected, but that this effect is less severe than, e.g. the welfare consequences of injuries, such as bruises and dislocated or broken bones.

Although domestication and subsequent genetic selection of poultry have reduced the magnitude of their fear responses, poultry still perceive humans as predators (Gerritzen and Raj, 2009). A recent Swiss study (Gerpe et al., 2021) on 15 commercial flocks of end-of-lay hens from aviaries found plasma corticosterone concentrations were 16% higher during depopulation than at baseline, indicating stress from catching and handling. Cloacal temperature was 0.47°C higher and comb temperature was 3.4°C lower in depopulated hens – highly significantly different from baseline measures. In addition, the respiration rate of depopulated hens was five chest movements per minute higher than during baseline (41 vs. 36). The authors concluded that most birds (90%) showed moderate adverse effects from the catching and crating process.

Considering the available evidence, the AHAW Panel concluded that handling stress is a welfare consequence inherent to transport, and all domestic birds are likely to experience this welfare consequence, mainly during catching and crating of the loading stage, during unloading at the arrival stage and during the uncrating stage.

The severity of the handling stress will depend on the birds' genetics and familiarity with people and handling. Birds have different levels of reactivity towards handling and might be impacted differently by catching, crating, loading and unloading. The handling process is likely less stressful for poultry, accustomed to handling (manual weighing, injectable vaccinations, etc.). However, also the nature of previous interactions with humans is very important as rough handling increases fear reactions towards humans and decreases the ability to establish positive human–animal relationships (Laurence et al., 2014). Pullets may receive their final vaccination(s), which adds to the stress of catching and handling.

Domestic birds are kept in different husbandry systems, from various types of cages to floor systems on litter or on slatted floor, with or without outdoor access. In free-range or organic systems birds might be more active and have more space, resulting in them being more difficult to catch. Cage and multi-tier housing systems pose challenges for catching hens.

In the most common procedure, catchers grasp the birds by one leg and carry them in an inverted position to the containers. Handling of broilers in an inverted position leads to increased plasma corticosterone levels compared to birds handled upright (Kannan and Mench, 1996). The aversiveness of inverted carrying has been shown in experiments on end-of-lay hens by elevated plasma corticosterone concentrations in hens removed from conventional cages three at a time and carried in an inverted position from the house, compared with those removed individually and crated before removal from the house (Knowles and Broom, 1993). Birds that are caught carefully and held in an upright position present less agitation and stress compared to inverted birds that are caught by the legs (Broom and Knowles, 1989; Carvalho, 2001; Langkabel et al., 2015; De Lima et al., 2019).

Considering the available evidence, based on their expert opinion, the AHAW Panel concluded that inversion will increase the severity of handling stress compared to handling birds in an upright position.

Catching machines are increasingly replacing manual catching for broilers and turkeys and function by using rubber fingers to sweep the birds onto a conveyor belt which transports and loads the birds into the containers. Catching systems developed for turkeys require that the birds are manually herded towards the conveyor belt of the loader, which will transfer the birds directly into the containers or to the front of the container in which the birds will be inserted manually. Mechanical catching systems have the potential to reduce handling stress for birds and humans alike (Lacy and Czarick, 1998; Knierim and Gocke, 2003), and several are commercially available for broiler chickens (Mitchell and Kettlewell, 2004a,b). Delezie et al. (2005, 2006) have reported a quicker decline in corticosterone

levels and reduced duration of tonic immobility following mechanical compared with manual catching and inverted handling. The heart rate and corticosterone levels of the broilers caught mechanically return to basal levels more quickly (Duncan et al., 1986) than those caught manually. A possible explanation for the lowered stress is that the birds are not inverted during mechanical catching. However, in a study by Nijdam et al. (2005), in paired comparisons of commercial depopulation by hand or mechanically on eight farms, no differences in physiological indices of stress, such as corticosterone levels, were found. However, increased experience of the operators may improve performance, as found by Knierim and Gocke (2003) during a year-long survey of automated broiler handling. They noted a reduction in loading time by a third, and significantly decreased rates of injury, likely explained by experience and adjustments to conveyor speed which was reduced from 1.4–1.6 m/s to 0.8–1.0 m/s. There is evidence suggesting inclines should be avoided where possible as Scott and Moran (1992) found significant increases in loss of balance, wing flapping and alarm calls by hens conveyed up or down slopes rather than horizontally. Scientific evidence supports that turkeys walking to the mechanical modules had a lower level of damage and elicited lower mean and maximum heart rates compared to birds loaded manually (Prescott et al., 2000).

In a study of laying hens, levels of fear, measured by tonic immobility, were lower in hens moved for 20 m on a conveyor belt than those of hens carried the same distance in an inverted position by hand or on a processing shackle. Still, the study did not include upright carrying by humans (Rutter et al., 1993).

Considering the available evidence, based on expert opinion, the AHAW Panel concluded that mechanical catching can potentially reduce the severity of handling stress compared to manual catching and inversion if it is operated correctly.

Processes associated with pretransport handling represent significant stress in game birds (capture of the birds, close contact with humans, transfer, placement in transport containers). While the physiological processes in pheasants and rearing practices are similar to those of some domestic poultry, a number of studies (Voslarova et al., 2006; Suchy et al., 2007; Chloupek et al., 2009) have described considerable differences in stress response between domestic poultry and pheasants, which are primarily wild-living animals. According to Bedanova et al. (2018), the greatest stress associated with pheasant transport is the handling of the birds and close contact with humans, and this stress load is higher in older pheasants than in younger birds (8 vs. 16 weeks).

3.7.1.2. ABMs

ABMs that are considered feasible for assessing handling stress in domestic birds during transport are given in Table 7. The welfare consequence of handling stress may be measured by observing escape attempts, piling up and alarm calls, which are behaviours generated in response to predators with the aim of reducing detection and capture.

Tonic immobility is a fear-potentiated response induced by physical restraint (Jones and Faure, 1981), and in the tonic immobility test, the bird is restrained and quickly turned on its back. Although immobility testing is useful to identify level of fear in birds, it is easier to apply in experimental or farming conditions than during catching and crating.

Table 7: ABMs for handling stress in domestic birds

ABM (animal categories)	Definition and interpretation of the ABM
Escape attempts	**Definition:** Moving, running or flying away or attempts to do so including wing flapping and pecking or scratching the catcher, often accompanied by vocalisations (Graml et al., 2008) or alternatively freezing or tonic immobility. The number of escape attempts correlates with fear (of humans) (Hemsworth et al., 1993). **Measurement:** Number of escape attempts or number of birds showing escape behaviours. **Sensitivity** is high since most of the birds experiencing handling stress are likely to escape or try to escape. **Specificity** is moderate as even in the absence of handling stress some birds may show escape behaviour due to other welfare consequences (e.g. sensory overstimulation).

ABM (animal categories)	Definition and interpretation of the ABM
Piling up	**Definition:** Birds crowding against and on top of each other during the catching process (from EFSA AHAW Panel, 2019). Escape behaviours during the catching process can lead to the animals accumulating in a corner of the cage or barn. **Measurement:** Number of birds or events. **Sensitivity** is moderate since not all the animals experiencing handling stress will show this behaviour. **Specificity** is moderate as even in the absence of handling stress piling up can be seen due to other welfare consequences (e.g. cold stress).
Distress calls	**Definition:** Single or repeated short and loud shrieking (screaming) at high frequencies (Manteuffel et al., 2004) (from EFSA AHAW Panel, 2019). When animals are caught to be put in a container, or when animals are in the containers which are moved and loaded, they might vocalise with a change in frequency (Ginovart-Panisello et al., 2020) as well as the acoustical spectrum of calls that indicate stress (Herborn et al., 2020) or alarm. **Measurement:** Number of calls or proportion of birds presenting alarm calls. **Sensitivity** is moderate, as only a proportion of animals experiencing handling stress will show distress calls. **Specificity** is moderate because birds not experiencing handling stress can show distress calls for other welfare consequences.

The ABMs escape attempts, piling up and distress calls are highly to moderately sensitive and specific to assess handling stress in a group of birds during catching and crating of the loading stage, during unloading at the arrival stage and during the uncrating stage. However, they are not feasible in transit.

3.7.1.3. Hazards

Staff and equipment

Humans are the main source of handling stress since they are perceived by the birds as predators (Gerritzen and Raj, 2009). Depending on the quantity and quality of previous human contact and on the handling (calm vs. rough), the level of handling stress will vary from low to very high. Equipment (caching machines, forklifts, conveyor belts) may also contribute to handling stress.

Rough handling

Rough or inappropriate handling of birds, e.g. by grasping the bird by the neck or by one leg or wing, will exacerbate the handling stress of birds. Rough handling may imply also that the containers are mechanically or manually moved inappropriately, resulting in tilting, dropping or shaking at loading or unloading in a way that causes handling stress. High speed of catching will increase the risk of rough handling.

The type and design of housing systems may impact the risk of rough handling due to difficulties in catching the birds. Catching end-of-lay hens in systems such as aviaries is often made difficult by the furniture and structure associated with the perches and nest boxes, and the fact that birds have more opportunity to escape (FAWC, 2019). The removal of hens from cages can be even more difficult since these do not usually allow easy access.

Inversion

Inversion refers to holding birds in an upside-down position. As birds do not have diaphragms, inversion can provoke compression of the heart and lungs by the viscera and might compromise breathing and cardiac activity. This causes stress, fear and wing flapping behaviour in an attempt to return to the upright position.

3.7.1.4. Preventive measures

Staff and equipment

Handling stress due to staff and equipment cannot be prevented, only mitigated since catching, crating and uncrating are necessary procedures when moving domestic birds from the husbandry system to containers for transport.

Rough handling

Handling during catching, crating and uncrating should be performed smoothly. This can be achieved by a good training of the staff to acquire the knowledge and skills required to perform their allocated tasks efficiently, letting them know that animals are sentient beings that can suffer pain and fear and therefore should be treated correctly to avoid negative welfare consequences (EFSA AHAW Panel, 2019).

Inversion

The preventive measure is to avoid inversion. Birds should be carried upright by holding the wings against the body (not carried by their legs).

3.7.1.5. Corrective and mitigative measures

Staff and equipment

Positive human–animal relationship during rearing will habituate birds to human presence and mitigate handling stress during catching and crating.

The interaction between staff and birds can be mitigated with mechanical catching and crating. Catching and movement of birds need to be planned and executed well to minimise chances of handling stress. Drops from one conveyor belt to another need to be avoided as they tend to cause injuries and wing flapping. The angles between two conveyor belts should be as wide as possible and any inclines should be minimised. Continually monitoring bird behaviour and adjusting equipment (e.g. reducing belt speeds) is important. Sides may need to be fitted to conveyor systems where birds are more active strains and if inclines cannot be avoided (e.g. in depopulating the Patio® system).

Rough handling

As best practice to mitigate the effects of this welfare consequence, birds should be herded quietly and carefully (in loose-housing systems), and not be swung, thrown or dropped. Loading should be performed smoothly and horizontally to prevent the tilting of containers causing birds to pile up or bunch (Grilli et al., 2015). The catching speed should be constant and appropriate to minimise handling stress.

Containers (crates) used for the movement of birds should be placed as close as possible to the birds to minimise the distance birds are carried. This will also reduce the risk of staff fatigue. Furthermore, staff rotation and rest breaks are important to reduce fatigue, which can lead to poor bird handling practices. Therefore, staff training and rotation, use of well-designed containers and correct setting up of equipment are the most effective mitigative measures. Another way is to slow down the catching process.

The selection of trained catching teams with a certificate of competence will mitigate handling stress and prevent injuries. According to the husbandry system where birds are maintained, specific preparations are required before catching and loading. Of course, catching and loading preparation also depends on the animals' characteristics, but in general:

- Manually caught poultry in cage-free systems should be gently driven into smaller areas where they can be temporarily contained or funnelled by portable hurdles for easier catching and loading. In three-dimensional systems (e.g. aviaries), birds should be carefully caught from upper levels, not driven down.
- Birds in cages (e.g. end-of-lay hens) should be loaded directly into the containers by their cages, avoiding inversion.

Inversion

Although inversion should be avoided where possible, and time spent inverted be minimised by bringing the containers closer, stress and injury may be reduced by handlers using their legs as breast supports to calm the birds after inversion and minimise wing flapping.

3.7.2. Injuries

As explained in Section 3.7, the term 'Injuries' refers to two welfare consequences which are here grouped together: (i) bone lesions (including fractures and dislocations) and (ii) soft tissue lesions and integument damage.

3.7.2.1. Description

This welfare consequence implies that the animal experiences negative affective states such pain and discomfort due to physical damage to the integuments, including plumage, skin or underlying tissues, e.g. bruises, bloodspots, scratches and open wounds; dislocated joints or fractures. For birds transported in containers, injuries are usually inflicted during catching and crating and the welfare consequences prevails throughout the stages of transport. When birds are transported for slaughter, the welfare consequences will last until the birds are slaughtered. In birds transported to other farms, the welfare consequences might last several days or weeks in relation to the severity of the injury. The severity of this welfare consequence will range from moderate (scratches) to very high (fracture), since injured animals experience pain and suffering will prevail and often increase with time.

Based on histological examination Griffiths (1985) concluded that 40% of the bruises recorded at the slaughterhouse originated from catching and crating. A study in Portugal by Saraiva et al. (2020) of bruises arising in the preslaughter stage in broilers, found the mean prevalence of bruises on wings, legs and breasts to be 3.37%, (with high variability between batches ranging from 0.43% to 8.29%). Twenty-five batches presented bruises in more than 4.0% of birds. Bruises were more frequent on wings (3.06%) compared with the legs (0.19%) and breasts (0.12%) in broilers of average weight 1.85 kg and handled manually. Increased journey duration did not increase the risk of bruising, which indicates that damage occurred on farm during loading (Saraiva et al., 2020). On the other hand, in turkeys, the duration of transport between farm and slaughterhouse has been positively correlated with the prevalence of some carcass lesions (McEwen and Barbut, 1992).

Kittelsen et al. (2015a,b) investigated the effects of preslaughter handling procedures on the frequency of wing fractures. Wings were examined for fractures in 11,609 broilers from 12 different flocks slaughtered in two slaughterhouses in Norway, one using biphasic CO_2 stunning and one using electric water-bath stunning. The same broilers were examined (i) in lairage, representing fractures attributed to catching and transportation; (ii) after evacuation of the transport containers and shackling (for electrical water bath stunning only); and (iii) post-stunning. The mean frequencies of wing fractures were 0.8% in the lairage; 2.9% after shackling prior to stunning (electrical water-bath stunning only); and 2.35% after stunning. From this research, it was concluded that regardless of the stunning method, more fractures significantly occurred during preslaughter handling at the slaughterhouse than during catching and transport, but also that fractures occurring during catching and crating result in prolonged suffering and are thus considered more serious in risk assessments of broiler welfare (Kittelsen et al., 2015a,b).

In a study by Kittelsen et al. (2018), broilers were either caught by both legs and carried inverted to the drawers or caught under the abdomen and carried in an upright position. One major finding in this study was a strong tendency ($p = 0.06$) towards more wing fractures when the birds were caught by two legs compared to the abdominal and upright method. Another finding was that the abdominal and upright method was faster and gave a lower and more consistent number of birds per drawer than handling by both legs. However, few broilers were included in this study compared with a commercial catching of larger flocks, and the authors concluded that further studies are needed to assess the effect of the catching method on the crating time.

Considering the available evidence, based on expert opinion, the AHAW Panel concluded that inversion and rough handling will increase the risk of injuries compared to handling birds in an upright position.

When broilers are caught in inverted position, holding them by two legs, compared to one leg, reduces the frequency and severity of haemorrhaging in the thigh (Wilson and Brunson, 1968). The removal of end-of-lay hens from cages holding the birds by two legs compared with a single leg reduced fractures from about 13% to 5% (Gregory et al., 1992).

Mechanical catching systems have the potential to reduce injuries. Knierim and Gocke (2003) found a lower percentage of bruises, broken and dislocated wings after a mechanical catching employing a three-rotor sweeper and conveyor system compared to manual catching (3.1% vs. 4.4% of birds). Machine catching significantly reduced the prevalence of fresh bruises, leg and wing bone fractures, and bone dislocations, of all categories studied (FAWC, 2019). The improvement in the number of bruises ranged from 23% to 31%, the improvement in the number of fractures ranged from 48% to 70% and the improvement in the number of dislocations ranged from 20% to 50%. The authors noted that the prevalence of injuries in mechanically caught birds significantly decreased with greater experience by the catching team in the machine's use; the same was not true for manually caught birds (FAWC, 2019). However, other studies reported more bruises, and broken or dislocated wings after mechanical catching

(Musilová et al., 2013). In a study by Nijdam et al. (2005), a higher percentage of DoA was reported after mechanical catching but they did not find a difference in bruises.

Considering the available evidence, based on expert opinion, the AHAW Panel concluded that mechanical catching can potentially reduce the risk of injuries compared to manual catching and inversion if it is operated correctly.

Hens in enriched cages are generally removed individually or in groups of two or three by pulling or lifting them out by one leg to form a bunch in each hand. Injuries might be caused during removal, due to hits against the cage entrance, food trough, perches or nest boxes. Hens may also hit cages or roof supports as they are carried down the narrow aisles of a battery house (Knowles, 1991). Hens can be injured as well when introduced into the crates. When passing through the openings, hens often open wings to resist entering and one or two wings may be broken at this point. Moreover, due to relative inactivity within cage systems and breeding for a high rate of laying, they are likely to present a variety of skeletal problems resulting in increased bone fragility and susceptibility to fracture (Whitehead and Fleming, 2000; Lay Jr et al., 2011). A large study on catching and crating of hens housed in aviaries in Switzerland (Gerpe et al., 2021) found 8.1% of hens sustained fractures and severe muscle damage. A small study comparing end-of-lay hen pathology from different housing systems, found that injuries, including fractures, after transport were almost exclusively for hens depopulated from cages rather than deep litter (barn) or free range (Keutgen et al., 1999).

Considering the available evidence, based on expert opinion, the AHAW Panel concluded that due to the bone fragility and susceptibility to fracture, the catching and crating of end-of-lay hens represents a particularly high risk of injuries.

3.7.2.2. ABMs

ABMs that are considered feasible for assessing injuries in birds during transport are given in Table 8.

Table 8: ABMs for injuries in domestic birds

ABM (animal categories)	Definition and interpretation of the ABM
Hanging, non-functional limbs, severe lameness, protruding bones	**Definition:** Birds with abnormal postures or movement, such as hanging, non-functional limbs or severe lameness, or with protruding bones. **Measurement:** Number of birds showing signs of broken or dislocated bones. **Sensitivity** is high as most of the injured birds will show hanging, non-functional limbs, severe lameness or protruding bones. **Specificity** is high as not injured birds will not show hanging, non-functional limbs, severe lameness or protruding bones
Bruises	**Definition:** Bruising is an injury that occurs after trauma, it results from a hematoma and is often without rupture of the skin. Recent bruising appears red; between 12 and 24 h after trauma, the bruise is often dark red to purple. Bruising that occurs during rearing can potentially be identified by a green colouration that occurs 24–48 h after trauma (Cockram et al., 2020). The bruises are difficult to see on the live animals but become more visible after plucking the carcass (feather removal during the slaughtering process). **Measurement:** Number of birds showing bruises. **Sensitivity** is moderate as birds might be injured without having bruises. **Specificity** is high since non-injured birds will not have bruises.
Wounds	**Definition:** Wounds comprise all lesions to the skin (skin lesion being a wound that has not yet completely healed), ranging from minor superficial punctiform spots, to scratches, to large open wounds that go deeper than the skin (Welfare Quality Network, 2019). They may occur during inversion or due to rough handling. **Measurement:** Number of birds showing wounds. **Sensitivity** is moderate as birds might be injured without having wounds. **Specificity:** is high since non-injuries birds will not have wounds.

ABM (animal categories)	Definition and interpretation of the ABM
Body parts entrapped between or inside containers (proxi)	**Definition:** entrapped wings, legs, necks or heads of birds between container doors, drawers or between stacked containers
	Measurement: number of birds with body parts entrapped in or between containers
	Sensitivity is moderate as birds might be injured without having body parts entrapped between or inside containers.
	Specificity is high as non-injured birds will not be entrapped between or inside containers.

ABMs of injuries, such as hanging, non-functional limbs, severe lameness, protruding bones, bruises and wounds are highly to moderately sensitive and specific during catching and crating, loading, unloading and uncrating of birds.

In addition, the presence of body parts entrapped between or inside containers, although not a direct ABM, can be used as a proxy to assess injuries in birds.

3.7.2.3. Hazards

Rough handling

Rough handling (e.g. grasping the bird by the neck or by one leg or wing) during catching and crating increases the risk of wing fractures and dislocated joints in broilers (Nijdam et al., 2004; Jacobs, 2016; Jacobs et al., 2017a,b). Bird impacts with hard surfaces like the edges of containers can lead to bruises or more severe injuries. This will mainly occur when staff does not pay enough attention while placing birds in the containers.

Different types of containers exist (see Section 3.3.2) and their design, mainly the shape, size of the door or drawer opening, has an impact on the ease of placing the birds into the containers. A major cause of bruising in the broiler breast are crating the birds quickly through a small opening at the top in the transport crates, and a deregulated mechanical catch system, which places the birds in the transport drawers (Grandin, 2015).

Humans have been identified as the main origin of rough handling (EFSA AHAW Panel, 2019). The quality of employees (attitude and knowledge of staff) and their proper supervision largely determines how many birds are injured (Kettlewell and Turner, 1985). Staff working under time pressure and people not given enough time for breaks and rest will increase the risk of rough handling. Inadequate number of handlers has also been identified as the origin of rough handling (Marahrens et al., 2011) and a main hazard responsible for the occurrence of bruises.

Inversion

Handling of broilers in an inverted position leads to more leg and wing fractures (Kannan and Mench, 1996), as well as dislocated joints.

Inadequate settings or too high loading speed by mechanical catching

Non-optimal settings or too high loading speed by mechanical harvesting and loading machines will increase the incidence of injuries.

Jamming or crushing of heads, wings and legs in containers

If birds put their heads, wings, legs or toes through the grid of the container, the protruding parts may get stuck between two containers or in the door during manual or mechanical handling of containers, resulting in discomfort, pain and injuries.

The risk of damage (e.g. broken wings, injuries to the back and thigh, bruises, etc.) is greater in crates with a small opening on the lid (Figure 5a) in comparison to container systems (Figure 5b,c).

Injuries may also occur when there are sharp projections inside the containers due to broken plastic or metal surrounds (AVMA, 2016; OIE, 2019).

3.7.2.4. Preventive measures

Rough handling

Avoiding rough handling of birds will reduce the risk of injuries. Training of staff to acquire the knowledge and skills required to perform their allocated tasks efficiently, but without damaging the birds, is identified as the most important preventive measure. Proper supervision of the catchers can also result in improved handling and low levels of injuries (Ekstrand, 1998).

Injuries due to hitting or pushing birds against the edges of the crate or container entrance can be prevented by using crates or containers with large doors or openings.

Containers (crates) used for moving birds should be placed as close as possible to the birds to minimise the distance they are carried. This will also reduce the risk of staff fatigue. Furthermore, staff rotation is important to reduce fatigue, which can lead to poor bird handling practices.

Loading should be performed smoothly and horizontally to prevent tilting of containers that cause birds to pile up or bunch (Grilli et al., 2015).

Inversion

Carrying birds in an upright position with their breast supported will reduce the risk of dislocated joints and fractures in legs and wings. The results of Kittelsen et al. (2018) showed that the abdominal and upright method did not provoke broken legs, and was associated to less placement of birds on their back in the drawers and less DoA.

Inadequate settings or to high loading speed by mechanical catching

Settings of catching machine should be adjusted in order to prevent injuries at catching, moving and crating.

Jamming or crushing of heads, wings and legs in containers

Careful supervision is needed when placing the birds into the containers and before and after loading and unloading the containers.

For this reason, great care must be taken upon loading the birds into and unloading them out of transport crates. Management and loading speed are critical when using crate systems.

Containers in good conditions, without broken plastic or metal parts protruding inwards, reduce the possibility of injuries and bruises. In drawer systems, a common problem is head entrapment. This is caused by rough loading on the farm, or unloading in the slaughterhouse, or/and poor design of the drawer rack (AVMA, 2016). The hazards 'rough handling of the containers' and 'jamming or crushing heads and legs in containers' can both appear at this stage and lead to pain and fear.

3.7.2.5. Corrective and mitigative measures

Rough handling

There are no corrective measures once birds are injured. The welfare consequences of injured birds can be mitigated by identifying injured birds during the processes of catching and crating and prevent them from travelling.

Inversion

If birds are inverted, the risk of bone lesions (dislocated joints or fractures in legs or wings) can be reduced by catching and carrying them by both legs, prevent wing flapping and by minimising the duration of inversion, by placing containers close to the birds.

There are no corrective measures once birds are injured. The welfare consequences of injured birds can be mitigated by identifying injured birds before the processes of catching and crating and prevent them from travelling.

3.7.3. Restriction of movement

3.7.3.1. Description

Restriction of movement occurs when a bird has insufficient physical space to move freely (e.g. walk, stretch its wings), sit or stand comfortably. Domestic birds may experience negative affective states such as pain, fear, discomfort and/or frustration when they are unable to behave normally. For example, experiments by Lagadic and Faure (1987) found that laying hens would work to gain access

to greater floor space and Nicol (1987) noted rebound behaviour after hens had been confined in small cages. Restriction of movement is a welfare consequence inherent to the transport of domestic birds, as they are confined in containers and the space allowance in all dimensions (available floor area and height) is low. In addition to a high prevalence, the duration of this welfare consequence is from crating on farm to the emptying of containers at the destination. This means that, in addition to the journey time, animals are in the containers before the journey starts and during lairage at destination, which can be many hours. The severity depends on the space available for the animal to adopt natural postures, rest comfortably, thermoregulate and move around. Severity is regarded as high when all animals in a crate cannot adopt natural sitting or resting postures at the same time with their heads comfortably extended, and moderate when they are unable to shuffle around within the container and hold the wings slightly away from the body.

Restriction of movement may impact flocks of birds differently according to the conditions they have experienced on farm and with the category of bird. Some birds, such as fast-growing broilers, do not express much locomotory behaviour (Schwean-Lardner et al., 2012) and have a low space allowance on farm. Thus, the restriction of movement in containers may be perceived by broilers as less adverse than for other birds such as game birds or hens, which are more active and often provided with access to outdoor environments. Experimental studies show an effect of the degree of restriction of movement on the stress response in game birds when confined in small transport containers. Suchy et al. (2007) and Voslarova et al. (2006) found that the levels of stress associated with catching, crating and 4-h transport of 9-week-old pheasants (mean body weight 0.8 kg) depended on the floor space allowance within the crate. A floor space allowance of 195 cm^2/kg of body weight induced a stress response (significant increase in plasma corticosterone concentrations, decrease in the heterophiles/lymphocytes (H:L) ratio), whereas an allowance of 290 cm^2/kg of body weight did not induce significant changes in blood physiology when compared to pretransport levels, suggesting such game birds may benefit from a larger space allowance.

The height of the containers also affects the restriction of movement. Vinco et al. (2016) conducted a field trial under commercial conditions to compare the welfare of large broilers (average weight 3.4 kg and head height when standing 38 cm) transported in cages of two different heights, 23 cm (not allowing them to stand up) and 46 cm (allowing them to stand in a natural position). Video scan analysis proved that during transport (vehicle moving) there was no difference in the proportion of birds (4%) attempting to stand up between both types of crates. All birds maintained a sitting position when the vehicle was in motion even in the crates with sufficient height to permit standing. From the video records, it was possible to see that during stops, some of the birds stood up. Once moving restarted, and in particular during loading and unloading of the crates, these birds, in an attempt to maintain their balance in a standing position, reacted with vigorous wing flapping and climbed on each other.

Ellerbrock and Knierim (2002) measured the standing height (defined as the highest point of the back) of turkeys of different weights and found that it was 49 cm for 8.4 kg turkeys of 11 weeks, 55.9 cm for 15.9 kg turkeys of 17 weeks and 55.6 cm for 20 kg turkeys of 21 weeks. The slight reduction in height at 21 weeks compared to 17 weeks was explained by reduced plumage in the older birds. Most common containers for the transport of turkeys have an internal height of 30 to 40 cm that clearly does not allow the birds to assume a standing position. Vinco et al. (2016) carried out a field trial under commercial conditions, where the welfare of turkey toms transported in cages of three different heights (39 cm, 77 cm and 116 cm) was compared. Average bird weight was 18.9 kg, the head height of the siting bird was on average 36 cm and its back height in the same position 27 cm. Thus, the birds were able to sit with their head in a natural position in the lowest height crates (39 cm). The head height of the birds standing was between 75 and 82 cm and birds were therefore able to stand with some limitation in the 77 cm high crates and to stand comfortably in the 116 cm high crates. The stress parameters used for the animal welfare evaluation (H:L ratio and corticosterone level) appeared to support the beneficial effect of standard crates (39 cm) over the use of higher ones where they have the possibility to stand. Post-mortem results confirmed the same, the provision of extra head space to stand resulted in an increase in injuries including scratches.

The responses of 11 kg female turkeys transported for 76 min over an 86-km route in crates at commercial height of 38.5 cm were compared with those transported in crates 77 cm in height, which enabled a standing position (Di Martino et al., 2017a). Floor space allowance averaged 109 cm^2/kg. The authors recorded five rising attempts/bird/h in response to vertical constraint (in containers of 38.5 cm) versus under one rising attempt/bird/h in the higher crates. Behaviours such as mounting, wing-flapping and balance loss were more frequently observed in crates enabling a standing position

and 36% of the female turkeys maintained a standing position in the higher crates during transport. Based on the higher incidence of traumatic lesions and haematological stress indicators in the crates, enabling standing position, the study concluded that increased vertical space allowance was likely to impair rather than to improve welfare with a crate height of 77 cm in comparison to 38.5 cm for female turkeys weighing 11 kg. These findings concur with the results of a Canadian study by Wichman et al. (2012), who examined the welfare of male turkeys of 11–12.5 kg live weight transported 4 per crate of either 40 cm or 55 cm in height (floor space was 118–134 cm^2/kg). Observations on the birds' behaviour during lairage, as well as recordings of carcass damage and meat quality were carried out after four commercial journeys of 40–90 min duration (total time in crates was 4–7 h). Turkeys in 40 cm crates panted more and lay down more than birds in 55 cm crates during lairage, which was maintained at 10–16°C. About 20% of birds from the 55 cm crates had scratches on their backs, which was significantly more than birds from the 40 cm crates (~ 5%). In a previous experiment of Wichman et al. (2010), no differences in physiological measures (aspartate aminotransferase, creatine kinase and H:L ratio) were found between turkeys not transported but confined for 6 h in crates of either 40 or 55 cm in height. Their movement was very restricted in the 40 cm crates, whereas in the 55 cm crates it was possible for the turkeys to stand up and move around almost as much as if kept in free height, even if they were not able to stretch their necks while standing. Thus, a 55-cm height allowance restricted head movement of 11–12.5 kg turkeys.

3.7.3.2. ABMs

ABMs to assess the welfare consequences regarding the restriction of movement are defined in Table 9. Both of them are specific and sensitive during crating and before loading, to assess restriction of movement in birds.

Table 9: ABMs for restriction of movement

ABM	Definition and interpretation of the ABM
Sitting posture	**Definition:** All birds are sitting at the same time in a natural position without overlapping. If the birds cannot sit all at the same time and/or overlap, they will experience restriction of movement.
	Measurement: The body posture of birds can be visually inspected at loading, unloading and during stops. Nevertheless, it is difficult to assess the body posture of birds while they are inside the containers, as only the birds in the periphery of the outer containers can be assessed.
	Sensitivity is high. If movement is restricted, not all birds will sit in a natural position and some of them will overlap.
	Specificity is high. Birds without restriction of movement will sit at the same time without overlapping.
Head posture	**Definition:** Birds sit with their necks extended without their heads touching the ceiling. If the head is touching the ceiling birds will experience vertical restriction of movement.
	Measurement: The head position of birds can be visually inspected at loading, unloading and during stops. It is difficult to assess the head position of birds while birds are inside the containers, as only the birds in the periphery of the outer containers can be assessed.
	Sensitivity is high. If vertical movement is severely restricted, birds will not be able to extend their necks and their head movement will be constrained.
	Specificity is high. If vertical movement is not severely restricted, birds will extend their necks to keep their heads up.

3.7.3.3. Hazards

Insufficient space allowance and height in the containers are the hazards affecting the restriction of movement of birds during transport.

Insufficient space allowance

One of the main hazards for restriction of movement is insufficient floor space, i.e. too many birds being loaded per container. Furthermore, too high stocking density can increase the risk of heat stress (see section on heat stress).

As emphasised by Ellerbrock and Knierim (2002), body space (i.e. the area occupied by the body) is the absolute minimum required by each bird and is the only space which can be measured directly. Planimetric studies for different species measure the area covered per bird. A planimetric study by Giersberg et al. (2016), using overhead photography, measured the floor space covered by broiler chickens depending on their live weight and position. From this study, it appears that the floor space covered (cm^2) by the broilers in a squatting or sitting position, has a close linear correlation (correlation coefficient = 0.82) with live weight (kg), expressed in the equation area (cm^2) = 91.80 + 120 × live weight (kg). Also using planimetric photographic techniques, Ellerbrock and Knierim (2002) investigated the static space requirements of male meat turkeys and the area covered by them in a standing position. They found that there was a strong relationship (correlation coefficient = 0.86) between their live weight and the area covered, and a formula was derived for the calculation of the area covered by turkeys weighing between 7 and 21 kg: (cm^2) = 252 × live weight ($kg^{2/3}$).

Animals occupy space in three dimensions but as it is hard to measure volume, their live weight is often used as a proxy measure because it follows a close relationship with the space occupied by an animal, which may be described by allometric equations. In this opinion, space allowances are given as cm^2 per bird, estimating them from allometric equations for space allowance:

$$\text{space allowance}\,(cm^2/bird) = k \times W^{2/3},$$

where k is a constant varying for different livestock and postures and W represents live weight in kilograms (Petherick and Phillips, 2009). Using an exponent of 2/3 assumes that all domestic birds have a similar shape. Therefore, allometric equations provide estimates of space requirements rather than definitive measurements of the area covered by a bird when sitting or changing position.

Petherick and Phillips (2009) suggest a k-value of 270 to estimate the minimum floor space allowance to enable mammals to simultaneously adopt a resting position. Baxter (1992) proposed a k-value of 290 instead of 270 for poultry to simultaneously adopt a sitting/squatting position based on studies from Bognor et al. (1979) and from Dawkins and Hardie (1989) who both measured the space occupied by hens. The difference in calculating occupied space between poultry (requiring k-value of 290) and other domestic animals (e.g. pigs requiring a k-value of 270) can perhaps be explained by the birds' plumage (Baxter, 1992).

The allometric equation with a k-value of 290 provides more space than derived from planimetric measurements (except for pullets and well feathered hens which cover more space with their feathers) and therefore the possibility to change posture, shuffle around and slightly lift their wings away from the body.

For heavier turkeys, there is less agreement between predicted and measured allowances. Using a constant k-value of 290 in the allometric equation, light turkeys of 9 kg live weight would require 140 cm^2/kg and of 22 kg would require 104 cm^2/kg live weight. For a light turkey this space allowance is about 15% more than the planimetric results in a standing position and for a heavy turkey is approximately 20% more than the area covered according to the planimetric results reported by Spindler et al. (2016), who found that the measured space required for standing broilers is only slightly less than for sitting.

Using the imprecise technique of a grid superimposed over videos of well-feathered end-of-lay hens sitting in their cages indicated that a mean 318 (range 290–335) cm^2 were required for hens (Hy-line brown) to lie (Mench and Blatchford, 2014), which, at the study mean weight of 1.6 kg, equates to 199 cm^2/kg. Planimetric measurements of hens by Spindler et al. (2016) found that poorly feathered hens occupied on average 242 cm^2/kg when sitting and moderately feathered hens about 290 cm^2/kg (assuming an additional 17% space compared to standing birds).

The allometric equation for sitting broilers (k-value of 290) provides results that are in line with the planimetrically measured floor space covered by ducks in a sitting position and with poorly feathered laying hens of approx. 1.9 kg, but the space prediction is less than that occupied by well-feathered hens and pullets (Spindler et al., 2013, 2016).

Results from different space allowance in studies for different bird categories and weights are presented in Table 10, second column. The equivalent space requirements based on the allometric equation with a k-value of 290 (Baxter, 1992) has been calculated. Based on planimetric data from the literature using overhead photography, laying hens and pullets, especially when well feathered, cover more floor space than predicted by using weight-based equations.

Table 10: Space allowance measurements and stocking density required for different bird categories and weights on farm or during transport

Category and weight of bird	Space allowance (cm²/kg) (from literature – based on mean planimetric measurements)	Space allowance (cm²/kg) (calculated using the allometric equation $cm^2 = 290 \times$ live weight $(kg)^{2/3}$)
Pullets, 1.5 kg	310[1]	254
Laying hens 1.7 kg (poor feather cover)	242[2]	243
Laying hens, 1.8 kg (average feather cover)	268[2]	239
Laying hens, 1.9 kg (lying in their cages)	199[3]	235
Broilers, 1.7 kg	188[2]	243
Broilers, 3.2 kg	161[4]	198
Peking ducks, 3.0 kg	212[2]	202
Muscovy ducks, 4.4 kg	199[2]	178
Turkeys, 8.6 kg	123[5] (standing birds)	143
Turkeys, 15.9 kg	106[5] (standing birds)	116
Turkeys 22.5 kg	80[2]	104

1: Spindler et al. (2013).
2: Spindler et al. (2016).
3: Mench and Blackford (2014).
4: Giersberg et al. (2016).
5: Ellerbrock and Knierim (2002).

The available floor space in the containers can affect the risk of injuries. Miklos (2008) reported that space allowances for broilers and hens of 207 cm²/kg or more led to higher levels of traumatic injuries (0.51%) in journeys in Hungary averaging 7 h duration than a reduced space allowance, with associated levels of 0.31%. Miklos (2008) also reported reduced levels of injury for transport in cold weather (0.26% vs. 0.48%), suggesting that huddling behaviour was protective against trauma, however, their retrospective study couldn't determine whether space allowance was reduced by industry practices in cold weather. The analysis of retrospective slaughter data from 64 short-journey consignments in Portugal of broilers (1.85 ± 0.26 kg) (Saraiva et al., 2020) linked increased levels of bruising with increased space allowance: the probability of more than 4% bruising prevalence increased linearly from about 20% at 180 cm²/kg to around 60% at 230 cm²/kg. Saraiva et al. (2020) suggested that transport containers with less space per bird can be more suitable in preventing bruising as birds support each other's bodies, reducing falling or the need to spread the wings and legs to keep balance. Therefore, it is suggested that a high space allowance may increase the risk of injuries in birds. However, in the same paper the author demonstrated that the bruises did not increase with transport duration indicating that bruises were more likely to have occurred on farms during catching, crating and loading.

Insufficient height

Insufficient height of the containers prevents birds from adopting natural sitting postures with the head extended and the possibility to move and change posture within the container.

Kinematic analysis of laying hens kept in cages indicate heights of 35 cm are sufficient for standing with the head in a natural position (Mench and Blatchford, 2014). However, the description of current practice provided by the German competent authority[4] is that '*For young laying hens, the minimum height of 23 cm is not sufficient in any case, but is a common transport practice (all containers of well-known poultry marketers have exactly this dimension). Even when poultry are squatting, the tail*

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[4] Obtained through a public consultation of the draft scientific opinion.

feathers stick-out through the grid floor into the boxes above', indicating that height of containers are sometimes too low to allow this position, at least for laying hens.

Although the consequences in terms of welfare in birds unable to stand in containers with restricted height allowance have not been extensively studied, it is reasonable to assume that the current practice of transporting poultry in containers which prevent them from standing in a natural position, thus restricting their movement considerably, leads to negative affective states such as frustration and discomfort. On the other hand, according to the Consortium of the Animal Transport Guides Project (2017) and the Farm Animal Welfare Council (2019), *'higher crates, where birds are able to stand, result in falling or piling up, leading to injuries'*. Therefore, there is a need for more research to determine bird preferences for both floor space and head height during transport in containers and the effect of space allowance on trauma, behaviour, air movement, ventilation and thermal stress for various species and categories of poultry.

3.7.3.4. Preventive measures

Restriction of movement is a welfare consequence inherent to the transport of domestic birds, as they are confined in containers and therefore it cannot be prevented, only mitigated.

3.7.3.5. Corrective and mitigative measures

Insufficient space allowance

The severity of the restriction of movement can be mitigated by providing enough floor space per bird in the container to ensure all birds can adopt a comfortable sitting position at the same time, and be able to shuffle around. The AHAW Panel concluded that based on the available planimetric results and the fact that animals during transport need more space than the covered area to move, change posture and also to thermoregulate (*see Section 3.7.6 on heat stress*), minimal available floor space for birds can be calculated using the allometric equation: space allowance (cm^2/bird) = 290 × body weight (kg$^{2/3}$) (adjusted based on Baxter, 1992) which sometimes provides birds with more space than the planimetric results. For game birds, pullet and laying hens up to 2 kg, especially when well feathered, more floor space than predicted by using the allometric equation is needed and the planimetric measurements are preferable. Minimum space allowances recommended by the AHAW Panel to mitigate restriction of movement are given for various categories of poultry, taking the lowest live weight of the range as a precautionary principle (e.g. for broilers of 2–3 kg the calculation was done for 2 kg). Related stocking density is also provided in Table 11. In the table, the equation is used to calculate the space (cm^2/bird) for a bird of a certain live weight. For practical use the results of the equation are transformed to kg/cm^2 by dividing the outcome of the equation by the average live weight of the birds.

Table 11: Recommended space allowance (cm^2/kg) and related stocking density (kg/m^2) for different bird categories at different live weights during transport

Bird category	Live weight of bird (kg)	Space allowance (cm^2/kg) (calculated with the allometric equation)	Stocking density (kg/m^2) (based on space allowance)
Quails	250 g	458	21
Pullets, game birds	Up to 1.5	310*	32
End-of-lay hens (average feather cover)	Up to 2	268*	37
Broilers, end-of-lay hens (poor feather cover)	Up to 2	231	43
Broilers, end-of-lay breeders, Ducks	2–3	202	49
Broilers, end-of-lay breeders, Ducks	3–4	184	54
Broilers, Ducks, Geese	4–5	171	58
Turkeys, Geese	Up to 9	140	71
Turkeys, Geese	9–15	119	84
Turkeys, Geese	15–22	105	95

*: This is instead calculated with the planimetric equation because pullets require more space than that resulting from the allometric equation.

The AHAW Panel also concluded that the proposed space allowance represents a balance between providing adequate space and reducing the risk of injuries. From the perspective of restriction of movement alone, these allowances are the minimum to mitigate the welfare consequences and birds will still experience restriction of behaviour and negative affective states such as frustration. However, space allowance implies additional welfare consequences which need to be considered together with the duration of confinement (see sections on thermal stress, resting problems and injuries).

Staff (i.e. the catchers) should ensure the correct number of birds are loaded according to the recommended space allowances. If any container is overcrowded, some birds should be taken out.

Insufficient height

Based on the available evidence, the AHAW Panel concluded that the height of the containers should allow the birds to keep their heads in a natural raised position when sitting and to be able to change position, without allowing them to stand in natural position. Minimum heights of the containers recommended by the AHAW Panel are provided for the different animal categories in Table 12. The recommended heights are based on expert opinion following the limited evidence that shows that the heights of some commercially used containers are not sufficient for certain categories of birds.

The minimum recommended height for turkeys was retrieved from the EURCAW-Poultry-SFA to the query reference (001–2022) (https://www.eurcaw-poultry-sfa.eu/en/minisite/sfawc/questions-eurcaw-q2e) that reported the minimum height of transport cages for which turkeys of different weights can sit with the head in a comfortable position (i.e. extended without touching the ceiling), able to change position without risk of injuries, thus with sufficient size and height to maintain this position but preventing them to stand up and climb on each other. The observations were performed on 43 turkeys, from 11.5 to 19.2 kg. Due to the high variability between turkeys of the same weight category, the minimum recommended height is the maximum height of the turkey assessed in each category. This height will also allow air circulation above the birds back while sitting if the vehicle is ventilated.

Table 12: Current commercial heights of the crate and minimum height recommendations from the EFSA experts according to domestic bird category

Bird category	Weight of bird (kg)	Current height (cm)	Minimum recommended height (cm)
Broilers	Up to 3.4	23–25.5	23
Broilers	Heavier than 3.4	23–25.5	25
Turkeys	From 11 to 13	31–42	40
Turkeys	From 14 to 19	31–42	45
Laying hens	Up to 2	23–25.5	25
Pullets	Up to 1.5	23–25.5	25

3.7.4. Sensory overstimulation

3.7.4.1. Description

Sensory overstimulation, including motion stress (see Section 3.7.5) occurs when one or more of the five senses becomes overstimulated:

- **Auditory system (hearing)**: Loud and/or sudden noises. Continuous high intensity/loud sounds. Simultaneous sounds from multiple sources
- **Visual system (sight)**: High intensity (bright) lights, rapidly changing images, flashing or strobing lights. Environments with rapid movement resulting in rapidly changing images or novel images.
- **Olfactory and gustatory systems (smell and taste)**: Powerful or noxious odours or tastes. Toxic or caustic gases or vapour.
- **Somatosensory system (touch, pain and position/location/mechanical activity)**: Includes the following sensors.

- ○ **Mechanoreceptors** respond to mechanical stimuli such as the bending or stretching of receptors.
  - ○ **Proprioceptors** respond to movement, action and location.
  - ○ **Thermoreceptors** respond to temperature changes.
  - ○ **Nociceptors** respond to stimuli that result in the sensation of pain.

- **Vestibular system** (equilibrioception): Intense motion or acceleration, vibration, disturbances in balance. Consequences, loss of balance, dizziness, disturbed locomotion, nausea and motion sickness.

Because of the importance of motion stress during transport this welfare consequence and the sensory systems involved in motion stress, the vestibular system together with the proprioceptors of the somatosensory system will be dealt with in a separate chapter. The other sensors of the somatosensory systems are considered together with the welfare consequences (i.e. nociceptors and mechanoreceptors with pain and thermoreceptors with thermal stress).

Sensory overstimulation, meaning that the animal experiences stress and/or negative affective states such as fear, and/or discomfort and/or stress and/or stress due to visual, auditory or olfactory overstimulation by characteristics of the physical environment, is regarded a clearly relevant welfare consequence for all animals transported in containers. The birds are exposed to sensory overstimulation during the stages of loading, journey, arrival and uncrating; therefore, the welfare consequences can prevail throughout the entire transport operation. The severity of sensory overstimulation is considered as moderate, meaning that the welfare of the animals is affected, but that this effect is less severe than, e.g. the welfare consequences of injuries, such as bruises and dislocated or broken bones. All birds might be subjected to it.

When one or more of the senses is/are stimulated excessively, the result can range from induction of pain to a degree of discomfort that imposes a stress on the individual that may precipitate behavioural changes and demands associated with fight or flight and escape. These responses must be considered undesirable in relation to animal welfare and therefore such risks should be reduced or avoided.

When the sensory system receives inputs, the central nervous system integrates the afferent information and initiates appropriate efferent outputs to ensure control and homeostasis. Stimulation of the receptor systems may, upon occasion, be excessive and this may be regarded as sensory overload. This can result from a large magnitude stimulation of a single sensory pathway (e.g. an excessively loud noise or extremely bright light) or may be induced by high intensity, simultaneous stimulation of more than one sensory system. Excessive stimulation or overstimulation may result from high intensity of stimulation or from long duration or continuous stimulation of one of more sensory systems by high intensity stimuli.

The consequences for animal welfare depend upon the intensity and duration of the stimuli and whether or not any stimulus results in pain or extreme discomfort. For lesser magnitude or lower duration stimuli the effects may induce fear, confusion, disorientation, frustration and in some animals, nausea and sickness. There may be a number of physiological stress responses resultant upon the excessive sensory stimulation including cardiovascular and respiratory changes, thermoregulatory responses, gastro-intestinal effects and changes in blood chemistry and stress biomarkers.

In addition to the primary problem of sensory overstimulation and the induction of aversion in the transported birds these factors can exacerbate the effects of other concurrent transport stressors such as handling stress, injury, thermal stress, restriction of movement and prolonged hunger and thirst.

3.7.4.2. ABMs

As sensory overstimulation induces a fear response, the ABMs are similar to those for handling stress (see Section 3.7.1.2), i.e. 'escape attempts' and 'distress calls'. However, assessment of these ABMs is not currently feasible during the journey stage.

3.7.4.3. Hazards

Unexpected loud sound/noise

Animals will get used to the noise of the running engine. However, sudden alterations in sound will constitute a threat to the animals' well-being and will induce fear to the birds. The catching and

crating process as well as the journey in the vehicle are stages with loud noises originating mainly from machines, the manipulation or movement of containers, or people who shout while catching the birds. There are no available data on noise level that relates to welfare consequences.

Visual stimuli

Sudden changes in environmental conditions including rapid alterations in visual environments e.g. light levels or rapidly changing images will all constitute a threat to the animals' well-being. Lighting intensity might impact bird reactivity and affect welfare outcomes (EFSA, 2004). Daylight is many orders of magnitude brighter (typically 1–25 thousand lux) than the levels in most domestic poultry housing, which is generally below 20 lx.

Noxious gases

Exposure to dust and ammonia due to disturbance of the litter during catching may impact upon birds prior to and during loading. In transit noxious gases may enter the load from vehicle exhaust fumes on busy roads when vehicle is moving slowly or is stationary.

3.7.4.4. Preventive measures

Unexpected loud sound/noise

Excessive sound or noise should be avoided or minimised in all stages of animal production, handling and transport. Therefore, strategies should be employed which reduce the risk of loud and/or unexpected sound and noise.

It is important to limit unexpected loud noises because they lead to fear and decrease the coping capacities of birds. The preventive measures will consist in staff education and training (i) to make them aware that noise at the birds' level should be avoided and (ii) to make them avoid shouting and making noise with the equipment and identify and eliminate the sources of noise. In addition, the machine should be setup correctly to avoid excessive noise.

Entering the bird house and approaching the birds calmly with minimal noise to minimise disturbance. Limiting sudden and extreme noise of equipment as much as possible. Careful selection of people with appropriate skills and the right attitude or training to acquire skills appropriate to the tasks and species of birds, would help to minimise fear when handling animals.

The noise produced by the vehicle through engine operation, air movement around and through the load and road surface contact can be minimised (although not entirely prevented) by the use of improved suspension systems and vehicle enclosure or insulation (use of curtain or solid sides with high quality ventilation). In the future the use of electric vehicles may reduce the engine sounds significantly.

Where possible, avoid overhanging trees and hedges that may hit the vehicle or trailer (HSA[5]).

Visual stimuli

Excessive visual overstimulation should be avoided or minimised in all stages of animal production, handling and transport. Therefore, strategies should be employed which reduce the risk of visual overload. Dimming of lights or catching during dark hours of the day could help to minimise flightiness and associated welfare risks. Careful selection of people with appropriate skills and the right attitude or training to acquire skills appropriate to the tasks and species of birds, would help to minimise fear when handling animals.

Catching and crating on farm with calming measures such as dim blue light (440–490 nm) or low lighting. Dimming of lights or catching during dark hours of the day could help to minimise flightless and associated welfare risks. The blue light still allows the workers to see and handle birds with precision, leading to less stress for the animals (Adamczuk et al., 2014). Avila and Abreu (2003) reported that blue light is used for catching birds as it overrides the visual capacity of the birds and they remain still, facilitating the movement of the catcher. The combination of partially dark ambience with the use of blue light has a calming effect on birds, reducing their excitement (seen as wing flapping) by 56%.

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[5] https://www.hsa.org.uk/or Humane Slaughter Association, Improving Standards in Animal Welfare at Slaughter, in Markets and during Transport (hsa.org.uk)

Noxious gases

The risk of exposure to noxious gases may be prevented by vehicle enclosure (closed curtain sides) and ensuring adequate ventilation (including mechanical ventilation).

3.7.4.5. Corrective and mitigative measures

No mitigative and corrective measures are feasible during transport, only preventive measures. Once the birds have been crated and the vehicle is moving it is difficult to decrease the sensory overstimulation of the birds.

3.7.5. Motion stress

3.7.5.1. Description

During journey, animals experience motion sickness, stress and/or fatigue due to the forces exerted as a result of acceleration, braking, stopping, cornering, gear changing, vibrations and uneven road surfaces during transport. During loading and unloading with the forklift, birds might experience acceleration, vibration and impact with the cages. Motion stress is regarded as a clearly relevant welfare consequence in the journey stage. Prevalence is high, as motion stress is likely to affect all animals in a moving vehicle. Duration depends on journey duration and onset of motion stress. Severity was regarded as moderate but increasing over time and eventually leading to fatigue.

Stress resulting from motion in transported animals may be attributed to exposure to vibrations on the vehicle, animals being subjected to a range of acceleration changes and a risk of impacts (sudden acceleration or deceleration). There is an extensive literature relating to the welfare impacts of vibrations upon animals in transit. It is well recognised that vibrations, accelerations, impacts may result in poor postural stability, muscle fatigue, exhaustion and in some cases motion sickness.

Motion sickness is a common response in some species of farm livestock including birds, during transport. Despite the paucity of data on livestock, there is sufficient evidence to believe that motion might affect animal welfare when animals are transported by road or sea (Santurtun and Phillips, 2015). In some mammals (e.g. pigs), motion sickness is characterised by signs referable to stimulation of the vestibular and autonomic nervous systems, including excessive salivation and vomiting (although not in birds). Affected animals may also yawn, whine or show signs of uneasiness and apprehension; severely affected animals may also develop diarrhoea. Motion sickness is seen during travel by land, sea or air, and signs usually disappear when vehicular motion ceases. In some animals who are repeatedly transported fear of the vehicle may also become a contributory factor due to the development of a conditioned response to the event; signs may be seen even in a stationary vehicle.

Birds (in common with most vertebrates) possess a well-developed vestibular system which may mediate any motion sickness. However, there are important differences between birds (including poultry) and other vertebrate groups that must be considered. Poultry are incapable of active (diaphragmatic driven) vomiting which is a key sign of motion sickness in other animals and so motion induced distress may be less obvious.

Secondly, the vestibular system of birds (including poultry) is very highly developed to ensure the maintenance of focus on objects (control of the axis of gaze; Cullen and Sadeghi, 2008) despite substantial body movements or motion and as such the sensitivity of this system and of the whole bird to motion induced distress cannot be easily predicted. There are three major vestibular reflex pathways regulating eye movements and balance essentially without involving cortical structures (Wilson and Maeda, 1974). The vestibulo-ocular reflex (VOR) regulates eye muscles to maintain gaze during head movements, the vestibulo-collic reflex (VCR) controls the neck muscles to support the head during movements and the vestibulo-spinal reflex (VSR) controls the muscles of the body and limbs to maintain posture and balance. These control systems are highly developed in birds (more so than in humans) which produces the well-recognised phenomenon of the 'steady cam vision' of domestic poultry. It is not known how these advantageous adaptations influence the birds' perception of movement or acceleration or how the outputs might interact with other special senses e.g. vision.

3.7.5.2. ABMs

In the context of transport there are no feasible ABMs. Loss of balance could be measured but during transport might not be feasible due to the animals not being able to stand up.

3.7.5.3. Hazards

Stimulation of the somatosensory and vestibular systems will result from vehicle motion and acceleration and exposure to vehicle vibrations resulting from movement and operation of the vehicle.

Both acceleration and vibration are a product of vehicle design and structure, mechanical operation, road surfaces and motion.

Acceleration

Displacement of an object is a vector quantity, which means that displacement has a size and a direction associated with it. The velocity of the object through the domain is the change of the location with respect to time. The average velocity is the displacement divided by the time interval. The acceleration of the object through the domain is the change of the velocity (positive or negative) with respect to the time interval. The average acceleration or deceleration is the change in velocity divided by the time interval.

It is not clear what impacts acceleration may have upon the vestibular systems of poultry. Furthermore, exposure to acceleration, deceleration and sudden braking will induce postural instability, increase the requirement for muscle activity to restore stability and will increase the risk of impacts and injuries and fatigue.

Vibration

Vibration is the movement of a body about its reference position. Vibration occurs because of an excitation force that causes motion. There are two basic types of vibrations which animals experience on a vehicle, up and down and side to side (HSA[6], Figures 16 and 17). Individual vibration signals combine to form a complex time waveform showing overall vibration. Vibration has been shown to alter animal behaviour and induce physiological changes as well as to cause effects at the cellular and molecular level. For these reasons, both environmental factors have a considerable potential to alter animal welfare status (Reynolds et al., 2019). Determining the specific levels of vibration that will alter well-being is complex, as species will respond to different frequencies and have varying frequencies where they are most sensitive. Vibratory movement has a direction (generally in three planes), a magnitude (how far) and a velocity (how quickly – what rate). Vibration in each of the three planes normally applicable must be considered to determine if there are differences to bird sensitivity to each plane of vibration. There is significant research demonstrating that certain frequencies of vibration often encountered on commercial transport vehicles are aversive to birds. The vehicle, with its engine running, will vibrate in small, rapid, regular movements. The animals will get accustomed to it over time and these vibrations should not cause a problem (HSA advice). However, vibration below 5 Hz is perceived as particularly aversive by chickens (Randall et al., 1997; Abeyesinghe et al., 2001) and this coincides with the fundamental frequency of most vehicles used for poultry transport, which is 1–2 Hz. The vibration during the transport causes fear, anxiety and mental stress on chicks (Carlisle et al., 1998). Oscillation and vibration frequency during transport of broilers varies from 0.5 to 50 Hz and that amount of vibration has been considered to represent adverse conditions for the birds (Randall et al., 1997).
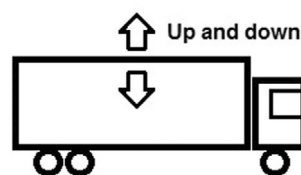
Figure 16: Up and down vibration in vehicles. Source: HSA
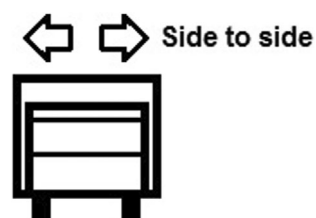
Figure 17: Up and down vibration in vehicles. Source: HSA

Further research is needed to quantify and characterise the vibrational profiles on commercial poultry vehicles and those present in transport containers to determine if vibration may constitute a significant risk to welfare in transit. It is also important to identify how the vibration regime experienced by poultry during transport may be modified if specific frequencies or magnitudes of vibration are considered to be detrimental to welfare and/or production.

The factors contributing to on-board vibration experienced by poultry in transit are:

1) Vehicle design and structure – Ride characteristics and ride comfort.
2) Vehicle suspension type and function.
3) Container and modular design and structure.
4) Nature of road surfaces and road types and location (small or larger roads with hills, bends and interrupted progress leading to variable speed ad frequent and/or sudden stops).
5) Driver style and training.

Vehicle ride

The vehicle ride results from the interaction of vibration and accelerations with the structure of the vehicle and in particular the suspension system. Therefore, it is distinct (although results from) the environment vibration. Fundamentally understanding the vehicle ride characteristics can provide the basis for reducing the stressful stimuli.

A feature of the impact of vibration in transport in humans is the phenomenon of tonic vibration reflex. This involves involuntary muscle contraction that occurs in response to direct or indirect application of vibration to a skeletal muscle (Hagbarth and Ecklund, 1968). Based on this, the International Organization for Standardization (ISO, 1997) proposes health guidance zones for whole-body vibration exposure bounded below by an exposure action value (EAV) and above by an exposure limit value (ELV). The vibration levels between the EAV and ELV may cause health risks, while vibration levels above the ELV are likely to cause health issues. However, there are not details of the induced pathologies.

Studies carried out by Peeters et al. (2008) in pig transport revealed that a 'wild' driving style produced longitudinal and lateral accelerations that affected heart rate, salivary cortisol and behaviour, but no correlations were found between vibrations, muscle fatigue and the non-ambulatory condition. Morris et al. (2021) suggested that vibration forces experienced by pigs during transportation may result in post transport 'non-ambulatory condition' through the induction of muscle fatigue. Morris et al. (2021) collected data of the three-axis acceleration, temperature and relative humidity profiles from six locations within commercial vehicles transporting slaughter weight pigs. Data over the entire transport journey for each load were processed and averaged. The study presented a mathematical approach that could readily be developed for application to other livestock including domestic fowls and rabbits. The approach is fundamentally based upon the techniques outlined in ISO 2631-1 (1997) and ISO 2631-5 (2018) technical standards for evaluating human exposure to whole-body vibrations, as there are no guidelines as how to analyse livestock data. While the reference values in these standards are used to model human comfort and health, the techniques themselves can be used to quantify the vibration environment without reference to the species experiencing the vibration. The main quantity used to evaluate vibration exposure was the root-mean-squared (RMS) acceleration value.

The values in Table 13 indicate the comfort level at each range of values of RMS classified by the corresponding definitions for humans (Values taken from ISO 2631-1, 1997) and applied to pig transport. Further research is required to allow validation of the application of different comfort states to the values of RMS for domestic birds and rabbits.

Table 13: Comfort level at each range of root-mean-squared (RMS) acceleration value

RMS, m/s^2	Comfort level
< 0.315	Not uncomfortable
0.315–0.63	A little uncomfortable
0.5–1.0	Fairly uncomfortable
0.8–16	Uncomfortable
1.25–2.5	Very uncomfortable
> 2.0	Extremely uncomfortable

3.7.5.4. Preventive measures

The reduction or prevention of motion stress can only be achieved by changes to vehicle design and structure or by attention to driving style, type of road and vehicle operation and maintenance. According to HSA, motion stress due to acceleration can be limited by:

- accelerating slowly and smoothly.
- anticipating hazards.
- avoiding sudden braking.
- smooth changes of gear.

Excessive up and down movement can be limited by:

- maintaining correct tyre pressures.
- maintaining the vehicle's suspension system.

Side to side movement can be limited by:

- correct maintenance of the vehicle.
- driving according to the quality of the road.
- taking corners with care.
- avoiding stopping or overtaking.

Many of the features of the vehicles and their basic operation cannot be altered to modify the vibrations imposed on transported birds. The nature of the suspension (e.g. leaf, air or hydraulic) may offer a strategy that can 'improve the ride' but the fundamental frequencies of vibration may not alter significantly. However, it may be possible to recommend a preferred suspension type i.e. the use of air or hydraulic suspension in preference to leaf suspension although this still has to be fully evaluated by experimentation rather than by intuition. Other recommendations and advice will include the driving style (e.g. calm driving avoiding sudden acceleration and breaks) and the training necessary to improve the driving style. It may also be possible to examine the design of containers and modules to reduce the risk of untoward vibrational regimes or to attenuate their transmission.

There are now several technologies available to allow monitoring of vibrations and accelerations on vehicles and these would allow recording and reporting of these variables on commercial poultry journeys. The effective use of such technology would only be possible if limits could be defined for acceptable ranges and limits for the frequencies and magnitudes of vibrations and accelerations that might affect the welfare of the birds in transit. Finally, for effective regulation the variables in question must be amenable to control or manipulation. This might require changes to vehicle, module and container design and might not be controlled within a journey. Thus, variables such as driving style, braking, accelerating and cornering/vehicle speed might be usefully monitored and regulated whereas vibrations resulting from natural frequencies of the vehicle bed and structure or from road surfaces might not be amenable to control. Advice might be issued relating to the choice of routes or types of road (surface) to be followed but only recommendations relating to the type of suspension to be employed on livestock vehicles would have an effect upon fundamental vibrations.

The advent of Microelectromechanical Systems (MEMS) devices for vibration and acceleration measurement and recording should provide the basis for commercially applicable systems (Looney, 2022). Thus, devices may be attached to the animal transport vehicles beds or chassis and to the transport containers or compartments to allow continuous monitoring and recording of vehicles motion, vibration and driving style. This will facilitate improvements in vehicle design and operation and driver training.

3.7.5.5. Corrective and mitigative measures

Mitigative measures will include all comments relating to driving style, driver training and education, attention to acceleration and breaking, possible improvements in vehicle bed and suspension system designs and minimisation of stressful co-factors arising from other aspects of sensory overstimulation.

3.7.6. Thermal stress

3.7.6.1. Description

Poultry are homoeothermic with the ability to maintain their body temperature constant within a narrow range. The core body temperature (CBT) of inactive birds is ~ 41–42°C (Mount, 1979), as the normal CBT of a bird can fluctuate by ±1°C (Dadgar et al., 2010).

Thermal stress means that the animal experiences stress and/or negative affective states such as discomfort and/or distress, when exposed to high or low effective temperatures. The effective temperature perceived by an animal is a combination of environmental conditions affecting the rate of heat exchange between the animal and the physical environment or surroundings. These are air temperature (or dry-bulb temperature), humidity, air velocity and radiant flux (Rashamol et al., 2019). The rate of heat exchange between an animal and its environment is also affected by the characteristics of the animal itself e.g. the surface temperature, water vapour density at the exchange surface and the properties of those surfaces and their area and orientation relative to air movement. These will all influence the effective temperature and should, in theory, be considered during transport.

The approach of this Opinion is based on thermal physiological concepts as described by EFSA (2004) (Figure 1) and originally formulated by Mount (1974). The term thermoneutral zone (TNZ) is defined as the specific range of environmental temperatures within which the energy requirement of an animal is minimal and constant. In the TNZ, body temperature is kept constant through the regulation of heat loss (Figure 18).
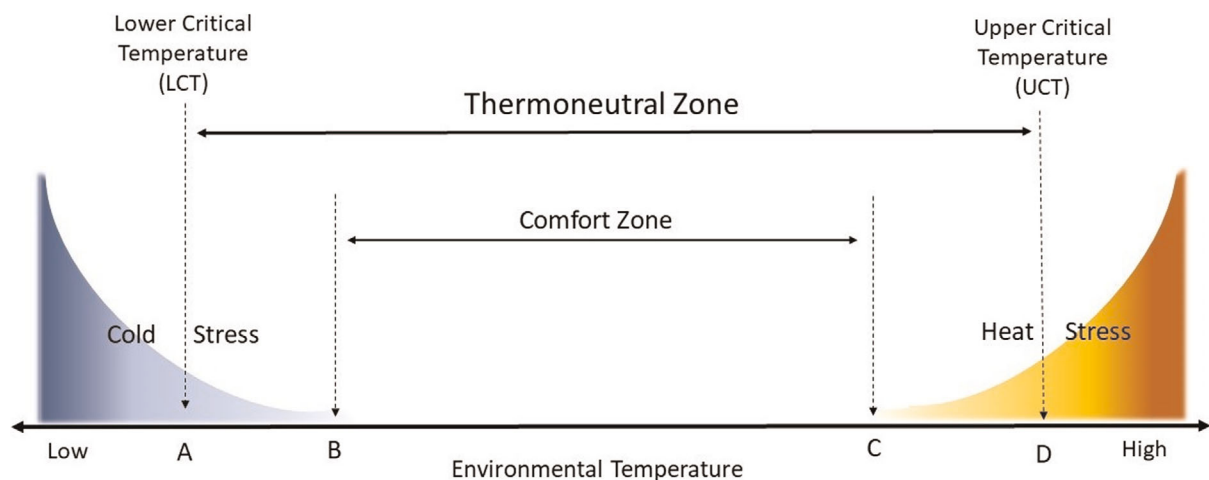
Figure 18: Schematic representation of the thermal neutral zone and the comfort zone as a function of the environmental temperature, considered especially relevant for the assessment of animal welfare during transport (Adapted from EFSA, 2004)

The TNZ is delimited by the lower critical temperature (LCT) and the upper critical temperature (UCT).

Within the TNZ, the thermal comfort zone (TCZ; between B and C in Figure 18) is defined as the environmental temperature which ensures (1) maintenance of a constant normal body temperature, (2) heat production is constant and (3) any thermoregulatory effort is not 'unpleasant'. In the TCZ, neither metabolic rate nor animal behaviour are activated in any way to keep body temperature within the normal range (Silanikove, 2000; Hodgson, 2014; Rauw et al., 2020). Above the TCZ (between points C and D in Figure 18), or below the TCZ (between A and B) poultry respond to an increasingly warm or cool environment, by making behavioural changes such as extending their wings, seeking shade or huddling and at the same time physiological adjustments such as reducing or increasing blood flow to their comb, wattle and other appendages (Chang et al., 2018). Although these changes do not measurably affect their metabolism (heat output), the animal may be aware of them and its affective state altered.

In poultry, the range of temperatures that the TNZ encompasses are not the same in all circumstances: rather, it depends on genetics and physiological status, body weight, the degree of feather cover, acclimatisation, feed and dehydration status (Dawson and Whittow, 2000; Tao and Xin, 2003) as well as other environmental factors (Lara and Rostagno, 2013). As an example of acclimatisation, in an experiment exposing broilers to various in-house temperatures, Arieli et al. (1980) found that a 10°C change in the mean daily temperature during the experimental period caused a 3°C change in the upper critical temperature (UCT) and an 8.5°C change in the lower critical temperature. Thus, birds transported under similar temperatures to those experienced on farm are more likely to be able to tolerate them.

Furthermore, the scientific literature underpinning the model shown in Figure 18 is based on studies involving a certain level of feed intake and freely available water under stable or resting conditions. As reviewed by Bracke et al. (2020), care should be taken when extrapolating findings obtained from experiments aimed at determining optimal rearing/housing rather than transport conditions. During commercial transport, animals are often exposed to factors that may act as stressors and/or limit their possibility to thermoregulate using mechanism present in non-transported control conditions. Prevalent transport conditions for poultry include feed and water deprivation, exposure to vibration and motion forces, low space allowances and highly variable ventilation rates.

In conclusion, the welfare consequences heat stress and cold stress are defined to include the associated stress, and/or negative affective states, that progressively develop when an animal is no longer in the TCZ. As explained, in the TCZ animals require no or minimal thermoregulatory effort. Above point C, animals are at risk of experiencing heat stress owing to increasing thermoregulatory efforts and above point D animals will experience heat stress associated with substantial efforts to maintain CBT. Likewise, below point B animals are at risk of cold stress and below point A animals will experience cold stress associated with substantial efforts to maintain CBT.

3.7.6.2. Thermal stress in passively ventilated vehicles

Currently the majority of vehicles used to transport juvenile and adult poultry are passively ventilated (Kettlewell et al., 2001a,b; Kettlewell and Mitchell, 2001a,b; Mitchell and Kettlewell, 2008). Air flow over the surface of the vehicle results in a pressure gradient in which there is lower pressure towards the front sides of the vehicle than at the rear sides and tail. There may be higher pressure on the front (forward face or headboard). The net effect is that in a passively ventilated vehicle air movement will tend to involve entry of air towards the rear, forward movement of air within the load towards the front of the vehicle and exit of air at the front sides of the load.

For all passively ventilated vehicles when the vehicle is stationary there is no driving force for ventilation other than buoyancy or free convection (the stack-effect) or external factors like cross winds. The buoyancy and free convection regimes will tend to create a thermal gradient within the load with the upper locations being warmer than those below. In more open configurations, cross winds may provide some beneficial effects. The problems when stationary will be exacerbated in vehicles operating with tightly fastened curtains with minimal gaps for air to enter, exit and circulate within the load.

Even those fitted with fans to aid ventilation often have the airflow dictated by two principles. The stack-effect causes heated air to rise and colder air to descend and is the dominant means in stationary vehicles. When moving, the stack effect continues to operate, particularly in areas of the load with low ventilation but is overlaid by the air flows which operate around and within a vehicle in motion. Both of these drivers of air flow may be influenced by uncontrollable external factors such as wind. There is little opportunity to control movement of air or regulate ventilation within non-curtained (open) passively ventilated vehicles except by the provision of perforated grills at appropriate inlet and outlet locations to facilitate natural pressure driven air flows (Kettlewell and Mitchell, 2001a,b; Mitchell and Kettlewell, 2008).

The most basic design is where the vehicle operates without curtains in warmer weather and with (usually) non-permeable curtains in colder weather. In fully open vehicles, flow is more difficult to predict or quantify as there are many routes through which air can enter and leave the load despite the pressure profile.

The presence of closed curtains minimises air entry and exit along the sides of the vehicle and the route for air flow will be determined mainly by the location of any openings in the structure (e.g. gaps under or around the curtains and the rear tailboard if this has openings). This results in uneven distribution of thermal conditions within the load with hot-spots (red) and cold-spots (blue) indicated in Figure 19 below. These principles apply also to vehicles upon which transport containers may be individually sheeted or curtained (Mitchell and Kettlewell, 1998; Kettlewell and Mitchell, 2001a,b; Mitchell and Kettlewell, 2008).

This area may become a cold zone in winter if insufficient or inappropriate curtaining is applied
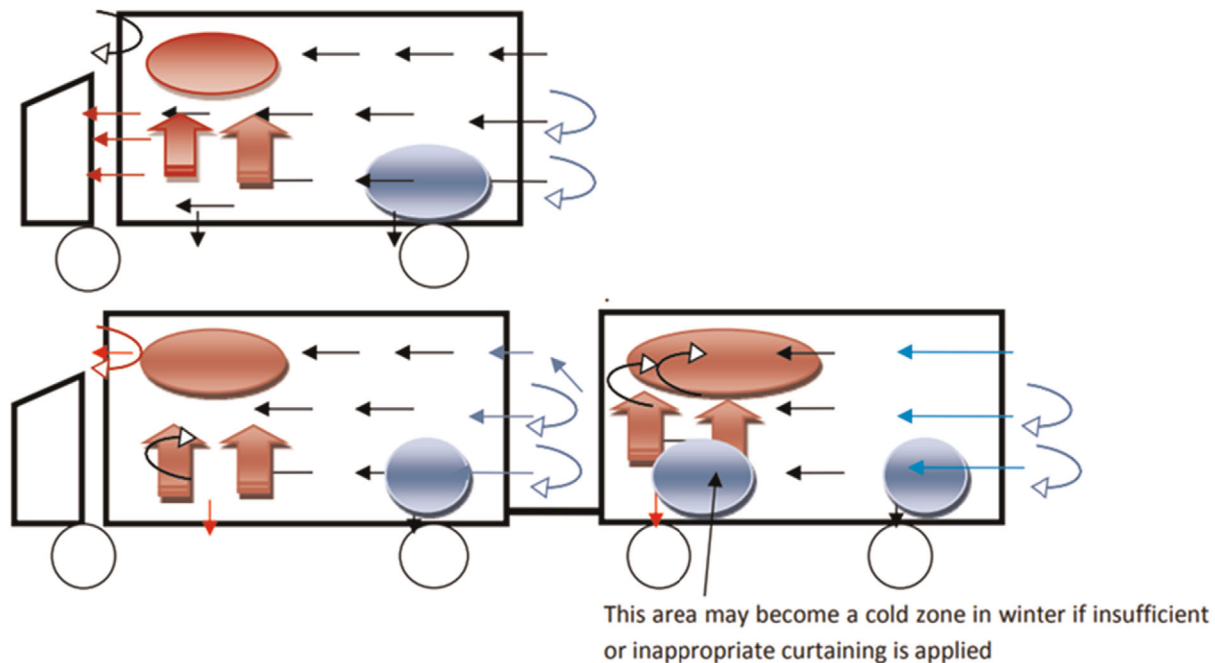
Figure 19: Predominant patterns of air flow (black arrows) in a moving passively ventilated vehicle fitted with curtains. Red areas indicate where air heated by the birds accumulates and blue areas show cold spots. Source: ANSES

All vehicles that are passively or mechanically ventilated rely on human intervention to adjust apertures, open or close curtains and select fan speeds to achieve optimal ventilation for the condition and class of bird loaded and according to the prevailing weather conditions. No current designs of vehicle transporting animals in containers can provide uniform ventilation for each animal if they are naturally ventilated. Thus, in cold weather, it is quite common for some animals within a load to be heat stressed when others are cold stressed at the same time (Knezacek et al., 2000). The thermal microenvironment can also fluctuate considerably during the transportation period in most locations.

3.7.7. Heat stress

3.7.7.1. Description

The animal experiences stress and/or negative affective states such as discomfort and/or distress when exposed to high effective temperature. The duration of exposure to heat stress can be for the entire journey or for parts of the journey. Severity will increase over time as animals experience distress and eventually fail to cope and die. Heat stress is a major welfare concern when transporting domestic birds, with prevalence being higher in the summer season.

As illustrated in Figure 18, in the TCZ there is no risk of heat stress. The risk of heat stress starts when the animal is beyond its comfort zone (at some point above point C) and progressively increases. Domestic birds will start to show behavioural (e.g. stretching wings) and physiological changes (e.g. vasodilatation). Blood vessels to the extremities (feet and comb) will vasodilate, causing increased blood flow, which can aid in dissipating heat through convection and radiation through the bare skin (Whittow et al., 1964; Midtgård, 1989). The UCT (D in Figure 18) describes the point above which animals can no longer maintain its body heat balance by conduction, convection and radiation (above D), and must significantly increase their use of physiological mechanisms to prevent a rise in body temperature above normal, e.g. vasodilation and evaporative heat loss, which requires energy expenditure and involve metabolic rate increases (Silanikove, 2000). As poultry lack sweat glands, most of the heat loss occurs through the respiratory route (Donald and William, 2002) through a process of evaporative cooling by the vaporisation of moisture from the damp lining of the respiratory tract (lungs and air sacs) (Gupta, 2011). As temperatures increase, the rate of panting increases. Under conditions of high temperature, the normal respiratory rate of adult domestic fowl (23 breaths/min) may increase by up to tenfold in response to thermal stress, reaching up to 273 breaths/min

(Kassim and Sykes, 1982). Evaporative heat loss through panting is effective only in the short-term: if panting is prolonged, there is a high risk of birds experiencing prolonged thirst and dehydration.

At the point where the capacity to lose heat is exceeded by the heat accumulation, the core body temperature will begin to rise. With continued exposure to heat, the animal's ability to cope is exceeded and it will die, as a homoeothermic animal can only survive a core body temperature a few degrees above its normal body temperature (Sjaastad et al., 2016). In broilers it has been reported to significantly increase as a result of exposure to temperatures above 30°C for 2 h (Sandercock et al., 2001; Menten et al., 2006). The point of fatality varies between individuals and species. When the CBT has increased to 44–45°C in birds, convulsions, uncoordinated movements and gradual loss of consciousness occur. If the temperature remains in this high range for a prolonged period, irreversible brain damage develops leading to death. The main cause of death (Bremner and Johnston, 1996) is due to acute and congestive heart failure (Barontini et al., 1999). Both environmental temperature and humidity are important factors affecting metabolic exhaustion and dehydration, which can cause heart failure (Elrom, 2001).

Heat stress can also alter the blood physiology of birds. Changes in parameters such as blood glucose, acid–base balance parameters, blood gas concentration, dehydration markers and the heterophil to lymphocyte (H/L) ratio may indicate that birds were subjected to thermal stressors during transport (Lalonde et al., 2021).

Fast-growing broilers have increased sensitivity to high ambient temperatures owing to their increased heat production (Sandercock et al., 2006; Renaudeau et al., 2012).

In turkeys (Havenstein et al., 2007), the larger size of the toms presents challenges in their ability to thermoregulate due to their relatively smaller surface area from which to dissipate heat (MacLeod and Hocking, 1993; Dawson and Whittow, 2000; Chiang et al., 2008). Ambient temperatures higher than 26°C were found to considerably increase DOA in turkeys (Di Martino et al., 2017a) – the temperature within the transport containers was likely at least 30°C. However, turkeys are known to be less sensitive than broilers to changes in environmental conditions (Yahav, 2000).

In comparison with chickens, meat quail have higher thermal comfort temperatures and may be more tolerant to heat stress because of a greater surface/volume ratio, suggesting greater ability to dissipate heat generated by metabolism (Sousa et al., 2014).

Relative humidity (RH) is a measure of the percentage saturation of the air with water vapour at a specific temperature in relation to the maximum water vapour that the air could potentially contain at that temperature. The water vapour content of the air is important because it has an impact on the rate of evaporative heat loss through the skin and respiratory tract (Bohmanova et al., 2007). When the ambient temperature is above the animal's comfort zone, a high level of humidity in the air will reduce evaporative heat loss and efficacy of panting and therefore make the animal feel hotter.

3.7.7.2. Indices based on temperature and humidity to predict heat stress

Several indices based on ambient temperature and relative humidity have been developed to predict thermally stressful environmental conditions. One is the temperature–humidity index (THI) that is very commonly used for livestock. A very similar index is the apparent equivalent temperature (AET), that has been used by Mitchell and Kettlewell (1998) and Mitchell (2006a,b) to assess heat stress during transport of broilers. AET is itself derived from the dry-bulb temperature (T) and the relative humidity (RH) expressed as water vapour density or pressure, according to the following equation:

$$AET = T + + \frac{\left(10^{(30,5905-(8.2\times(Log(\tau+273.16)\div Log(10)))+(0.0024804\times(\tau+273.16))-(3142.31\div(\tau+273.16))-(3142.31\div(\tau+273.6)))}\right)\times(RH\div100)}{((0.0006363601\times(T+273.16))+0.472)\times0.93}.$$

The associated physiological stress is shown in three gradations: the safe zone when the AET is below 40; the alert zone between AET of 40 and 65 and the danger zone with an AET above 65. Figure 20 shows the three zones and their corresponding combinations of T and RH.
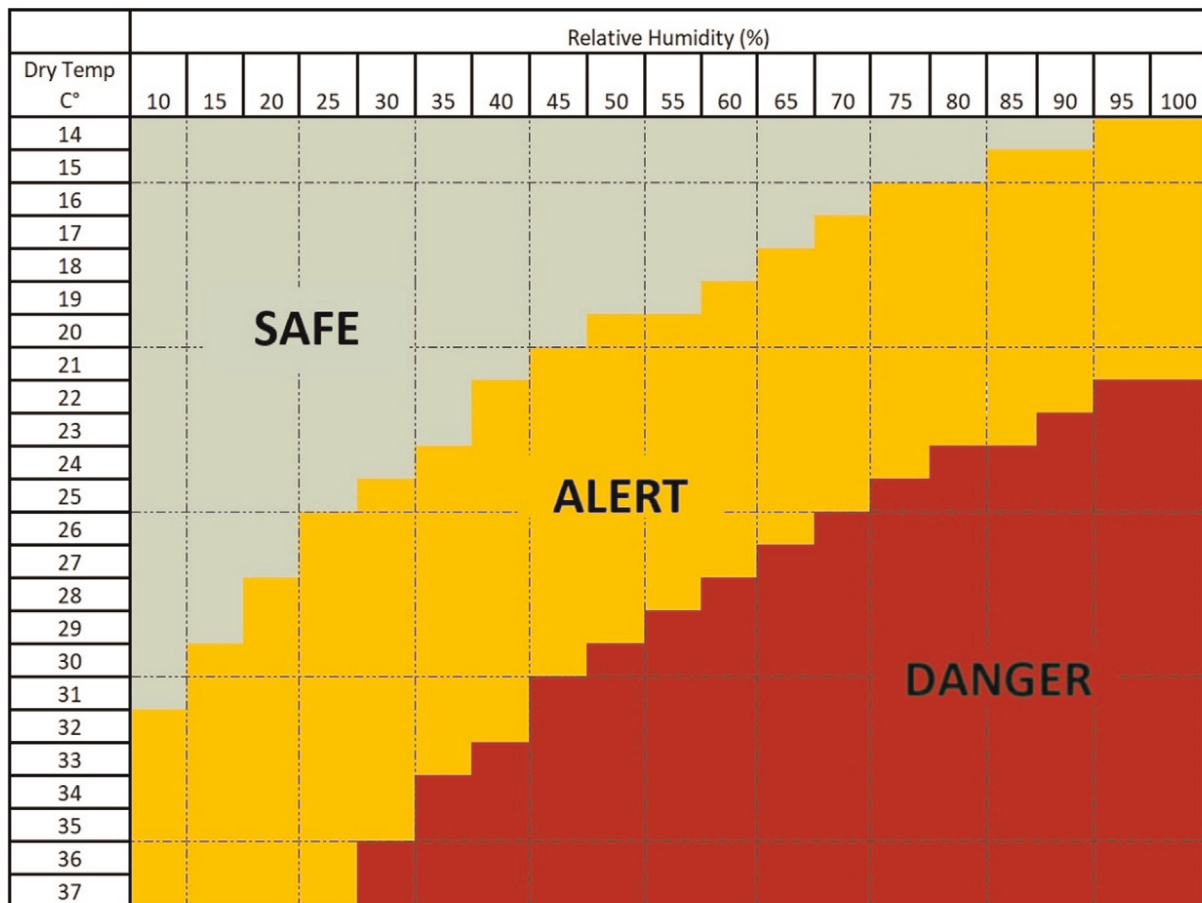
Figure 20: Thermal Comfort Zones for broiler transport defined by apparent equivalent temperature (AET) by Mitchell et al. (2004, 2005, 2006)

The safe zone corresponds to the thermal comfort zone, although it is not exactly the same as Figure 18 since different ranges of humidity are taken into consideration. These ranges of 'Safe' combinations of dry-bulb temperature and relative humidity will impose minimal heat stress under typical broiler transport conditions (i.e. crates or containers and commonly used vehicles).

Combinations falling within the Alert zone (between C and D in Figure 18) will impose some demand for thermoregulatory effort with a small degree of hyperthermia and acid–base disturbances, but thermoregulatory success will be high, with little change in deep body temperature. If they have the possibility, birds will move away from conspecifics and lift their wings, as an attempt to maximise space usage within the crate to ensure that the maximum body surface area is exposed to the environment (Mount, 1979; Weeks et al., 1997). However, in the context of transport, possibilities to do this are limited and the effectiveness of these responses might be impaired by the transport condition (e.g. lack of ventilation and high stocking rate, which reduces body surface area for heat exchange and increases conductive gain from contact with conspecifics). Therefore, the risks of exceeding thresholds of heat stress are increased during transport (Villarroel et al., 2011; Bracke et al., 2020).

In the danger zone (Figure 20). Thermal Comfort Zones for broiler transport defined by apparent equivalent temperature (AET) by Mitchell et al. (2004, 2005, 2006), corresponding to above D, Figure 18), thermoregulatory demands will be high, increasing further with the imposition of greater thermal loads. Thermoregulatory success will be reduced, i.e. deep body temperature will increase, reflecting hyperthermia. Poultry will lose the ability to keep their CBT and the risk of mortality will increase (Mitchell, 2006a,b).

Table 14 shows examples, calculated with the AET equation, indicating that as relative humidity levels rise inside the transport container the corresponding air dry-bulb temperatures marking the upper limit of the safe zone (ALERT) or the upper limit of the alert zone (DANGER) zones will decrease.

Table 14: Examples indicating that as relative humidity levels rise inside the transport container the corresponding air dry-bulb temperatures marking the upper limit of the safe zone (ALERT) or the upper limit of the alert zone (DANGER) zones will decrease. See Figure 20

RH %	Container air dry-bulb temperature (°C) corresponding to AET ALERT	Container air dry-bulb temperature (°C) corresponding to AET DANGER
10	32.5	48
15	30	44
20	28	41
25	26.5	38.5
30	25	36.5
35	23.7	34.5
40	22.5	33
45	21.5	31.5
50	20.5	30.5
55	20	29.5
60	19	28.5
65	18	27.5
70	17.5	26.5
75	16.5	25.5
80	16	25
85	15.5	24
90	15	23.5
95	14.5	22.5
100	14	22

Obviously the longer the journey duration the greater impact a given integrated thermal load will have upon the stress experienced by the birds and therefore their welfare.

The validation and application of AET (Mitchell and Kettlewell, 1998, 2004a,b, 2008; Mitchell, 2009) is specifically for broiler chickens under commercial transport conditions and stocking densities of relevant journey durations. There are good reasons to assume that recommendations for acceptable temperatures and humidity developed from the application of AET may be extended to other poultry categories and species but with appropriate modifications taking account of differences in body weight and surface area, metabolic status and capacity, physiological age and status and insulation (fat and feather cover). The thermal limits for transportation of turkeys have received little research attention. Despite a general belief that the thermoregulatory capacity of turkeys may be greater than that of broilers this may only have relevance if the birds are transported at the same body size. In commercial practice, turkeys are transported to slaughter at much higher body weights and over a greater range of body weights. This increase in absolute total heat and moisture production will impact upon the container thermal micro-environment and may render the birds more likely to experience heat stress at similar ranges of temperature to other poultry. The upper limits for temperature and AET should take these differences into account. There may be also some small difference between broiler breeder thermal requirements and those of slaughter birds despite differences in age of travel and physiological maturity.

Another index is based on enthalpy and is employed as a qualitative indicator of thermal environment of livestock such as poultry, cattle and hogs in tropical regions (Rodrigues et al., 2011). Enthalpy is a physical quantity that has as input variables the dry-bulb temperature, the relative humidity of the air and the local barometric pressure (Campos et al., 2019). Enthalpy has been used by Dos Santos et al. (2017) to assess transport stress related to meat quality traits in broilers. Rodrigues et al. (2011) formulated the Enthalpy Comfort Index (ECI) equation considering an average barometric pressure of 890 mmHg.

$$h = 1.006t + \frac{RH}{pb}10^{(7.5t/237.3 + t)} \times (71.28 + 0.053t),$$

where h = ECI (kJ/kg of dry air); t = dry-bulb temperature (°C); RH = relative humidity (%); pb = Local barometric pressure (mmHg). The ECI was categorised into comfort (35.0–48.0 kJ/kg),

warning (48.1–57.6 kJ/kg), critical (57.7–66.1 kJ/kg) and lethal (66.2–90.6 kJ/kg) zones for broilers from the sixth week of age based on Queiroz et al. (2012). The zones of comfort, warning and critical correspond to the zones of safe, alert and danger derived from AET.

De Oliveira et al. (2021) have employed ECI to classify broiler transport journeys in terms of the threat to bird well-being. The highest ECI observed under tropical conditions (wet season) was 70.6 ± 6.5 kJ/kg and exceeded the broiler comfort zone and was considered within the lethal zone. For the same season, the ECI for other shipments was 58.1 ± 9.7 kJ/kg and was categorised in the critical zone. It was concluded that higher humidity in the environment is harmful to broiler thermal comfort. Values of ECI for other journeys studied was found to be 46.9 ± 4.5 kJ/kg during the dry season and was within the comfort zone limit. A further development of application of enthalpy to the assessment of animal welfare in transit has been presented (Villarroel et al., 2011; Miranda-de la Lama et al., 2021). This work described the use of time derivatives of temperature and enthalpy as non-invasive indicators of welfare of pigs in transit. This analysis has yet to be applied to the transport of poultry.

In conclusion, several indices based on dry-bulb temperature and relative humidity have been developed to measure high effective temperature inside the transport containers. Two of these indices are the apparent equivalent temperature (AET) and enthalpy comfort index (ECI). The AHAW Panel, considering the available evidence and based on their expert opinion, concluded that:

- For AET: The upper limit of the comfort zone (C) is estimated to be below an AET value of below 40. If AET remains below this threshold, broilers and probably other domestic birds are unlikely to experience heat stress during transport (safe zone). The upper limit of the thermoneutral zone (D) is estimated to be at an AET value of 65. Between AET values of 40 and 65, there will be some demand for thermoregulatory effort, but thermoregulatory success will be high with little change in deep body temperature during a limited period of time. Above AET of 65, the bird's mechanisms to cope with heat stress will become less effective and the risk of severe heat stress increases (danger zone). The combinations of relative humidity and dry-bulb temperature giving rise AET values for the safe, alert and danger zones can be found in Table 14 and Figure 20 and above.
- For ECI: The comfort zone is estimated to be below an ECI value of 48.0 kJ/kg. If ECI remains below this threshold, poultry are unlikely to experience heat stress during transport (comfort zone). If ECI exceeds this threshold, there will be an increasing risk of heat stress (warning zone). The thermoneutral zone is estimated to be at an ECI value of 57.6 kJ/kg. Above this threshold, the bird's mechanisms to cope with heat stress will become less effective and the risk of severe heat stress increases (critical zone). ECI can be calculated using the formula reported above.

3.7.7.3. Specific findings on heat stress in end-of-lay hens

Although heat stress is less common than cold stress in end-of-lay hens, it can be sufficiently severe to cause death in hot weather. For example, Petracci et al. (2006) found increased DoA to be associated with higher summer temperatures in Italy. It might be difficult to recommend different limits dependent upon feathering of end-of-lay hens as the feather status can vary from bird to bird in the same consignment and it would be difficult for catchers or others to specifically identify the feather status at depopulation or point of departure. Poor feather cover increases heat loss and may not constitute a problem at higher temperatures and humidities, although hens with high feather loss will have experienced stress during production making them less able to cope with any transport stressor.

Estimates of TNZ for hens on farm and experiments where feed and water are offered do not reflect the conditions during transport but are mentioned here as specific evidence is limited. For laying hens in housing with controlled ventilation, the thermal comfort zone was estimated to be 21–28°C (Castilho et al., 2015) at levels of relative humidity between 40% and 70% (Ferreira, 2016). This closely matches the recommendation of Weeks et al. (1997), who used a heated model hen transported among live hens, that a comfortable range for end-of-lay hens in transit is between 22°C and 28°C when air movement within the containers is between 0.3 and 1.0 m/s. The AHAW Panel cannot recommend ventilation rates as it depends on the design, stocking density and layout of the containers, the weather conditions and the air inlets and outlets of the vehicle.

Recent evidence from Beaulac et al. (2020) used a controlled experiment to simulate transport conditions by exposing fasted hens in crates (with a space allowance of ~ 184 cm²/kg) to various temperature and humidity combinations and durations up to 12 h. They observed panting at 30°C with

significantly more seen both at high humidity (80% RH) compared with drier 30% RH and in well-feathered compared with poorly feathered hens. Negligible levels of panting were seen at or below 21°C. Although no mortality was recorded for exposure to 30°C for up to 12 h, the hens were more restless, and their blood physiology and meat quality parameters indicated significant dehydration.

Weeks et al. (1997) recommended air temperature near poorly feathered hens should be in the range 22–28°C to achieve thermal comfort, but they did not measure humidity levels. There are no comparable figures for well-feathered hens, but expert opinion suggested a range from 22–25°C is likely to represent their TNZ during transport. Ribeiro et al. (2020) experimentally exposed individual laying hens on full feed and water to several wind speed, humidity and temperature combinations. Results confirmed that hens kept at 24°C and 60% RH or at 28°C and 40% RH at wind speeds between 0.2 and 1.4 m/s were within their TNZ. The recommended upper thermoneutral limit for all end-of-lay hens is suggested to be 28°C and 60%. The interpolation of this limit into the AET table or graph indicates that this would be classified as falling within the alert zone for broilers.

In conclusion, in the absence of direct validation of the AET through experimental studies on end-of-lay hens it is reasonable to extrapolate from broilers to end-of-lay hens and similar ranges of temperature and humidity of those recommended for domestic birds can be suggested for end-of-lay hens.

3.7.7.4. ABMs

Panting is a feasible ABM that is considered feasible for assessing heat stress in birds during transport (Table 15).

Table 15: ABMs for heat stress in domestic birds

ABM	Definition and interpretation of the ABM
Panting	**Definition**: Breathing with short, quick breaths with an open beak (Welfare Quality®, 2009) (from EFSA AHAW Panel, 2019).
	Measurement: Observation of the proportion of birds showing this behaviour in a representative sample of birds. It is recommended that 20 crates containing broilers should be observed at various locations in the vehicle (front, middle, rear) or in lairage. The number of birds that pant can be quantified according to the following formula: % of birds panting = (number of birds panting)/ (number of birds per container × number of containers monitored)) × 100% (Butterworth, 2009). A percentage of panting exceeding 1–6% (Berghout et al., 2018 for broilers) is indicative of heat stress.
	Visual inspection is feasible at unloading or in lairage. Containers on the edge of load can be inspected also at loading and during stops.
	Sensitivity is high, as birds will pant when exposed to high effective temperatures and panting occurs only during severe heat stress in broilers (de Jong et al., 2012) and in end-of-lay hens (Ribeiro et al., 2020).
	Specificity is high, as the absence of severe heat stress is associated with normal respiration rates (absence of panting).

3.7.7.5. Hazards

Too high effective temperature

Too high effective temperature is the principal hazard for heat stress. As mentioned above, between AET values of 40 and 65, there will be some demand for thermoregulatory effort, but thermoregulatory success will be high with little change in deep body temperature during a limited period of time (alert zone) and above AET of 65, the bird's mechanisms to cope with heat stress will become less effective and the risk of heat stress increases (danger zone). The ventilation in the vehicle (particularly when stationary) is a highly influencing factor on temperature during transport.

When in motion, air tends to move from the back to the front of the vehicle. The general rule is that the risk of heat stress will be highest close to the final air exit points in all types of vehicle (Figure 19). When stationary, heat and moisture will tend to accumulate in the entire load and problems in any of the hotspots illustrated in Figure 19 may be exacerbated depending on the duration of the standstill. On warm humid days, the heat and moisture load imposed in the thermal core of the poultry vehicle or trailer will be greater than on an average or cool day. Using curtains under these conditions would most likely exacerbate the problem.

In hot and humid environmental conditions, poor ventilation will exacerbate the perceived effective temperature. Ventilation plays a key role in the dissipation of heat and humidity as produced by

animals during transport (Schrama et al., 1996; Consortium of the Animal Transport Guides Project, 2017). A higher level of humidity in the air will worsen the effect of high temperatures. A higher air speed (in practice: active or fan ventilation) will reduce this effect. Ritz et al. (2005) reported an increased level of mortality in broilers during loading, unloading and lairage due to the reduced ventilation, but not during the journey where vehicle motion provided sufficient air movement from passive ventilation.

Specific findings for hazards in end-of-lay hens

For end-of-lay hens, Richards et al. (2012) modelled data from loggers in eight positions within loads of end-of-lay hens transported in modules, and confirmed that both when travelling and in lairage, some parts of the load tracked outside air temperatures whereas others were dominated by bird heat owing to uneven ventilation. Conditions also varied within modules, with the upper and central drawers (unsurprisingly) being warmer.

Figure 21 from Richards et al. (2012) illustrates the temperature variation within one drawer of hens in a module placed at the top, front of the load indicating likely heat stress for short periods during loading when temperatures exceeded 30°C, but overall, the microclimatic temperature was mainly within the estimated TNZ. This was for a commercial transportation under temperate conditions in the UK. With hens in containers at high stocking density and, in most of the load, very little air movement, levels of humidity are likely to be high, limiting the effectiveness of evaporative cooling (panting).
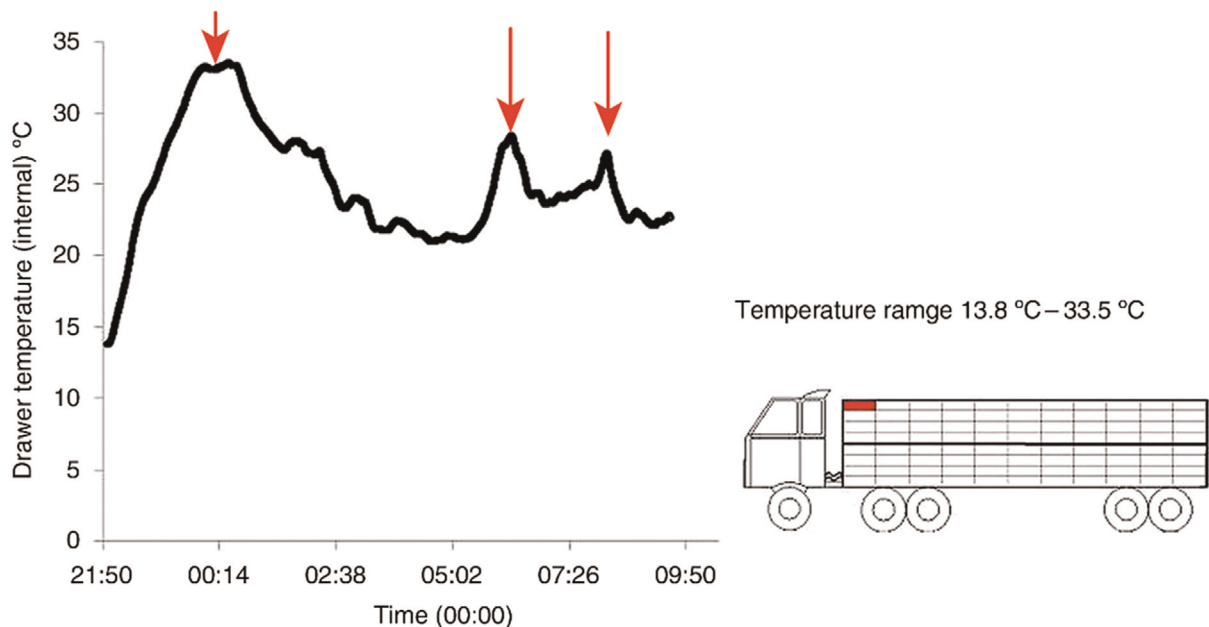
Figure 21: Thermal profile from internal logger position b (top, front). The first arrow indicates the beginning of transport, the second at unloading and the third the middle of the lairage period (From Richards et al., 2012)

Insufficient space allowance

Space allowances have important effects on heat exchange. Insufficient space allowance is detrimental in hot weather. A primary cause of the problem is the increased heat and moisture production within the container or the load/vehicle resulting from a greater biomass in the load. In addition, limited space within the transport container limits the expression of behavioural thermoregulation and exploitation of changes in posture and orientation to enhance heat loss e.g. raising wings and generally uncovering thermal exchange windows of less feathered skin. The close proximity of other birds further limits heat loss as direct contact with other birds reduces the effective surface area of each individual available for heat loss. The mass of birds also presents an effective obstruction to air flow through the perforated container sides, base and top thus reducing both ventilation for the removal of heat and moisture and direct convective cooling.

3.7.7.6. Preventive measures

Preventive measures for too high effective temperature

To prevent domestic birds to experience heat stress, they should travel in the safe zone, corresponding to AET values below 40. Birds should not travel in the danger zone. Related to the possibility of travelling in the alert zone, Mitchell and Kettlewell (2004a,b) studied combinations of T and RH falling in the alert zone and found they resulted in minimal or moderate physiological response reaction to heat stress in birds in transport simulations of 3 h duration. For this reason, based on their expert opinion, the experts of the WG suggest that birds could be transported when in the alert zone for journey durations up to 4 h (which is the typical duration of journey for domestic birds and does not differ too much from the 3 h transport simulations).

The combinations of relative humidity and dry-bulb temperature giving rise to AET values for the safe, alert and danger zones were calculated and can be found in Figure 20 and Table 14.

To monitor AET, vehicles should be equipped with sensors recording dry-bulb temperature and relative humidity inside or close to the containers at several locations to include the top 'hot spot' at the front, the 'cold spot' at the outside rear and bottom of the load and representative points in between. The driver should monitor the microclimate (combination of T and RH) of the load and adjust the ventilation if the AET exceeds the comfort zone levels.

Convective heat loss (including increased air movement within the containers) is the most effective means of managing heat stress for animals in transit. At too high effective temperatures increased ventilation of the load is required to remove excess heat and moisture production from the birds (thus reducing the effective thermal load with particular attention to regulating the in-crate humidity or water vapour density). In passive ventilated vehicles, the removal of the curtains will increase ventilation and may allow the birds to stay in their thermoneutral zone when the vehicle is in transit but not when it is stationary. There is scope also to improve thermal comfort by modifying vehicle design and operation by incorporating mesh panels within the curtains and fitting adjustable vents to both the headboard and tailboard (Figure 22). Locating vents with sliding covers in the headboards and tailgate, will enable air flow to be adjusted according to weather and bird conditions.

Figure 22: Mesh panels within the curtains of a vehicle and adjustable vents to both the headboard and tailboard (Courtesy of Malcolm Mitchell)

The structure and permeability of curtains may be altered and/or the trailer structure may be modified to enhance the efficiency of air entry or exit driven by the pressure profiles. Thus, replacing solid curtains with curtains containing permeable sections will facilitate air movement. The use of permeable mesh sections allows air exchange but will exclude or limit water ingress. Permeable curtains may be used on the sides of the vehicle and/or at the rear. Various designs and configurations are in current commercial use e.g. lateral permeable strip sections running the full length of the vehicle.

Ventilation may be further modified by optimisation of air extraction towards the front of passively ventilated vehicles by the creation of an 'effective plenum chamber' behind the front headboard. The sides of this plenum are left open and correspond to the position of the low-pressure region on the

moving vehicle. Enhanced and preferential air extraction takes place through this defined outlet. The coupled use of this strategy with perforated or permeable curtains greatly improves and controls the passive ventilation of the load. A number of vehicle or trailer designs using these approaches are currently in commercial use. To aid passive ventilation, vehicles should have solid roofs, which can be raised when stationary in hot weather, including during loading except in the case of containers fixed to the trailer with a central longitudinal passage (also called liners). In this case, the roof should be open above this passage.

Other preventive measures can be put in place when planning transportation in vehicles with passive ventilation. Hottest hours of the day should be avoided (Warriss et al., 2005). Reducing the number of birds per crate or drawer will reduce heat production and improve air flow around the birds. When animals are transported in the alert zone (i.e. at AET levels between 40 and 65) leaving parts of containers or whole modules empty especially at the hot spots will reduce the risk of birds being exposed to AET levels corresponding to the danger zone (AET values above 65).

Nevertheless, the modular and container structure of vehicles for broiler transport and for other categories of poultry makes it difficult to achieve good ventilation of all birds in the bio-load using only passive ventilation systems.

Air circulation and ventilation rate are better controlled in fan ventilated vehicles, where the movement of air into, within and out of the vehicle container is controlled by a combination of suitably positioned mechanical fans of sufficient capacity, and natural apertures which should enhance established natural air pressure gradients. A major advantage of mechanical ventilation is that air flow may be adjusted in response to external conditions, number of birds transported and their physiological requirements. These vehicles have the major advantage that the ventilation is not dependent upon vehicle movement and improve the distribution of airflow in the actual load thus reducing the risk of thermal stress. Therefore, to achieve thermal comfort for all birds in transit, controlled and uniform mechanical ventilation is essential (Weeks et al., 2019). Mechanically ventilated vehicles and trailers have been designed and are successfully operated in other sectors of livestock transportation (e.g. cattle, pigs and sheep). Similar mechanically ventilated vehicles for poultry transport have been proposed and developed but as yet only few vehicles have been employed in commercial practice.

Controlled environment or air-conditioned vehicles or trailers can regulate or modify internal thermal conditions by appropriate heating or cooling. These are typically used for the transport of newly hatched or day-old chicks. They have the major advantage that the internal environment may be controlled precisely regardless of external weather or thermal conditions and does not rely upon vehicle movement. Controlled environment vehicles are not common in the EU for the transport of poultry for slaughter but should be employed as designs are available (Mitchell and Kettlewell, 1998, 2001, 2008).

New designs of module and crates have been developed by commercial companies to increase load ventilation and specifically to increase air movement in and around the crates and birds. Figure 23 shows mechanically ventilated vehicles.

Figure 23: Mechanically ventilated vehicle that uses curtains and a fan in the tailboard for ventilation (Courtesy of Malcolm Mitchell)

<u>Specificity for end-of-lay hens</u>

Feathering becomes very important when birds are exposed to lower temperatures, particularly if there is significant air movement and/or a risk of wetting. The lower limit for temperature for end-of-lay hens might be higher than for other categories of poultry as generally they have a poorer feather cover and limited metabolic capacity and reserves. As it might be difficult to recommend different limits dependent upon feathering of end-of-lay hens, it may be proposed that in crate temperatures for end-of-lay hens should be maintained above 18°C regardless of feather cover on the assumption that if this is higher than the limit necessary for thermoregulatory requirements it cannot impact the welfare of the birds (i.e. it will not cause heat stress) but will protect end-of-lay hens whatever their feather status.

Furthermore, wetting and excessive air flow should be absolutely minimised under cold conditions. It is recommended that any wetting of crates and drawers is avoided (e.g. washing of containers with inadequate drying or rain ingress during the journey to the farm). As the risk is greatest at the air inlet it may be necessary to avoid loading of these modules and crates with end-of-lay hens (leave empty).

Preventive measures for insufficient space allowance

Increased space may facilitate penetration of air movement into and between the containers and thus enhance convective cooling from the surface of the birds and evaporative cooling of the exchange surfaces including the respiratory tract. Common practice from industry is to increase space allowance by 10–25%. Another strategy is to leave some (upper/central) containers empty within the load to obtain a similarly beneficial effect. As these strategies are already employed by the industry in hot weather, the determination of the numbers of birds to be loaded per container and/or the numbers and locations of containers to be left empty had been determined by trial and error and it is not possible currently to make precise recommendations for quantitative strategies as no published research data are available.

To enable bird comfort and sufficient air circulation, there should be a gap above their backs at least equivalent to the distance from the back to the top of the head in a normal sitting posture. Moreover, to achieve sufficient ventilation within the load there ought to be gaps between the containers as, for poultry, there is evidence to suggest the height allowance within the containers should be sufficient for seated rather than standing postures. This is a compromise, weighing the disadvantages of an increased risk of incurring trauma from injuries with provision of greater space allowance – vs. an increased risk of inadequate ventilation (and heat stress during warm, humid weather) with reduced space allowance, which will also prevent birds from performing their full behavioural repertoire and, we believe, lead to some degree of negative affective states such as frustration and fear.

Pinheiro et al. (2020) concluded that the use of spacers between the transport crates can modify the ventilation patterns within the load and increase the air circulation between the crates. However, the internal ventilation within the crates was not demonstrably changed.

It is to be noted that due to the obstructions presented by the modular structures, containers (crates) and the bird themselves, it is not possible to indicate minimum air movement within the container.

3.7.7.7. Corrective and mitigative measures

In certain cases, misting and nebulisation can be applied to the birds to cool them down (Jiang et al., 2015), assuring appropriate ventilation. Partial surface wetting has been reported useful as a method of reducing heat stress, particularly when RH is low, because water evaporates by absorbing heat directly from the body of the bird, and also by absorbing heat from the surrounding air (Mutaf et al., 2008). Some studies (e.g. Douglas et al., 2019) indicate this is effective, whereas others do not (Luthra, 2017; Pinheiro et al., 2020).

If there are delays, vehicles should keep moving (go out on the road again) until birds can be unloaded into a lairage with appropriate environmental conditions as detailed in the EFSA Opinion on Slaughter of poultry (EFSA AHAW Panel, 2019). In order to reduce the risk of thermal stress all slaughterhouses should be equipped with holding facilities with banks of fans to ventilate the stationary vehicle or trailers prior to lairage or unloading. The provision of roofs and some wall structure will provide shade, will reduce direct solar radiation on the vehicles and will provide a better control of the direction of the forced convective flow. During loading and at arrival, when the vehicle is stationary, birds should be protected from the sun and ventilation provided. Vehicles should be parked

in the shade and the roof be raised during hot weather when loading alternate stacks or modules onto the vehicle can maintain good airflow around the birds. Containers should be spaced out and fans directed at the birds if panting is observed.

During loading, to mitigate heat stress the vehicle should be loaded from back to front and preferably completing the whole lower layer first (where 2-tier module systems are used). This is because heat rises and accumulates behind the front headboard so loading potential hot-spots last delays the potential onset of heat stress during the journey. Where fans are fitted to the vehicle, turn them on during loading and if birds are observed to be panting direct free-standing fans at the loaded birds.

Animal welfare assessment on vehicles during transport and at arrival in slaughterhouses should be directed close to the final air exit points, to the top row of drawers (centre) of the top modules, that is the location with higher risk of heat stress (Figure 19). The frequency of such assessments should increase at times of high ambient temperature and corrective measures must be put in place if birds show signs of heat stress.

3.7.8. Cold stress

3.7.8.1. Description

The animal experiences stress and/or negative affective states such as discomfort and/or distress when exposed to low effective e temperature.

The LCT (A in Figure 18) is the point in environmental temperature below which an animal experiences cold stress and must increase its rate of metabolic heat production to meet the heat loss to the environment. When environmental temperature drops below the LCT, the body must produce heat by increasing its metabolism (e.g. by shivering) in order to maintain its core body temperature. At the point where the heat-producing capacity is exceeded by heat loss, the core body temperature will begin to drop, eventually reaching fatally low levels. However, below the limit of the comfort zone (point B in Figure 18), animals are already at risk of cold stress.

Heat exchange via radiation can be a major contributor to cold stress. In very cold, clear weather significant heat can radiate to the sky. In cold conditions air movement will increase convective heat loss (and surface evaporative cooling if the birds are wetted) and this will be detrimental to the cold stress of the birds.

To keep warm, birds rely on heat generated from metabolism and from muscular activity. In the transport situation when feed is withdrawn, and birds are confined in containers their ability to generate heat is reduced and their LCT may be lower (Koh et al., 2000).

When the effective temperature is too low, the thermoregulatory capacity of the birds for homoeothermy is exceeded. Birds can die from hypothermia if the conditions are too cold or the birds are wet and cold (Caffrey et al., 2017). Hunter et al. (1999a,b) and Mitchell et al. (2001) and Mitchell (2006a,b) reported studies in which a model was developed which related physiological responses, in broilers in transport simulations, to low temperatures with superimposed air movement (0.7 ms^{-1}) and wetting during simulated journeys of 3-h duration. This model indicated that wetted birds experience moderate cold stress at air temperatures below 8°C and predicts that at −6°C or lower wetted birds in an air stream would experience severe and potentially fatal hypothermia. Even at relatively mild temperatures (e.g. 4°C and 8°C) wetting has a significant impact on feather insulation and evaporative cooling and this combination of conditions will induce time dependent hypothermia and increase the demand for thermoregulatory responses. Even at an air temperature of 12°C wetting and air movement were associated with hypothermia although only a small decrease in body temperature occurred when birds were kept dry.

In conclusion, cold stress is mainly induced by low dry-bulb temperature, although it is also influenced by air movement. Humidity does not have a big impact on thermoregulation demands for cold stress. Therefore, conclusions for cold stress are based on dry-bulb temperature (and are not based on indexes combining temperature and humidity).

The AHAW Panel, considering the available evidence and based on their expert opinion, concluded that the lower limit of the comfort zone is estimated to be at 10°C which is an average considering the above evidence for which 12°C did not induce significant cold stress responses, and 8°C did induce some moderate responses. If the effective temperature in the containers remains above 10°C, domestic birds are unlikely to experience cold stress during transport. Below 10°C, the bird's mechanisms to cope with cold stress will become less effective and the birds will likely experience cold stress.

3.7.8.2. Specific findings on cold stress end-of-lay hens

End-of-lay hens are at greater risk of experiencing cold stress than heat stress during the whole transportation process (loading, journey, lairage and unloading). They are often poorly feathered owing to wear and tear feather loss in cage systems and to injurious pecking in all systems (Knowles and Broom, 1990; Richards et al., 2012). The lack of insulation provided by feathers (including the opportunity to fluff the feathers) makes them more sensitive to cold (Leeson and Morrison, 1978). Furthermore, they are metabolically exhausted at the end of the egg laying production cycle with low energy, protein and calcium stores, and are therefore more prone to fatigue and bone breakage (Gregory and Wilkins, 1989; Gregory and Devine, 1999; Whitehead and Fleming, 2000; Richards et al., 2012) and lack reserves to draw on to keep warm by shivering. Their body weight is frequently low with minimal fat cover and musculature. In a UK study of 24 commercial flock depopulations, Weeks et al. (2012) found that poor feather cover, poor health, lower body weight and cumulative mortality of the flock was positively associated with an increased risk of DoA. Many of these factors would make the birds more susceptible to cold stress.

Cold stress is a likely contributing factor to the significantly higher DoA rate during winter and spring than during autumn and summer as reported by Çavuşoğlu and Petek (2021) for 31.7 million hens transported to one Turkish slaughter plant. Further, they found 4.02% weight loss in winter, significantly more than during autumn (3.46%) which could be due to shivering to try to maintain body temperature.

A large survey of the majority of end-of-lay hens slaughtered in the UK during 2009 found lower ambient air temperatures (in the range daily mean $-2.2°C$ to $23.1°C$) to be a risk factor for increased DoA (Richards et al., 2012). Similarly, in the Czech Republic lower ambient temperatures were associated with increased DoA for over 3,000 consignments totalling over 17.4 million birds between 2010 and 2017 (Večerková et al., 2019) with greater mortality during the cold winter months of January (0.72) and December (0.695%) compared with August (0.36%). They showed that below $15°C$, there is a significant increase in DoA, as well as below $0°C$.

Under commercial transport conditions, birds may be exposed to both wind and wetting at all stages of transport both of which have a profound chilling effect mimicking lower temperatures even when ambient temperature is well above zero.

The lower limit for temperature for end-of-lay hens might be higher than for other categories of poultry as generally poorer feather cover and limited metabolic capacity and reserves. As it might be difficult to recommend different limits dependent upon feathering of end-of-lay hens, the AHAW Panel proposed that in crate temperatures for end-of-lay hens should be maintained above $18°C$ regardless of feather cover on the assumption that if this is higher than the limit necessary for thermoregulatory requirements it cannot impact the welfare of the birds (i.e. it will not cause heat stress).

3.7.8.3. ABMs

ABMs that are considered feasible for assessing cold stress in birds during transport are given in Table 16.

Table 16: ABMs for cold stress in domestic birds

ABM (animal categories)	Definition and interpretation of the ABM
Huddling	**Definition**: Sitting close together in tight groups or clumps often with open space in between (Welfare Quality®, 2009) (from EFSA AHAW Panel, 2019) in order to share the body warmth of conspecifics.
	Measurement: Observation of the proportion of chickens showing huddling behaviour (Welfare Quality®, 2009). In the future, it might be possible to fix cameras to monitor positioning within the crate as birds could move away from inspectors and appear to be huddling, thus the behaviour should be distinguished from fear reactions. Continuous video recording could be observed by the driver and transmitted remotely.
	Sensitivity is high as chickens will clump together as a response to low environmental temperatures. Persistent huddling indicates cold stress.
	Specificity is high. Birds may also huddle in response to other welfare consequences such as fear, but these groups are looser with empty spaces in between.

ABM (animal categories)	Definition and interpretation of the ABM
Fluffing up of feathers	**Definition:** Erection or ruffling or bristling of feathers (Strawford et al., 2011) (from EFSA AHAW Panel, 2019). This is an autonomic response to trap air within the feathers to increase their insulative value.
	Measurement: The feathers of the birds are ruffled giving a rounded, fluffy appearance.
	Sensitivity is high, as piloerection always occurs in severe cold stress (autonomic response). Sick birds may also exhibit the behaviour, but this is in response to their elevated CBT and reduced feed intake, which makes them more sensitive to cold stress.
	Specificity is high, as thermally comfortable birds (in the TCZ) do not show piloerection
Shivering	**Definition:** Shaking slightly and uncontrollably (Strawford et al., 2011) (from EFSA AHAW Panel, 2019) as a response to generate additional body heat from the muscle reserves to preserve CBT The wings or body of the hen quiver or move from side to side in a rapid motion coupled with fluffed feathers (Beaulac et al., 2020).
	Measurement: Observation of the proportion of birds showing this behaviour in a representative sample of birds, especially checking containers located at the bottom and on the side of load. Manual inspection of these sections is feasible at loading, unloading and during stops.
	Sensitivity is high as shivering always indicates cold stress.
	Specificity is moderate as a lack of observed shivering does not indicate that cold stress is absent as individual birds vary in their capacity to use shivering for thermoregulation.
Cloacal temperature	**Definition:** Core body temperature measured by in the cloaca of the chicken (Mujahid and Furuse, 2009).
	Measurement: Measuring cloaca temperature using a thermometer in a representative sample of chickens. This can only be measured after arrival, during uncrating.
	Sensitivity is high. A cloacal temperature (measured by cloacal temperature) lower than 40°C indicates that chickens are too cold (Mujahid and Furuse, 2009; Maman et al., 2019).
	Specificity is high. When chickens are not too cold, they do not have a cloacal temperature below 40°C.

Among these ABMs, huddling, fluffing up of feathers, shivering and cloacal temperature are considered by the experts to be sensitive and specific for the assessment of cold stress.

3.7.8.4. Hazards

Too low effective temperature

The effective temperature perceived by an animal is a combination of the temperature, the humidity and the ventilation or wind speed. In the case of low effective temperature, humidity is not so relevant as for high effective temperature. The risk of cold stress during commercial broiler transport is limited and is the highest in extremely low ambient temperatures or at higher temperatures but in conditions where air movement and water can penetrate the load. The general rule is that the risk of cold stress will be the highest close to air entry points in all types of vehicles (Figure 19).

The heat production of broilers within the load allows warming to acceptable temperatures even at sub-zero external temperatures. Under such conditions the vehicle and load ventilation is designed to ensure the removal of additional or excess heat and water vapour and to deliver adequate fresh air to the birds within the load. Ventilation is concerned with the proper management of the heat and moisture loads generated during transport of birds (and other animals) in containers. As adequate ventilation requires specific air inlets and outlets in passive systems (open load or improved designs with defined apertures) or mechanical systems. There is a risk of cold stress for birds located at cold

air inlets if the external temperatures are very low despite the thermal micro-environment being adequately managed in all other parts of the load.

Inspection of the scientific literature reveals that a major issue for birds in transit exposed to low ambient temperatures is the risk of wetting (rain/snow ingress, melted ice, road spray at air inlets, wetting during catching and loading). Loading into wet containers in cold weather may cause initial chilling of the animals. During transport in cold and humid environmental conditions, lack of curtains to protect the birds and lack of heating system in the vehicle might result in rapid decrease of the perceived temperature. On the majority of passively ventilated vehicles, the air flow when moving tends to be from back to front due to the pressure profile when curtains or sides are closed. Adequate ventilation, regardless of ambient temperature, means that even with minimal ventilation vents minimised, there is still a risk of water entry from both precipitation and road spray. Such wetting is accompanied by increased convective cooling in the load region close to the air inlet. The combined effects of disruption of the feather insulation by wetting, enhanced convective cooling by air movement and increased evaporative cooling from the wetted surface. These thermal challenges combined with lower air temperatures greatly increase the risk of cold stress.

3.7.8.5. Preventive measures

Preventive and corrective measures of too low effective temperature

On all vehicles broilers might be considered safe from cold stress if the lowest temperature in the containers is 10°C or more.

Under cold climatic conditions, the vehicles should park in a sheltered position during loading and unloading. Lorries should be parked in warm areas and avoid wind in cool or cold weather. Vehicles and/or modules should have curtains, which can be used according to weather conditions. The design may need to vary according to both weather and class of bird transported and may need a combination of mesh and solid panels. The use of curtains would be merited in order to prevent the birds freezing at the rear of the vehicle. However, one would also have to consider the possibility of overheating the birds in the zone where they could become potentially very warm at the front and top of vehicles. Some vehicles in colder countries may have solid sides with defined inlet and outlet apertures which may function in a similar manner to modified curtained vehicles but give more thermal protection during cold weather.

Furthermore, the lower temperature limit can be maintained or exceeded by management of the ventilation rate and air distribution by adjusting vents and inlet outlet area ratios. At low external temperature the heat and moisture production of the birds within the load will allow elevation of in-crate temperature by reducing crate ventilation rate and mixing rate of on-board air with incoming cold air from outside the vehicle. Careful attention should be paid to simultaneous control of water vapour density and avoidance of unnecessary high air velocities at the air inlets. In addition, short term strategies can include not loading modules or crates located immediately adjacent to the air inlets.

Mechanically ventilated vehicles are unlikely to be associated with the risk of cold stress, as lower temperature limit can be maintained by reducing fan throughputs. Ventilation systems ensure low speeds of incoming air and a sufficient degree of diffusion and mixing with the internal air. This will avoid the risk of cold stress for birds located at cold air inlets if the external temperatures are very low.

Any wetting of crates and drawers should be avoided (e.g. washing of containers with inadequate drying or rain ingress during the journey to the farm. As the risk is the highest at the air inlet it may be necessary to avoid loading of these modules and crates with birds (leave empty).

Confined in transport containers at high stocking densities, birds' bodies are so close that they are in effect one large mass with mainly their backs and heads able to lose heat by convection and evaporation. Thus, comparatively high stocking rates reduce the risk of cold stress. It has also been shown that when broilers were exposed to cold temperatures, huddling behaviour increased (Strawford et al., 2011).

Avoiding the coldest hours of the day for transportation of animals will allow them to avoid extreme climatic conditions, which are especially problematic when vehicles are not equipped with curtains (European Commission, 2017b).

3.7.8.6. Corrective and mitigative measures

Cold stress can be mitigated by adjusting ventilation or closing curtains. Upon arrival at the abattoir lairage waiting times should be kept to a minimum and loads with more vulnerable birds (from longer

distances, less healthy or poorly feathered flocks) should be unloaded and processed without delay. Birds should be observed for signs or cold stress (huddling, shivering or cold to the touch) and the load managed accordingly with attention to climatic conditions.

3.7.9. Prolonged hunger

3.7.9.1. Description

Freedom from hunger and thirst is the first of the Five Freedoms (FAWC, 2019) that have been widely adopted as guiding principles of animal welfare. Birds transported in containers are usually deprived of feed for a period before being caught and crated. Due to this routine practice prolonged hunger may affect all birds, and prevalence is regarded as high. The duration of prolonged hunger will depend on onset and journey time. Once hunger starts, it will continue with increased severity until the bird is slaughtered or unloaded at the production site and provided with feed. The onset of experiencing hunger, meaning a craving or urgent need for food accompanied by an uneasy sensation (a negative affective state), is not considered a welfare concern. However, depending on duration of feed withdrawal, and also category of bird, body condition, temperature and additional stressors during transport, fasting will eventually lead to the welfare consequence of prolonged hunger as metabolic requirements are not met. Birds which have had free access to feed on farm and with high metabolism (broilers and turkeys) or depleted body reserves (end-of-lay hens) are at particular risk of experiencing prolonged hunger. On the contrary, meal-fed birds (e.g. pullets, free-range laying hens, game birds) may have developed larger crops, which can act as a feed store, delaying the onset of hunger. When the digestive tract is empty and/or physiological or behavioural ABMs of hunger are present, birds will feel hungry and be motivated to eat.

Feed deprivation is done for hygiene, practical and economic reasons, with no proven welfare benefit to domestic birds. Nevertheless, when containers have perforated floors, lowering the level of droppings may reduce feather soiling, which could benefit their welfare.

For birds transported to the slaughterhouse, feed withdrawal is practised at farm level, before catching, to allow time for the digestive system to empty before processing, leaving less ingesta and faeces for potential carcass contamination (Caffrey et al., 2017). Warriss et al. (2004) noted that the maximal clearance of the crop occurs within 4 h of withdrawal of both feed and water, or earlier (within 2 h) where only feed is removed and that the rate of defecation initially reduced but after 4 h of feed deprivation remained the same. Birds require water in order to digest feed and their drinking behaviour does not decrease until up to 4 h after feed withdrawal (Warriss et al., 2004) (see Section 3.7.10 on prolonged thirst).

Most of the literature on effects of feed withdrawal before transport and slaughter concerns broilers. Broilers are highly motivated to feed spending about 5.4% of their time eating and with free access to feed they eat frequently, typically 53 feeding bouts in 24 h (Weeks et al., 2000) although Warriss et al. (2004), reported 26 bouts in 24 h in a smaller study. Feed withdrawal therefore thwarts their normal behaviour and their onset of experiencing hunger as a negative affective state is likely to develop after a short period of feed deprivation.

The rate of feed passage through the gastrointestinal tract is fairly rapid and is similar in chickens and turkeys (Hillerman et al., 1953), where a significant reduction in gut contents occurs within 4–6 h of feed deprivation (Warriss et al., 2004; Caffrey et al., 2017). Weight loss in the first 3–4 h is due mainly to the elimination of excreta and corresponds mostly to undigested feed. With the advance of fasting, body weight loss corresponds mainly to the utilisation of body reserves.

Feed withdrawal affects several metabolic processes as it quickly exhausts the main energy supplies necessary for the broilers to cope with the conditions to which they are subjected. Glycogen reserves are very small in broilers. Deprivation of feed for 5 h resulted in depletion of glycogen stores in the liver (Savenije et al., 2002), which is the primary store available for maintaining blood glucose levels. Warriss et al. (1988) found also depletion of liver glycogen to negligible levels within 6 h, and this may result in the perception of hunger (Knowles et al., 1995). Subsequent to utilisation of glycogen stores, feed withdrawal causes a shift from anabolism to catabolism and a reduced metabolic rate (Buyse et al., 2002). The largest mobilisation of nutrients to meet the maintenance energy requirements results from lipolysis and proteolysis. These findings indicate that after 6 h, metabolic requirements are no longer met and so the welfare consequence of prolonged hunger starts.

Faeces are composed of undigested feed and endogenous components that are constantly secreted regardless of feed intake, such as gastric, pancreatic, hepatic and intestinal mucous secretions, as well

as mucosal sloughing from different digestive segments. In addition, faeces are always eliminated together with urine especially in birds with access to water.

A close relationship exists between the period of fasting and the dry matter within the gastrointestinal tract. During prolonged fasting, the proportion of faeces not originating from the feed increases, altering the chemical composition and physical characteristic of the excreta. Therefore, increased contamination occurs with excreta originating from long fasting periods due to their higher viscosity. In chickens, extremely watery excreta have been found with withdrawal periods longer than 12 h (Chen and Moran Jr, 1995), probably from an accentuation of intestinal cell breakdown (Gomes et al., 2008). This condition is likely to affect the health and welfare of the broilers.

Gomes et al. (2008) reported a linear body weight loss during a 12-h feed withdrawal period of 0.20% and 0.36%, for broilers with and without access to water, respectively. This highlights the importance of dehydration for total weight loss. More recent work on broilers transported in Belgium (Jacobs et al., 2017a,b) reached similar results.

From the available evidence that broilers subjected to feed withdrawal periods longer than 6 h will exhaust liver glycogen reserves, the AHAW Panel concluded with 50–100% certainty (more likely than not) that they will experience prolonged hunger. Furthermore, the AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) that domestic broilers subject to feed withdrawal periods longer than 12 h will experience prolonged hunger as well as intestinal cell breakdown which is detrimental to their health and welfare.

In turkeys, the rate of passage of feed through the gastrointestinal tract is fairly rapid and is similar to broiler chickens. About half of the total content of a meal is excreted within 4–5 h (Imabayashi et al., 1956) and about 75% of it is excreted within 12 h (Duke, 1986).

Generally, research indicates that for hygiene and economic reasons, the optimal total feed withdrawal time, is between 8 and 12 h prior to slaughter, as this withdrawal period yields the lowest occurrence of carcass contamination and carcass yield losses (Wabeck, 1972). Less than 8 h may result in faecal residues in the digestive tract. After 12 h of feed withdrawal, the intestine is almost entirely rounded as it fills with gas and gas may bubble out if the intestine is nicked. The entire gut lining is broken down and sloughed lining is found in much of the intestine (Savage, 1998). Although intestinal sloughing seems to be more marked in broilers compared to turkeys, and intestinal strength will begin to decrease after 12 h in both species when intestinal 'suffering' appears to be evident. The AHAW Panel considers this as sign of prolonged hunger that is extremely detrimental to animal welfare.

As for broilers also for turkeys, serum triglyceride concentrations are responsive to feed and water withdrawal. In a study by Huff et al. (1996), serum triglyceride concentrations were reduced from control levels within 16 h of feed deprivation in market age heavy strain turkeys. In another study (Bacon et al., 1988), 12 h feed withdrawal was sufficient to cause a significant reduction in blood plasma concentrations of triglycerides. Serum triglyceride concentrations decreased rapidly also in turkeys that were full fed and deprived of water. This may be an indirect effect of the concomitant decrease in feed consumption that occurs in water-deprived turkeys, which can be as much as 79% (Proudman and Opel, 1981; Augustine, 1982; Leeson et al., 1988).

Considering the available evidence, the AHAW Panel, based on expert opinion, concluded that similar conclusions to broilers are reached for turkeys. They conclude with 50–100% certainty (more likely than not) that feed withdrawal periods longer than 6 h will lead to turkeys experience prolonged hunger and with 90–100% certainty (from very likely to almost certain) that feed withdrawal periods longer than 12 h will lead to turkeys experience prolonged hunger.

In game birds there is no need to remove feed as they are not sent directly to slaughter. Similarly, pullets are typically not feed-withdrawn before transportation, as they are not destined for slaughter (Schwartzkopf-Genswein et al., 2012).

In end-of-lay hens, under simulated transport conditions, Beaulac et al. (2020) found the blood physiology altered with time between 10 and 18 h without feed; changes in blood CO_2 demonstrated a greater negative value for hens in 18 h than for hens in 10-h deprivation while the sodium changes had larger positive values with increasing duration. Both haematocrit and haemoglobin level changes were negative for hens in the 10-h deprivation and positive for hens in the 18-h deprivation, with hens in the 14 h being intermediate but not differing from either. There is no scientific data of the effect of less than 10 h feed deprivation, and end-of-lay hens may already experience prolonged hunger with feed withdrawal periods lower than 10 h.

Following 24 h feed deprivation, layers (Damme et al., 1987) have similar to broilers a fasting metabolic rate of about 30 kJ/h (Noblet et al., 2015) as they may be laying at an 80% rate even at the

end of the commercial laying period. The experiments of Damme et al. (1987) did not provide data for shorter periods of fasting, relevant to transport, but did note that poorly feathered birds had increased heat loss of up to 11 kJ/h indicating that they would experience prolonged hunger sooner than well-feathered hens.

Furthermore, end-of-lay hens are still productive with laying eggs, which requires considerable metabolic effort and feed intake. Having laid almost one egg/day for a prolonged period of a year or more, they are metabolically exhausted with few body reserves. That prolonged hunger is aversive to hens may be informed by studies of induced moulting of hens (typically at 80 weeks of age), a management procedure of restricting nutrients or fasting the hens which prolongs the productive laying period. This leads to significant increases of behaviours indicative of frustration, including aggression, and feed-seeking compared with unfasted controls (Forkman et al., 2000; Mazzuco et al., 2011).

Considering the available evidence, the AHAW Panel, based on expert opinion, concluded with 66–100% certainty (from likely to almost certain) that, under thermoneutral conditions, end-of-lay hens will experience prolonged hunger after feed withdrawal from at least 10 h. However, no evidence exists about a possible earlier onset of prolonged hunger.

From an animal welfare perspective, there is no benefit of fasting birds before transport. However, from a hygienic perspective, non-fasted birds will defecate during the journey and increase feather soiling with increased risk for public health. To reduce the level of droppings during transport of birds to the slaughterhouse, on-farm feed withdrawal can be practiced, always considering that the total time of feed deprivation. Four hours of feed deprivation before transport provides a significant reduction in gut contents, that can be accelerated with the intake of water until the time of catching and crating.

3.7.9.2. ABMs

There are no feasible ABMs to assess prolonged hunger during transport.

3.7.9.3. Hazards

Too long feed deprivation

As presented above, the hazard for prolonged hunger is the feed deprivation that is performed routinely as a hygienic measure. Feed deprivation can be performed at different points in time on the farm before catching and crating of the animals.

For broilers and turkeys, deprivation of feed for 6 h results in depletion of glycogen stores in the liver (Warriss et al., 1988; Savenije et al., 2002) and will start to experience prolonged hunger and a negative affective state, which will increase with time so that by 12 h, the welfare consequence of prolonged hunger is almost certain and leading to a weakened condition in the bird.

End-of-lay hens are generally meal fed and have a period of darkness (typically 8 h) at night which is when they are often caught and crated. Thus, they may have a full crop with food reserves to sustain them during transport. During depopulation from large colony cage housing, only part of the flock of end-of-lay hens can be removed each day. The houses are designed to distribute feed to all cages at the same time and it is not cost efficient to distribute feed when half of the house is empty. For this reason, feed withdrawal periods could be long. Exacerbating this is the current practice in the EU which involves a large proportion of end-of-lay hens being transported for longer distances owing to few slaughterhouses that can process them. Hens transported longer distances to slaughter had a greater proportion of body weight loss compared with broilers (Çavuşoğlu and Petek, 2021). Weeks et al. (2012) reported risk factors for DoA in end-of-lay hens to include distance travelled, low body weight, poor feather cover and low ambient air temperatures.

For end-of-lay hens, under thermoneutral conditions, deprivation of feed for 10 h results in prolonged hunger. As end-of-lay hens often have low body weight and fat cover, and may be poorly feathered, their risk of experiencing hunger during transport is increased particularly in conditions leading to cold stress, where energy demand will increase.

3.7.9.4. Preventive measures

To prevent the risk of broilers and turkeys to experience prolonged hunger during transport, the total time without feed should not exceed 6 h.

The longer the on-farm fasting time, the longer will be the total feed withdrawal time related to transport. Therefore, to prevent prolonged hunger during transport, on-farm feed withdrawal should

be avoided (for birds sent to other farms) or reduced to a minimum (for birds sent to slaughter) and the total withdrawal time should always be considered. Nijdam et al. (2005), concluded that to continue feeding broilers until catching resulted in higher body weight at the slaughterhouse and less stress (i.e. a reduction in a negative energy balance).

Communication with live bird catching teams and hauliers and with the slaughterhouse or destination farm (European Commission, 2017a, 2018) for planning and coordinating the arrival of live birds, will reduce transport duration by minimising loading times and waiting time upon arrival of live birds and prevent prolonged hunger.

3.7.9.5. Corrective and mitigative measures

Domestic birds subjected to feed withdrawal periods longer than 6 h will start to experience prolonged hunger and a negative affective state, which will increase with time so that by 12 h or more the welfare consequence of prolonged thirst is almost certain. To mitigate it, the total time of feed deprivation should not exceed 12 h.

To mitigate prolonged hunger in end-of-lay hens, the total time without feed should not exceed 10 h.

Provision of feed during transport could correct the hazard and mitigate prolonged hunger. However, if animals are to be fed in containers, the systems for feeding must be designed in a way to enable all birds to access to sufficient amounts of feed, and water (or liquid/wet feed) must be provided concurrently. If this is not the case, the risk of prolonged hunger in all birds is not reduced. Unless all animals can access feed from their original position in the container, the space allowance, including height, must enable animals to move freely within the container to access feed. This is not common practice today, and it might also increase the risk of injuries (see Section 3.7.2).

The only feasible mitigative measure is to unload the animals and provide feed and water if they are transported to other farms, or slaughter them immediately.

3.7.10. Prolonged thirst

3.7.10.1. Description

Prolonged thirst means that the bird has been unable to get enough water to satisfy its needs and experiences craving or urgent need for water, accompanied by an uneasy sensation (a negative affective state), and eventually leading to dehydration as metabolic requirements are not met. Birds will therefore experience thirst after shorter periods of water withdrawal, thus 'prolonged' represents substantial water imbalance due either to a period without access to water or to water loss via panting or, as is common during transport, to both. Prolonged thirst is regarded as a clearly relevant welfare consequence for all animal categories due to high severity. Prevalence will vary, but most likely this welfare consequence will arise during the journey stage and severity will increase over time till dehydration will arise. However, evidence of dehydration in transported birds is scarce.

The feed provided for domestic poultry is dry, thus drinking behaviour is linked with feeding. Using RFID (Radio Frequency Identification) tags to monitor individual broiler behaviour at 35 days of age, Li et al. (2019) reported both feeding and drinking bouts in the same hour with time spent averaging 11.8 min at feeder and 2.8 min at the drinker. Warriss et al. (2004) found that slaughter age broilers had a mean 1.08 feeding and 1.81 drinking bouts/h but during a 24-h feed withdrawal the drinking behaviour reduced by 52% to an average of 0.93 bouts/h with no difference during the first 4-h period but a 36% reduction after 16–24 h off feed. In contrast, other experimental work has recorded a linear increase in drinking behaviour during 24 h of feed deprivation (Sprenger et al., 2009). There is a need for research that measures actual water consumption rather than the proxy drinking behaviour and to determine when domestic birds experience negative affective states and craving for water.

The minimum passage rate of fluids through the gastrointestinal tract of fed chickens or turkeys is 2–2.5 h (Dansky and Hill, 1952; Tuckey et al., 1958). The welfare implication of water deprivation duration for poultry has mostly been studied using physiological changes, with studies using up to 48 h of water deprivation or feed and water deprivation (Rault et al., 2016). Various physiological indicators of prolonged thirst have been measured such as those related to dehydration (osmolality, packed cell volume, plasma electrolytes), metabolic status (glucose and lactate concentrations) and stress physiology (corticosterone and vasotocin concentrations) but inconsistent outcomes were obtained resulting from differences in strain, sex, age, production stage and environmental conditions between studies (Koike et al., 1977; Arad et al., 1985; Stallone and Braun, 1986; Knowles et al., 1995; Saito and Grossmann, 1998; Iheukwumere and Herbert, 2003).

Vanderhasselt et al. (2013) experimentally withdrew water from broilers for 0 (control), 6, 12, 24 and 48 h. They concluded that 'of all indicators measured, plasma chloride concentration is the most suitable to detect the effects of dehydration during transport'. After 6 h of water withdrawal, plasma chloride concentration was increased but it did not rise further with longer withdrawal. The best indicators of medium-term water deprivation (6–12 h) were creatinine and sodium. Haematocrit followed approximately the same pattern as creatinine during the first hours of water withdrawal but showed no further increase when withdrawal exceeded 12 h.

Considering the available evidence, the AHAW Panel, based on expert opinion, concluded with 66–100% certainty (from likely to almost certain) that broilers subjected to water deprivation periods longer than 6 h will experience prolonged thirst as demonstrated by the increase of plasma chloride as sign of dehydration. Broilers subjected to water withdrawal periods longer than 12 h will show an increase in creatinine and therefore the AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) that they will experience prolonged thirst which is detrimental to their welfare. There is a lack of evidence regarding indicators of thirst following water withdrawal.

In turkeys, Duke et al. (1997) removed feed together with water for 4–8 and 12 h. Even after 12 h water removal, there were no evident signs of carcass dehydration. This low level of dehydration was not, however, statistically significant, as indicated by non-significant changes in packed cell volume (PCV) from blood samples.

In conclusion, considering the limited available evidence, the AHAW Panel, based on expert opinion, reached similar conclusions for turkeys as for broilers although they are less susceptible to dehydration.

For end-of-lay hens, there is little scientific evidence regarding when hens experience negative affective states and craving for water. Haskell et al. (2004), using water deprivation in an operant conditioning test, showed that 2 h of water deprivation induced redirected aggression towards a subordinate hen. Similar aggressive behaviours have been demonstrated in laying hens deprived of water for 6 h (Forkman et al., 2000) as well as pacing, excessive preening or redirected pecking in earlier work by Duncan and Wood-Gush (1972) of feed or water-deprived hens.

Rault et al. (2016) used a motivation test based on passing through a narrow, vertical gap to access water to assess the behavioural changes in laying hens following various durations of water deprivation. Hens were highly motivated to drink after 12 h of water withdrawal (the shortest duration tested) and were willing to squeeze through even the narrowest gap to access water. However, the exposure to additional stressors during transport, including too high effective temperature, will lead to end-of-lay hens becoming thirsty prior to 12 h of deprivation. In a study designed specifically to determine responses of hens to cold and heat exposure as well as simulated transport durations of 4, 8 and 12 h, Beaulac et al. (2020) found signs of dehydration both with increased duration of water deprivation and with exposure to heat (30 °C combined with 30% or 80% RH). Both changes in blood sodium and live shrink increased linearly with time, being significantly different at each time point, and were also significantly greater for heat stressed hens owing to moisture loss through panting (which was also observed at 30°C). Moreover, end-of-lay hens are still productive, with a high proportion of them producing eggs in the oviduct or laying during transport and eggs are ~ 77% water (UK Dept of Health[7]).

With 24 and 32 h off water lead to changes on most behaviours (e.g. drinking duration), whereas changes were seen in some behaviours at 18 h (e.g. location of the hen close to the drinker, reduced standing). They concluded that water deprivation induces behavioural changes such as drinking duration up to 24 h from water withdrawal. However, even under the experimental conditions in their usual cage housing,

Considering the available evidence, the AHAW Panel, based on expert opinion, concluded with 66–100% certainty (from likely to almost certain) that end-of-lay hens subjected to water deprivation periods longer than 6 h will experience prolonged thirst as demonstrated by changes in behaviour including an increase in levels of redirected aggression towards other hens. End-of-lay hens subjected to water withdrawal periods longer than 12 h have increased levels of blood sodium, are highly motivated to drink and work hard to access water. The AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) that they experience prolonged thirst.

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[7] https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/167973/Nutrient_analysis_of_eggs_Analytical_Report.pdf

3.7.10.2. ABMs

No ABMs are considered feasible for routine assessment during transport.

3.7.10.3. Hazards

Water deprivation for too long.

As presented above, the hazard for prolonged thirst is deprivation of access to drinking water. Considering that birds might be spending a considerable time in the transport containers before the journey starts (e.g. considering loading time) and in lairage after the journey ends (e.g. time spent in lairage before birds are uncrated), the total time without water potentially becomes very long, resulting in an increased risk of prolonged thirst. Domestic birds subjected to water withdrawal periods longer than 6 h will start to experience prolonged thirst and a negative affective state, which will increase with time so that by 12 h or more the welfare consequence of prolonged thirst is almost certain.

End-of-lay hens may have an increased risk of experiencing prolonged thirst as a consequence of producing eggs of high-water content during transport.

3.7.10.4. Preventive measures

Poultry should have easy access to water until they are caught and placed in containers.

To prevent the risk of poultry experiencing thirst during transport, the total time of water deprivation should not exceed 6 h in the safe or alert thermal zone.

Provision of water during transport might prevent, correct or mitigate prolonged thirst. However, if animals are to be watered in containers, the systems for watering must be designed in a way enabling all animals access and ingest sufficient amounts. If this is not the case, the risk of prolonged thirst is not reduced in all birds.

For end-of-lay hens, some approved vehicles are equipped with stainless steel pipe systems as a supply device, which can be filled with water from a large tank on the vehicle and ideally provide a drinking nipple per transport crate that protrudes into the side of the crate. However, due to the low space allowance, not all the animals of the containers can reach a drinking device that is unknown to them and take in a sufficient amount of liquid and nutrients there (German Competent Authority in their response to the Public Consultation, provided to EFSA).

Communication with live bird catching teams and hauliers and with the slaughterhouse or destination farm (European Commission, 2017a, 2018) for planning and coordinating the arrival of live birds, will reduce transport duration by minimising loading times and waiting time upon arrival and prevent prolonged thirst.

3.7.10.5. Corrective and mitigative measures

Domestic birds subjected to water withdrawal periods longer than 6 h will start to experience prolonged thirst and a negative affective state, which will increase with time so that by 12 h or more the welfare consequence of prolonged thirst is almost certain. To mitigate it, the total time of water deprivation should not exceed 12 h in the safe thermal zone. This time should be considerably reduced during hot weather. However, the extent of reduction of the period of water deprivation is not known.

The most feasible mitigative measure is to unload the birds and provide water or slaughter them immediately.

3.8. Transport duration

For the overall assessment of transport duration, it should be considered not only the journey duration (period the vehicle is in transit), but also the time the animals are in the containers and the prior feed withdrawal period on farm. Regardless of how optimal the conditions of the transport provided are, domestic birds can potentially be exposed to a number of hazards during transport that might, either on their own or in combination, result in impaired welfare consequences. The exposure to these hazards ends only when the transport ends and the animals are uncrated from the containers. Any aversive effects associated with exposure overstimulation or to restriction of water and feed are likely to increase with transport duration and could interact with other factors, such as temperature, that might also change during a journey. The recommended maximum transport time is based on an overall assessment across the highly relevant welfare consequences, and it is based on the scientific

evidence combined with expert opinion. The recommendation for transport time is based on the assumption that recommendations on microclimatic conditions and space allowance are followed.

Sensory overstimulation and motion stress: As soon as birds are crated and loaded into the vehicle, and during all time when the vehicle is moving, all domestic birds are to some extent exposed to motion stress and often also, at least periodically, to sensory overstimulation. As a consequence of the vehicle motion, animals experience stress potentially leading to fatigue and negative affective states such as fear and distress. The duration of the welfare consequence depends on transport duration and onset of vehicle motion. Because of the constant presence of motion stress, it is not possible to estimate a temporal cut-off for onset of this welfare consequence after initiation of the journey.

Prolonged hunger: The welfare consequence prolonged hunger is regarded as highly relevant in the transit stage. Prevalence is expected to be high, as no studies have documented the successful feeding of domestic birds in the vehicle during journeys. Depending on factors such as time off feed before journey start, domestic birds may not be hungry during the initial phase of the journey, but hunger will develop over time. The duration of prolonged hunger depends on time of on-farm withdrawal and transport duration until the birds are uncrated and slaughtered or provided with feed, and severity is expected to increase with increasing duration, as the need for feed becomes more and more problematic for the animals. Prolonged hunger may lead to exhaustion and a weakened condition. It is concluded that domestic birds subjected to total feed withdrawal periods longer than 6 h will start to experience prolonged hunger and a negative affective state, which will increase with time so that by 12 h or more the welfare consequence of prolonged hunger is likely to almost certain (90–100% certainty). It is likely to almost certain that end-of-lay hens travelling under thermoneutral conditions will experience prolonged hunger after 10 h of total feed withdrawal.

Prolonged thirst: The welfare consequence prolonged thirst is regarded as highly relevant in the transit stage. Prevalence may be high as water is not provided to the animals. So far, no documentation for proper intake of water, even in journeys on vehicles fitted with drinkers, are available. Depending on factors such as time off water before journey start and/or microclimatic conditions before and during the journey, domestic birds may not be thirsty during the initial phase of the journey, but thirst will develop over time. The duration of prolonged thirst depends on transport duration, and severity is expected to increase with increasing duration, as the need for water becomes more and more problematic for the animals. The available data do not allow a detailed determination of the interval between journey start and initiation of thirst, especially due to the lack of repeated sampling.

It is concluded that domestic birds subjected to water withdrawal periods longer than 6 h will start to experience prolonged thirst and a negative affective state, which will increase with time so that by 12 h or more the welfare consequence of prolonged thirst is likely to almost certain (90–100% certainty).

Other summarising considerations: In addition to the welfare consequences summarised above, the risk of animals experiencing pain and/or discomfort, as well as the severity of it, will also increase with transport time. This may happen if the animals had a pre-existing painful condition. Even though this should not happen, it is not always possible to identify pathological conditions in domestic birds while they are on the farm, as they are known to not show overt signs of, e.g. discomfort.

In addition, animals which did not show a health condition before the journey may get injured during the transport due to e.g. catching and crating, and the pain and discomfort from such conditions will continue, and likely worsen, until the animal can be uncrated. In this weakened state, domestic birds are often less able to cope with the extra challenges associated with transport, and their condition is likely to deteriorate with time and transport duration.

The pain and/or discomfort from both types of the above-mentioned health conditions are not expected to be prevalent, but for the affected animals the consequences may be severe and will develop over time. The duration of these negative affective states will depend on transport duration, as they cannot be terminated until the journey is stopped and the birds are uncrated. During a journey, such health conditions may lead to suffering. It is, however, not possible to establish a temporal cut-off for when pain and/or discomfort may start. Evidence on continuous welfare consequences involving stress and negative affective states suggests that limiting transport time would reduce the exposure to the hazards. Scientific evidence on how the progressively developing welfare consequences change with increasing transport duration is limited. Based on the presence of the continuous welfare consequences combined with the limited data on the progressively developing

welfare consequences, it is not possible to scientifically define a maximum journey duration that will not impair animal welfare.

In order to take into account all welfare consequences, continuous as well as progressively developing, 12 h can be suggested as a limit to transport duration (including on-farm feed and water withdrawal) until being uncrated when animals are kept under the suggested microclimatic and space allowance conditions, and no water or feed is available to the animals during that period. Importantly, this does not imply that the welfare consequences necessarily occur after a transport duration of 12 h, or that no welfare consequences occur before 12 h, as there are many factors other than transport duration that affect the risk of welfare consequences during a journey. In end-of-lay hens 10 h can be suggested as a limit to transport duration (including on-farm feed and water withdrawal) until being uncrated when animals are kept under the suggested microclimatic and space allowance conditions, and no water or feed is available to the birds during that period.

To reduce the transport duration, transport should be planned in advance form the stage of loading to arrival and uncrating (see Section 3.2.2). This includes the coordination of the different stages of the transport (e.g. itinerary for loading and unloading, location of any driver resting places/stops during the journey), estimation of their duration and time of arrival. Effective planning includes communication with live bird catching teams and hauliers and with the slaughterhouse or destination farm (European Commission, 2017a, 2018) for planning and coordinating the arrival of live birds. The potential welfare consequences and their possible hazards (such as analysis of weather forecast) should be identified, and preventive and mitigative measures implemented. Contingency plans will help the driver and the transport company to ensure the security and the welfare of the animals in case of emergency and to minimise transport time.

3.9. Iceberg indicators

Iceberg indicators are indicators that can be used to obtain a quick overview on possible welfare problems, as they may reflect several welfare consequences in an integrative manner. Their presence implies that not only the animals that show them will experience these welfare consequences.

They provide an overall assessment of welfare, just as the protruding tip of an iceberg signals its submerged bulk beneath the water's surface (FAWC, 2019). Iceberg Indicators are not exclusively linked to a specific welfare consequence but might be associated with the additive consequence of several welfare consequences.

The most relevant iceberg indicator in domestic birds at transport is dead on arrival (DOA).

3.9.1. Dead on arrival (DOA)

Any bird that is found dead in the container or at the spot is considered DOA (Welfare Quality[®], 2009, from EFSA AHAW Panel, 2019). DOA is normally expressed in percentage (%), that is the number of birds found dead in the containers at uncrating/total number of birds transported on the vehicle multiplied by 100. To be valid as iceberg indicator DOA should be assessed for each transport. For this to work, it will be necessary to identify birds from every individual journey separately. At arrival, the assessment of DOA is considered an iceberg indicator of the transport conditions. This ABM is not sensitive or specific to a single welfare consequence since it can have a different or a multi-factorial origin. However elevated levels of DOA are always indicative of poor animal welfare at one or more stages of transport. As it is strongly associated with several welfare consequences, this ABM has proved useful for inspection purposes.

DOA% in broilers varies considerably between studies (Table 17), with means ranging from 0.11% to 0.68% (Jacobs et al., 2017a) and several transports having no mortality.

Table 17: Literature overview showing reference, country of study, mean and range DOA percentage of broilers

Visser et al. (2014) (the Netherlands)	0.11 (0.04–0.26)
Chauvin et al. (2011) (France)	0.18 (0.00–1.40)
Haslam et al. (2008) (United Kingdom)	0.12 (0.00–0.64)
Petracci et al. (2006) (Italy)	0.35 (0.04–2.00)
Vecerek et al. (2006) (Czech Republic)	0.25 (unknown)
Ritz et al. (2005) (United States)	0.68 (unknown)

Warriss et al. (2005) (England)	0.13 (0.03–3.10)
Nijdam et al. (2004) (the Netherlands)	0.46 (0.00–16.61)
Warriss et al. (1992) (England)	0.19 (0.00–15.80)

DOA also shows high variability in other poultry categories and species.

Petracci et al. (2006), during a 4-year period, surveyed the incidence of DOA birds over 33 broiler, 11 turkey and 19 spent hen abattoirs representing the majority (around 70%) of the Italian poultry slaughter plants. The overall average incidence of DOA was found to be 0.35% in broilers, 0.38% in turkeys and 1.22% in spent hens. Di Martino et al. (2017b), surveyed DOAs of turkey and laying hens transported to three large abattoirs in northern Italy in a 3-year period, the median DOA was 0.14% in turkeys, and 0.38% in hens. Voslarova et al. (2007a) reported an overall mortality of 0.279% in turkeys during transport for slaughter in the Czech Republic during the period 1997–2006. Overall mortality among turkeys transported for slaughter in the Czech Republic from 2009 to 2014 was 0.147% (Machovcova et al., 2017).

Although ideally DOA should be 0%, and this is often achieved, variable levels of DOA are likely to occur during transport, sometimes with no apparent cause.

Different studies have shown a clear correlation between on-farm mortality and DOA (Haslam et al., 2008; Chauvin et al., 2011; Weeks et al., 2012, BenSassi et al., 2019). However, DOA is known to be even more related to transport than to farming conditions. Levels of DOA have been linked with environmental conditions (e.g. temperature and humidity), transport conditions (e.g. longer distance travelled; Oba et al., 2009), waiting time and animal-based variables such as genotype, sex and live weight (Nijdam et al., 2004; Chauvin et al., 2011; Jacobs et al., 2017a,b). Increases in temperature, transport distance, waiting time, broiler weight and the practice of thinning can all increase DOA (Villarroel et al., 2018).

Another major causal factor is traumatic injury (Gregory and Austin, 1992). Reduced mortality will therefore result from closer control of environmental conditions during transit and more careful bird handling to reduce trauma (Warriss et al., 1999).

Impact of restriction of movement on DOA

Chauvin et al., 2011 found the provision of more space allowance during transport (EFSA, 2004) was associated with reduced mortality in broilers, most likely due to reduced heat stress.

Impact of heat stress on DOA

According to Machovcova et al. (2017), it appears that heat stress can be a major factor in the birds' mortality. Since the highest death rates among turkeys transported for slaughter in the Czech Republic, from 2009 to 2014, occurred during the warmer seasons (14–21°C). Vieira et al. (2011) reported that the mortality incidence of broilers in Brazil in the summer was 0.42%, followed by spring (0.39%), winter (0.28%) and autumn (0.23%), average 0.33%.

Di Martino et al. (2017a) found that turkeys subjected to ambient temperatures higher than 26°C were found to have considerably increased DOA. The temperature within the transport containers was likely at least 30°C.

Data provided by the Netherlands Food and Consumer Product Safety Authority (Pladmin data on Laying hens, 2017–2021) for end-of-lay hens from the years 2017, 2018, 2019, 2020 and 2021 showed an increased DOA rate on days (except for year 2017) Figures 24 and 26). These findings are also confirmed in their advice on transport of finisher broilers.[8]

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[8] Advice of BuRO on the transport of finisher pigs and broilers during (extremely) high temperatures, The Dutch Office for Risk Assessment & Research (BuRO), 2020 https://english.nvwa.nl/about-us/documents/consumers/food/safety/documents/advice-of-buro-on-the-transport-of-finisher-pigs-and-broilers-during-extremely-high-temperatures
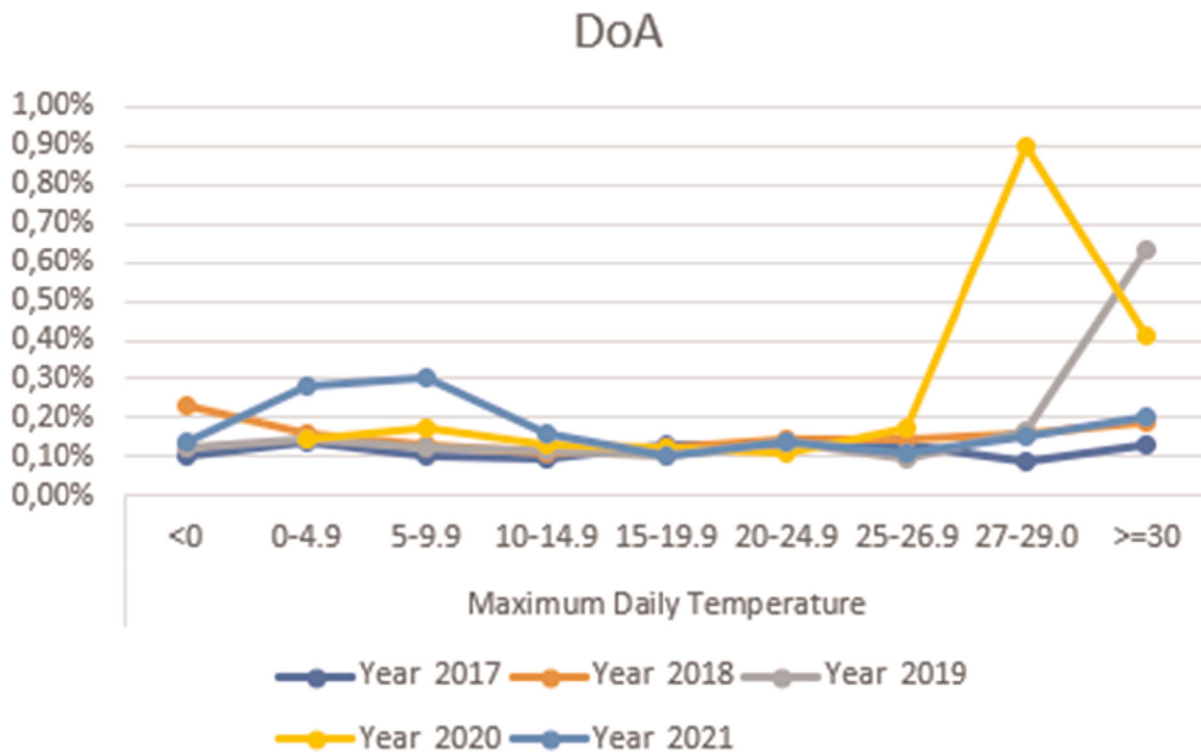
Figure 24: Average DOA per maximum daily temperature range between in 2017, 2018, 2019, 2020 and 2021 (graph produced by EFSA with data from the Netherlands Food and Consumer Product Safety Authority)

Impact of cold stress on DOA

Low ambient temperatures also appear to increase transport-related mortality. The lowest temperatures (−6°C to −3.1°C) observed in studies were associated with higher death rates (0.179%) in comparison to temperature intervals from −2°C to 1.9°C (0.079%) and 2 to 5.9°C (0.077%) (Vecerek et al., 2016). Weeks et al. (2012), in a large survey of end-of-lay hens slaughtered in the UK during 2009, also found lower ambient air temperatures (−2.2°C) to be a risk factor for increased DOA. Večerková et al. (2019) confirmed that the greatest number of deaths in end-of-lay hens occurred at a temperature from −6.0°C to −3.1°C (0.718%, i.e. 15% of all deaths) and at a temperature of −3.0°C to −0.1°C (0.655%; i.e. 18% of all deaths).

The greatest mortality (0.55%) was associated with transports carried out in winter months whereas the lowest death losses (0.30%) were found in chickens transported for slaughter in summer months Vecerek et al. (2016).

Impact of transport duration on DOA

Nijdam et al. (2004) examined the risk factors for DOA in Dutch broiler production and suggested that in particular, reduction of transport and lairage times might reduce DOA. Based on published figures for mortality during the rearing period, Warriss et al. (1992) estimated that journeys of up to 4 h increased mortality by about ten-fold and journeys over 4 h nearly 19-fold.

A study by Voslarova et al. (2007b) compared transport mortality rates (DOA) across poultry species for the period 1997–2006 in the Czech Republic. The study analysed data for broilers, hens and cockerels, turkeys, ducks and geese transported for slaughter. The correlations with journey distances in the categories up to 50 km, from 51 km to 100 km, from 101 km to 200 km, from 201 km to 300 km and over 300 km were determined. The highest mortality rates occurred in hens and cockerels (1.013%), followed by turkeys (0.272%), broilers (0.253%), ducks (0.103%) and geese (0.056%). Mortality rates correlated highly with transport distance. The lowest mortality rates were for the shortest transport distances; in broilers (0.154%), turkeys (0.164%) and hens and cockerels (0.595%) for a transport distance up to 50 km; and in ducks (0.069–0.111%) and geese (0.021–0.053%) for transport distances up to 300 km. Highest mortality rates in hens and cockerels (1.892%), turkeys (0.341%) and broilers (0.536%) were observed for transport distances over

200 km, while in ducks (0.147%) and geese (0.253%), highest mortality rates were with transport distances exceeding 300 km.

Vecerek et al. (2016) revealed an increase in transport-related mortality rates in broiler chickens transported for slaughter in the Czech Republic compared to a previous study by Vecerek et al. (2006). In the period from 2009 to 2014, overall transport-related mortality of broiler chickens was 0.37%. It ranged from 0.31% to 0.72%, the increase approximately corresponding to the increasing transport distance.

Di Martino et al. (2017b) found that in turkey hens, transportation lasting longer than 2 h was associated with higher DOA, which progressively increased with travel duration, remaining constant between 4 and 6 h and peaking at 8 h (median: 0.57%). Machovcova et al. (2017) reported that overall mortality among turkeys transported for slaughter in the Czech Republic from 2009 to 2014 was 0.147% The lowest mortality (0.023%) was found in turkeys transported for distances up to 50 km; longer distances were associated with increasing death rates, with the highest losses (0.543%) recorded for distances from 201 to 300 km.

Impact of the interaction of transport duration and thermal stress on DOA

Interaction transport time × ambient temperature appears to be one of the main factors associated with DOA (Nijdam et al., 2004). Under temperature extremes longer transport durations increase thermal stress and enhance the risk of DOA.

Voslarova et al. (2007a) have reported an analysis of DOA in hens and roosters in the Czech Republic. Over a 7-year period up to 2004, the mean DOA was 0.93%. DOAs in hens and roosters increased from 0.592% at transport distances up to 50 km to 1.638% at transport distances up to 300 km. Bird mortality was also influenced by the season of the year. Higher mortality rates were reported during the cold months of the year (October–April), indicating birds were cold stressed.

Večerková et al. (2019) found the overall mortality of hens during transport for slaughter in the period from 2010 to 2017 to be 0.516%, although the number of dead laying hens in individual consignments varied significantly. Lower outside temperature was connected with significantly increased hen mortality. The results show that the highest number of deaths occurred at a temperature of −6.0°C to −3.1°C and at a temperature of −3.0°C to −0.1°C, i.e. at the lowest temperatures in the monitored period. When transporting at temperatures above 0°C, there were significantly fewer deaths of transported hens. The lowest transport-related mortality was found when the average temperature was between 15°C and 21°C (the highest average temperature in the monitored period). Considering transport distance, the lowest mortality (0.338%) was found in hens transported for distances up to 50 km; longer distances were associated with increasing death rates, with the highest losses (0.801%) recorded for distances from 201 to 300 km (Večerková et al., 2019). These findings document the need for increased care for end-of-lay hens during their transport for slaughter in the winter at lower outside transport temperatures, in particular below 0°C (e.g. by adequate temperature regulation in the means of transport), and of hens transported over longer distances (if the transport distance cannot be reduced).

Voslarova et al. (2016) analysed data on Pekin ducks transported to slaughterhouses between 2009 and 2014 in the Czech Republic. Average transport-related mortality was 0.077%. The lowest transport-related mortality (0.052%) was found for distances shorter than 50 km. The highest mortality rates were found for transport distances of 101–200 km (0.105%). In addition, the season of the year significantly affected transport-related mortality in Pekin ducks. The highest death losses were found in the summer (0.090%), in comparison with transport-related mortality rates in any other season of the year. Shortening transport distances and maintaining a suitable micro-climate inside transport vehicles especially in the summer are thus two important factors that can contribute to reducing transport-related mortality in Pekin ducks in commercial practice.

Assessment of DOA

Inspectors of the Canadian Food Inspection Agency (CFIA) exercise closer scrutiny when the number of broilers or end-of-lay hens arriving DOA exceeds 1% and 4%, respectively. Ritz et al. (2005) commented that a typical USA industry goal is for DOA percentages to be below 0.20% in broilers and broiler breeders. According to animal welfare guidelines developed by the USA National Chicken Council (NCC, 2017) for broilers and broiler-breeders, when DOAs are over 0.5%, a corrective action is required. In the UK according to welfare assurance scheme standards (RSPCA, 2017) the threshold DOA in any load triggering an investigation is 0.5% for end-of-lay hens and 0.25% for broilers, although the goal is for decreasing proportions or no DOA.

DOA is widely collected in broiler slaughter plants throughout Europe as confirmed by the *Study on the application of the broilers Directive and development of welfare indicators: Final Report DG SANTE Submitted by Food Chain Evaluation Consortium* (European Commission, 2017c). The recording of DOA is a legal requirement of the Broiler Directive as it is considered a valid indicator of animal welfare on farm collected at slaughter (ANNEX III MONITORING AND FOLLOW-UP AT THE SLAUGHTERHOUSE), since birds with reduced welfare may be less likely to survive the journey.

Competent authorities in many MSs have set DOA thresholds for broilers, above which an investigation takes place to try to determine the cause; subsequent measures would then be taken against the transporter or the keeper as appropriate.

- Germany $\geq 0.5\%$.
- The Netherlands $\geq 0.5\%$ (signal threshold) $\geq 1\%$ (intervention threshold) (DOA% in broilers on average fluctuates around 0.14% year-on-year).
- Sweden $\geq 1\%$.
- Belgium $\geq 1\%$.
- Italy $\geq 1.5\%$.
- Spain $\geq 2\%$.

The use of DOA as an indicator is also considered by the poultry industry fairly uncontentious since it is a simple, objective, binary indicator. DOA is therefore among the core group of indicators used in industry/voluntary schemes.

To be a valid iceberg indicator, DOA should be collected and monitored for each individual transport. Birds might die during transport due to factors that are not attributable directly to the transport conditions. Therefore, the cause of DOA might not need to be investigated for each individual transport, but if it exceeds a certain level, that might be attributed to the transport conditions. The AHAW Panel suggested that this level might be 0.1% for all domestic birds.

4. Assessment of scenario 2: Road and air transport of day-old chicks

4.1. Introduction

The majority of domestic birds are transported as day-old chicks from hatcheries to production sites. The EU also exports day-old chicks to third countries. In 2020, the number of day-old chicks exported from the EU was more than 200 million. Transport is mainly by road, but air transport is also common practice.

Traditionally, broiler and layer chicks hatch in a hatchery. Upon arrival from the breeding farm, hatching eggs can be stored and disinfected before placement in the incubators. Hatching takes approximately 21 days, depending on the species, and is usually carried out in two steps: incubation phase (~ 18 days) and hatching phase (~3 days). Hatching time is spread across a 'hatch-window', typically 24–48 h (Careghi et al., 2005; Willemsen et al., 2010; Tong et al., 2015), which is dependent on age of the parent stock, egg handling, egg storage time and the incubation conditions (Decuypere et al., 2001). This may result in an age difference between chicks in the same batch of a day or more.

When hatched in hatcheries, following the hatchery procedures, chicks are subjected to a waiting period, most often without access to feed and water, before they are loaded on a vehicle and transported to the farm where they are unloaded and placed in the barn. In some alternative farming systems (e.g. the Patio system) broiler hatching eggs are transported to the farm and hatch there. The newly hatched chicks stay in their rearing barn, meaning they are not transported.

The current EU transport regulation (EC 1/2005) does not set a maximum journey time for transport of day-old chicks, but requires that suitable food and water shall be available in adequate quantities, save in the case of a journey lasting less than 24 h for chicks of all species, provided that it is completed within 72 h after hatching. That means that day-old chicks can be transported for 24 h without feed and water. Therefore, the so-called 'day-old chicks' can in reality be up to 3 days of age when they reach the farm.

It is important to recognise the unique nature of the transportation of newly hatched chicks. No other species is transported in the immediate post-natal period (EFSA AHAW Panel, 2011). Before hatching, chicks prepare themselves for their first few days of life by taking the yolk, a store of nutrients, into the abdominal cavity. The yolk 'comprises approximately 18% of the body weight (BW) of chicks and provides immediate post-hatch energy and protein for maintenance and growth' (MacLeod, 1980; Freeman and Manning, 1984). However, commercial broiler chicks have been bred, in

part, for unnaturally high metabolic rates, so the yolk may be nutritionally insufficient for these purposes. Absorption of the yolk should enable chicks to survive at least a short time without outside nutrients, but they are in most of cases transported without food or water, which appears to be challenging, at least for meat production chicks (HSUS, 2008).

4.1.1. Stages of transport

Transport of day-old chicks as described in this scientific opinion consists of five stages.

1) *Preparation* includes planning of the journey and assessment of fitness for transport.
2) *Loading* includes placing chicks in boxes (crating) and loading of boxes onto the vehicle.
3) *Journey* includes the movement of chicks by vehicle and intermediate stops along the way until the last place of destination is reached.
4) *Arrival* includes the period from arrival of the vehicle to unloading of the last box from the vehicle, in each destination farm.
5) *Uncrating* includes removing the chicks from the boxes.

4.2. Stage 1: Preparation of day-old chicks for transport by road and air

4.2.1. Description

The phase of preparation of day-old chicks for transport has the dual objective of preparing chicks for their delivery on farm and checking that they are fit for transport.

After hatching, chicks are taken from the hatching chamber in trays and go through a number of hatchery procedures, including separating chicks from eggshells and debris, sorting out second-grade chicks, vaccination, eventually sexing (layers and usually only slower-growing hybrids), counting and crating, which usually takes usually 2–4 h (Bergoug et al., 2013; Hedlund et al., 2019). Some animals can also be subjected to mutilations (e.g. beak trimming, toe clipping) in the hatchery. Separation and further processing are usually done by automated systems in integrated production such as boilers or layers and involves rollers and high-speed conveyor belts that transport chicks through the hatchery. Hatchery procedures in general have been shown to be stressful for chicks and may have long-term consequences on behaviour and stress reactivity (Hedlund et al., 2019).

Planning of the transport, including the decision about stocking densities in the box, and the determination of the length of the journey and the number of stops, is part of the preparation stage of transport.

Traditionally the chicks do not receive food or water from hatching until they arrive at the farm, so the chick uses the yolk sac as a source of nutrition (Nielsen et al., 2011). However, watery gel feed may be provided in some long transport (e.g. air transport) and for some species (e.g. day-old quail chicks).

4.2.2. Fitness for transport

4.2.2.1. Description

Please refer to Section 1.1 for domestic birds.

4.2.2.2. Conditions leading to day-old chicks being unfit for transport

The principal conditions which will make day-old chicks unfit for transport are:

- Poor chick quality.
- Inability to stand.
- Fractures (legs, wings) and dislocations.

The list is not exhaustive but is based on the limited published evidence and the opinion of the AHAW Panel.

Poor chick quality

The Pasgar score (Boerjan, 2002) may be used to determine chick quality based on five criteria: (1) Navel condition (black button or leaky navel); (2) Yolk sac (large size of the residual yolk sac); (3) Red hocks (red or swollen hocks); (4) Abnormal beak (red beak or nostrils contaminated with albumen); and (5) Low alertness. For each of the five criteria, one point is subtracted from 10, with chicks scoring 10 being free of any abnormality and 5 being the lowest score. Experimental work by van de

Ven et al. (2012) found high levels of mortality (62.5% at day 7) for broiler chicks with low Pasgar scores, and other quality criteria, including rare physical abnormalities such as an open skull, crossed beak or four legs, although such chicks are normally culled at the hatchery.

Tona et al. (2003) published a scoring system to evaluate chick quality (Table 18). The main difference with Pasgar's score was the addition of an eye condition (open and bright, open and not bright, closed), the down condition (clean and dry, wet, dirty and wet), the leg condition (normal legs and toes, one or two legs infected) and suboptimal navel quality (van de Ven et al., 2012).

According to Table 18, DOC might not be fit for transport when the scoring of any of the parameters is 0. This will occur when the activity is weak, when they are dirty and wet (there is a risk of any of them experiencing cold stress during transport), if they present body with swallowed large yolk and rather hard to touch, closed eyes, two infected legs, navel not closed and discoloured, large remaining membrane and very large remaining yolk.

Table 18: Assessment of different parameters for determining chick quality (from Tona et al., 2003)

Parameters	Assessment	Characteristics	Scores
Activity	Activity is assessed by laying the chick on its back to determine how quickly it returned to its feet. A quick spring back onto its feet was regarded as good but trailing back onto its feet or remaining on its back was assessed as weak.	Good Weak	6 0
Down and appearance	The chick body was examined for dryness and cleanness. It was regarded as normal if it is dry and clean. If it is wet or dirty or both (which can be a source of contamination), then it is not good.	Clean and dry Wet Dirty and wet	10 8 0
Retracted yolk	The chick was put on its back obliquely on the hand palm until abdominal movement totally stopped. The height of its abdomen was estimated. The consistency of the abdomen to touch was then estimated. If the height of abdomen was estimated to be higher and harder to touch than normal, then yolk retracted was regarded as large and consistent.	Body with normal swallowed yolk Body with large yolk and rather hard to touch	12 0
Eyes	The chick was put on the legs, and its eyes were observed. The state of brightness and wideness of the gape of the eyelids were estimated.	Opened and bright Opened and not bright Closed	16 8 0
Legs	The chick was put on its feet to determine if it remained upright well. The toes were examined for their conformation. If the chick remained upright with difficulty, articulations of the knees were examined to detect signs of inflammation or redness or both.	Normal legs and toes One infected leg Two infected legs	16 8 0
Navel area	Navel and surrounding areas were examined for closure of the navel and its coloration. If the colour was different from the skin colour of the chick, then it was regarded as bad.	Completely closed and clean Not completely closed and not discoloured Not closed and discoloured	12 6 0
Remaining membrane	Observation of the navel area allowed estimation of the size of any remaining membrane. The size of any remaining membrane was classified as very large, large or small.	No membrane Small membrane Large membrane	12 8 4
Remaining yolk	Observation of the navel area allowed estimation of the size of any remaining yolk. The size of any remaining yolk was classified as very large, large or small.	No yolk Small yolk Large yolk Very large yolk	16 12 8 0

<u>Inability to stand</u>

Any chick unable to stand is unfit to be transported.

<u>Fractures (legs, wings, etc.) and dislocations</u>

Chicks showing fractures and dislocated articulations should not be transported, since they are likely to experience pain.

4.3. Stage 2: Loading of day-old chicks

4.3.1. Description

This stage includes putting chicks in transport boxes at the hatchery and moving the chicks (in boxes) onto the vehicle and sometimes from a road vehicle to an aircraft.

Hatcheries make use of plastic or disposable transportation crates (Figure 25) with a standard size of $60 \times 40 \times 12$ cm, containing 90 chicks under normal circumstances, which equals a space availability of 26.7 cm^2 per chick. Day-old pheasants and partridges are handled and put in boxes of $49.7 \times 26.5 \times 12.7$ cm (Figure 26). These boxes contain approximatively 80 pheasants (16.5 cm^2/chick) and 100–120 (13.2–11 cm^2/chick) partridges – the stocking densities can be adjusted according to climatic conditions.

The crating of the chicks is usually automatic, via a conveyor belt that dispenses chicks in boxes. The boxes are then stacked on carts, leaving space between the two stacks for ventilation and placed in a holding room (lairage) with usually controlled temperature until loading of the vehicle (Lambrecht et al., 2020). In large consignments of chicks, typical for broiler chickens, laying hens and turkeys, the boxes are loaded by forklift onto the vehicle. For smaller productions, like quails or game birds, the crating of the chicks and loading of the boxes may be carried out manually.

Figure 25: Plastic crate used for transport of day-old chicks. The size is $60 \times 40 \times 12$ cm (Courtesy of Malcolm Mitchell)

Figure 26: Disposable transport box used for day-old chicks of pheasants during transport. The size is 49.7 × 26.5 × 12.7 cm (Courtesy of Virginie Michel)

4.4. Stage 3: The journey of day-old chicks by road and air – the journey

4.4.1. Description

Road transport

The vehicles used to transport day-old chicks are usually equipped with air conditioning systems, as well as temperature, relative humidity and gas sensors for environmental control inside the load.

Once loaded in the vehicle, the boxes with the day-old chicks can be transported directly to the farm, where they will be placed, or they may undergo different stops for the delivery of other chicks to several farms, depending on the type of production and on the number of animals to deliver. The chicks can also be transported to an airport and unloaded for subsequent air transport.

Lambrecht et al. (2020) documented current practices for transport of day-old chicks in Belgium. The journey reportedly takes on average 1.5 h, but can last up to 11 h. The use of a single vehicle for different farms means that day-old chicks destined for the last farm on the route will experience the longest journey times.

Air transport

High quality breeding stock are shipped mainly in the form of day-old chicks, turkey poults, ducklings and hatching eggs worldwide by air cargo over very long distances and therefore by plane. Due to the long journeys, food watery gels are provided (Mitchell, 2009).

Air transport contributes to additional challenges that can adversely affect chick welfare before, during and after flights. As extremely low temperatures and atmospheric pressure are found at the altitude commercial planes fly, cargo holds are pressurised, heated and ventilated to provide appropriate environmental conditions for live animal transport. Possibly due to the lack of environmental control (mainly climatic environment) during the holding stages preceding and following flights (loading/unloading/waiting), these periods can be even more detrimental to the chicks' welfare than the flights themselves (HSUS, 2008).

4.5. Stage 4: Arrival of day-old chicks

4.5.1. Description

At arrival, the day-old chicks are usually unloaded without delay. Indeed, in case of partial delivery, the vehicle has to continue the journey; therefore, the boxes that should stay on the farm are unloaded as soon as possible at each stop.

4.6. Stage 5: Uncrating of day-old chicks

There has been no research on the handling of chicks upon arrival, when they may either be gently tipped out or removed manually from the transport containers. Handling of chicks in the barn may

have an effect on chick welfare. Additionally, the way the barn is prepared (e.g. litter and air temperature, brooding systems, access to feed and water) will have an effect of chickens' welfare and health.

4.7. The highly relevant welfare consequences identified for transport of day-old chicks

As explained in Section 2.2.1, an exercise based on expert knowledge elicitation was performed to identify the highly relevant welfare consequences per each stage of transport of day-old chicks. Welfare consequences were not identified for the preparation stage, as they will mainly appear at later stages of transport.

Table 19 shows the highly relevant welfare consequences for each stage of the transport. The description and assessment of the welfare consequences, as well as their prevention and mitigation are provided in this chapter. For each welfare consequence, it is explained to each stage of the transport it applies.

Table 19: Welfare consequences identified as highly relevant for road and air transport to production sites. (n.a. = non-applicable, x = highly relevant, − = not relevant)

Transport scenario 2: Transport of day-old chicks by road and air to production sites					
Welfare consequence	*Preparation*	*Loading*	*Journey*	*Arrival*	*Uncrating*
Handling stress	*n.a*	x	−	−	x
Sensory overstimulation	*n.a.*	x	x	x	x
Motion stress	*n.a.*	−	x	−	−
Heat stress	*n.a.*	−	x	x	x
Cold stress	*n.a.*	x	x	x	x
Prolonged hunger	*n.a*	x	x	x	x
Prolonged thirst	*n.a.*	x	x	x	x

During the transport, chicks experience several welfare consequences that will worsen with the increase of journey length. Handling stress cannot be avoided since animals have to be crated and then loaded into the vehicle. Additionally, they might experience sensory overstimulation, and motion stress depending on journey conditions. They may also experience cold stress or heat stress depending on environmental and microclimatic conditions within their transport boxes. In the majority of cases, chicks are not provided with food and water at the hatchery or during the journey (except for some species such as quails or in certain types of transport such as air transport). As they usually do not have access to food and water since they hatched, they will start experiencing prolonged hunger and thirst as their yolk sac reserve decreases. Major causes of losses in transit and post-transport mortality and morbidity are dehydration and undernutrition (Xin and Lee, 1997).

4.7.1. Handling stress

4.7.1.1. Description

Handling stress in day-old chicks refers to stress and/or negative affective states such as pain and/or fear resulting from human or mechanical handling, in this context (e.g. when put in a crate). Therefore, this welfare consequence appears during the loading and uncrating stages.

After pulling (removal of the chicks from the hatcher) newly hatched chicks are crated (Hedlund et al., 2019), which usually takes 2–4 h and is usually done by automated systems involving rollers and high-speed conveyor belts (Knowles et al., 2004). Chicks are crated manually or mechanically (Lambrecht et al., 2020). No impact of hatchery processes on welfare or production of broilers was reported, but increasing drop heights and conveyor belt acceleration led to disorientation and discomfort in the chicks (Knowles et al., 2004; Giersberg et al., 2020).

For these reasons, the prevalence of handling stress (due to manual or automated manipulation when crating) is considered high, as all day-old chicks are subjected to it. The severity varies from medium to high depending on the number and quality of the handling procedures, and the duration varies from short- (only during the crating process) to medium-term duration. Therefore, this welfare consequence is considered as highly relevant for day-old chicks.

4.7.1.2. ABMs

Pharmacological and behavioural laboratory studies link expression of distress call with negative affective state. As such, there is an *a priori* expectation that distress calls on farms indicate not only physical, but also emotional welfare (Herborn et al., 2020). Using whole-house recordings on 12 commercial broiler flocks (n = 25,090–26,510/flock), Herborn et al. (2020) showed that early life (day 1–4 of placement) distress call rate can be linearly estimated using spectral entropy. Spectral entropy was predictive of important commercial and welfare-relevant measures: low median daily spectral entropy predicted low weight gain and high mortality, not only into the next day, but towards the end of production.

The orientation and posture of chicks on the conveyor belt as well as escape attempts and wing flapping can also be indicators of handling stress. The presence of chicks on the floor is a simple measure of the consequence of inappropriate conveyor settings and rough handling and therefore handling stress for chicks.

ABMs that are considered feasible for assessing handling stress in day-old chicks during transport are given in Table 20.

Table 20: List of ABMs for handling stress in day-old chicks

ABM (animal categories)	Definition and interpretation of the ABM
Orientation and posture on the conveyor belts	**Definition:** Orientation of the head (forward, backward, sideways) and retaining this position on the belt (Giersberg et al., 2020).
	Measurement: Measuring the orientation of the head and measuring the body posture (standing, sitting, lying) before and after the procedure (e.g. dropping) (Giersberg et al., 2020).
	Sensitivity is high as with handling stress (due to increased drop height and belt speed) changes in orientation and posture are more frequent (Giersberg et al., 2020).
	Specificity is low as when handling stress is not present other factors than the hatchery procedures may cause the chicks to change their orientation and posture.
Falling on the floor	**Definition**: chicks falling from different heights (from conveyor belts, boxes) directly on the floor. Chicks that fall on the floor may get injured and suffer from pain and fear. If they stay on the floor, they might be suffering additional welfare consequences such as cold stress, and may be accidently been walk over by personnel or machine and suffer from more severe injury and even die.
	Measurement: the number of chicks falling on the floor per time unit is counted in 'at risk' areas (manual or mechanical handling of the chicks).
	Sensitivity is low since chicks may suffer from handling stress without falling on the floor.
	Specificity is high, since in case of the absence of handling stress, no chicks will fall on the floor for other reasons.
Distress calls	**Definition:** A change in frequency (Ginovart-Panisello et al., 2020) as well as the acoustical spectrum of calls that indicate stress (Herborn et al., 2020). These include, single or repeated short and loud shrieking (screaming) at high frequencies (Manteuffel et al., 2004). Distress calls are a response to a range of environmental stressors and elicit food calling and brooding from hens (Herborn et al., 2020).
	Measurement: Vocalisations can be recorded to assess the level of distress call or other sounds in the barn. Ideally this should be analysed using specialised software (Herborn et al., 2020). Different parameters can be used such as the number of call bouts per time unit, the percentage of animals performing calls, or more easily the intensity of the sound level represented by decibels, frequencies and amplitude.

ABM (animal categories)	Definition and interpretation of the ABM
	Sensitivity is moderate since chicks experiencing handling stress may not always show distress calls.
	Specificity is moderate since even with no handling stress, the ABM maybe still present since chicks may perform distress calls due to other welfare consequence (e.g. cold stress).
Escape attempts	**Definition:** Attempts to move, run or fly away, including wing flapping, from a fear-provoking stimulus (Graml et al., 2008).
	Measurement: Observation of the proportion of chickens showing this behaviour in a representative sample of chickens.
	Sensitivity is moderate, as chicks will not always express escape attempts when experiencing handling stress.
	Specificity is moderate, as even in the absence of handling stress, escape attempts might not be fully absent and may occur due to other fear-provoking stimuli than handling, such as sudden loud noises.

ABMs for handling stress such as 'posture and orientation on conveyor belt', 'falling on the floor' 'distress calls', 'escape attempts' and are highly to moderately sensitive and specific for assessment while chicks are put in the boxes, either automatically or manually. When boxes are loaded into the vehicle, it will be difficult to assess the impact on chick welfare with these ABMs, since animals are hardly visible.

4.7.1.3. Hazards

Rough handling

The main hazard is rough handling of day-old chicks automatically or manually during catching, putting in the box, loading and uncrating. This hazard includes 'changes in velocity, drop height acceleration and speed of conveyor belts', 'poor design of components of the conveyor belts system' and 'rough manual handling'.

Change in velocity (e.g. falling from one belt on another belt with a different speed, or in the crate): A difference greater than 0.4 m/s between belts was identified as a risk for the welfare of chicks (Knowles et al., 2004). A drop height above 280 mm when switching from belt or when dropping in a crate (Giersberg et al., 2021) and acceleration and speed of belts (Knowles et al., 2004; Giersberg et al., 2021), with a speed of 27 m/min (Giersberg et al., 2021) were indicated as a risk factor for chick welfare. Poor design of the system components (where chickens become caught, trapped, smothered or crushed) (Knowles et al., 2004) are also recognised as at risk for chick welfare, as they can get hurt and injured. Manual rough handling of the chicks when placing them in crate is also a risk factor for their welfare.

The hazard origins are staff and equipment, originating from errors in operation, poor design and maintenance of handling equipment, conveyor belts going too fast or badly synchronised, leading to handling stress.

4.7.1.4. Preventive measures

With on-farm hatching, chickens are not subjected to handling processes (Van de Ven et al., 2009; de Jong et al., 2019, 2020; Souza da Silva et al., 2021; Jessen et al., 2021a) and therefore it is the best preventive way where transport will be avoided.

When day-old chicks have to be transported, handling stress cannot be prevented since catching, loading and unloading are necessary procedures when moving day-old chicks from hatchery to farm. Only mitigative measures can be applied.

4.7.1.5. Corrective and mitigative measures

Handling stress can be mitigated by training the staff to manipulate the chickens and the boxes containing the chickens with care.

Constant monitoring of crating and loading of boxes into the vehicle is advised to prevent day-old chicks from falling off the belts. Proper design and maintenance of the systems will ensure that chickens will not be trapped or caught in any part of the system. If birds fall on the floor, special

procedures should be available to indicate how to pick them up with care in order to mitigate negative welfare consequences. It is recommended to pick up day-old chicks on the floor with both hands supporting the whole body.

It is recommended to synchronise or reduce belt speed and the height between them. Belt speeds higher than 27 m/min and drop height between belts of more than 280 mm are a risk for chick welfare. The speed difference between belts should not be greater than 0.4 m/s and steep gradients of the belts should be removed.

4.7.2. Sensory overstimulation and motion stress

4.7.2.1. Description

Sensory overstimulation occurs when the chick experiences stress and/or negative affective states such as fear or discomfort due to visual, auditory or olfactory overstimulation by the physical environment. Developing chicken embryos sense photoperiodic, auditory and olfactory cues in their environment (Reed and Clark, 2011), so sensory overstimulation may occur both prior to hatching and after hatching. Sensory overstimulation and its relationship with motion stress have been described in Section 3.7.4.1.

Motion stress occurs when day-old chicks experience motion sickness, stress and/or fatigue due to the forces exerted as a result of acceleration, braking, stopping, cornering, gear changing, vibrations and uneven road surfaces during transport. During unloading with the forklift, chicks might experience acceleration, vibration and impact with the cages.

There is no published information about sensory overstimulation during handling of chicks. Therefore, we can use the knowledge available about sensory overstimulation on farm and assume that during handling before transport, day-old chicks will be submitted to light and noises that will expose them to sensory overstimulation.

This welfare consequence is regarded as a highly relevant in the loading stage. Prevalence is high, as sensory overstimulation and eventually motion stress is likely to affect all animals during crating and loading. Duration is normally brief, as this step is done quickly. But during the journey, sensory overstimulation and motion stress can last throughout the transportation period. Severity depends on the speed and gentleness (or roughness) of displacement and on the level of noise/light around the chicken boxes. There is no specific knowledge about impact of noise or other sensory overstimulation and motion stress during transport stages of day-old chicks.

Sensory overstimulation is present during all stages of transport from loading to uncrating and motion stress during journey duration.

4.7.2.2. ABMs

Fear due to sensory overstimulation can be measured by fear tests such as the tonic immobility test (Jones, 1986), whereas stress responses (e.g. noradrenalin, corticosterone) can be measured in blood, feathers or faecal samples (Weimer et al., 2018). However, none of these measures are feasible under commercial conditions.

Therefore, distress calls and escape attempts are ABMs that can be used to monitor sensory overstimulation and motion stress (see Section 4.7.1.2). But it has to be noted that sensitivity is low for both these ABMs. Indeed, they might not be present even in case of sensory overstimulation/motion stress and in case this welfare consequence is not present, the ABMs might be there, caused by other welfare consequences (e.g. handling stress).

4.7.2.3. Hazards

Sensory overstimulation can be due to sudden or unexpected loud noise or too bright light during placement in the transport boxes and loading on lorries manually or with a forklift.

Unexpected loud sound/noise

Sudden or loud noises can induce fear in the day-old chicks and decrease coping capacities. Day-old chicks can be exposed during transport (loading, journey and unloading) to an environment with loud noises originating mainly from machines, the vehicle, box manipulation or movement, and sometimes from personnel shouting. There is no available data on noise levels and their relationship to the welfare consequences.

Visual stimuli

Bright lights and sudden changes in light should be avoided since they might provoke fear in animals. Chicks could be submitted to sudden changes in light when going from an area to another and from inside to outside with very high intensity natural light.

Acceleration

As in other domestic birds, it is not clear what impacts displacement (velocity and acceleration and deceleration) may have upon the vestibular systems of day-old chicks. Furthermore, exposure to acceleration, deceleration and sudden braking will induce postural instability, increase the requirement for muscle activity to restore stability and will increase the risk of impacts and injuries and fatigue (see chapter vibration in motion stress).

Vibration

Vibration during loading, unloading and transportation have been shown to alter animal behaviour and induce physiological changes as well as to cause effects at the cellular and molecular level (see chapter vibration in motion stress).

4.7.2.4. Preventive measures

Unexpected loud sound/noise

Excessive sound or noise should be avoided or minimised in all stages of animal production, handling and transport. The preventive measures will consist in staff education and training (i) to make them aware that noise at the birds' level should be avoided and (ii) to make them avoid shouting and making noise with the equipment and facilities, and to identify and eliminate noise sources. Regarding facilities and equipment, machines as well as the vehicle should be setup correctly to avoid excessive noise and facilities should be noise proofed.

Visual stimuli

Excessive visual overstimulation should be avoided or minimised in all stages of animal production, handling and transport. Strategies should be employed which reduce the risk of visual overloaded. Uniform lighting at low levels sufficient for safe handling will minimise dear in day-old chicks.

Acceleration and vibration

Fast acceleration or drops during manual or automatic crating (conveyor belt) should be avoided. During the journey drivers should drive carefully to avoid sudden movements.

4.7.2.5. Corrective and mitigative measures

During handling and transport, day-old chicks will suffer from a certain level of sensory overstimulation, but some measures can help in mitigating the welfare consequences for birds. There is no scientific published evidence about this, but in order not to overstimulate chicks, the following methods can be applied:

- Use electric handling machinery in order to decrease the sound level.
- Handle the chicks quietly, gently and calmly.
- Use a uniform and low level of light.
- Avoid changes in light levels.

4.7.3. Thermal stress

Newly hatched chickens, until day 5–8 of age, behave as poikilotherms, which means that they cannot regulate their body temperature by themselves (Nichelmann and Tzschentke, 2002). Appropriate effective temperature (Nielsen et al., 2011) is therefore required to keep the body temperature of chicks at the desired level of 40–40.5 °C (Mujahid and Furuse, 2009; Maman et al., 2019). Because of the special temperature requirements of chicks during the first week of life, maintaining them in their thermo-comfort zone (see Section 3.7.7.1) is critically important and challenging (HSUS, 2008). Thermal stress may be the most severe welfare consequence during the transportation of newly hatched chicks.

Vieira et al. (2016) compared the thermoregulatory responses of newly hatched chicks exposed to cold (21°C), heat (38°C) and 'thermoneutral' conditions (35°C) for 1 h in commercial transport

containers placed in an environmental chamber. Body weight, respiratory frequency, mean surface and cloacal temperature were monitored. The position of the box in the chamber had a negligible effect on the thermal comfort of birds. The mean surface and cloacal temperature were significantly reduced with the cold treatment. No significant effect was seen with higher temperatures. However, the authors suggested that with longer simulated journey durations the higher temperatures might also have affected the chicks' thermoregulatory responses.

Maman et al., 2019 exposed post-hatch chicks during 12 h to three different temperatures where humidity (53–56%) and temperature were controlled. Day-old chicks with high body temperature (42.6°C) lost a greater percentage of body weight due to dehydration and exhibited lower organ weights after the 12 h exposure period, which was followed by significantly poorer broiler performance. There were no significant performance differences in between the chicks in the control (40.0°C) and low (38.1°C) body temperature groups. Authors concluded that day-old chicks are more sensitive to higher body temperatures than to lower temperatures during the post-hatch handling period.

Some studies have attempted to define thermoneutral environments for neonatal chicks on the basis of metabolic heat production and body temperature responses (Misson, 1976; Henken et al., 1987; van der Hel et al., 1991). However, these studies did not measure indicators of homeostatic effort which are better to define the physiological effect of the thermal micro-environment. Xin and Harmon (1996) examined the effects of a range of temperature and humidity (20–35°C and 40–17%) upon day-old chicks by measuring metabolic rate and mortality. They concluded that optimum or thermoneutral conditions occurred between 30°C and 32°C.

Having not yet a developed a system to regulate their body temperature and with a low weight (9–200 g depending on species – from quail to a goose) day-old chicks can be subjected both to cold and heat stress depending mainly on external climatic conditions, ventilation and microclimatic conditions in the box. The prevalence of animals submitted to these welfare consequences can be from low (if chicks are kept in thermal-comfort zone) to high in case they are submitted (during waiting, loading/unloading or transportation) to either too cold or too hot effective temperatures. The duration can be short (e.g. opening of the doors during unloading of a part of the chick boxes in winter) or long (e.g. effective temperature too high or too low during the whole journey). The severity of thermal stress depends on the effective temperature and the duration of exposure, which can be low leading to discomfort until very severe, leading to death (DOA). For these reasons, cold and heat stress have been selected as the highly relevant welfare consequences for day-old chicks.

In the immediate post-hatch body temperature and metabolic rate of the chicks increase (Freeman and Manning, 1984). However body temperature remains labile during exposure to suboptimal thermal environments (van der Hel et al., 1991) and due to the chick immaturity of thermoregulatory homeostasis hypothermia or hyperthermia may arise. Therefore, an extreme consequence of thermal stress, combined or not with other welfare issues like delayed access to feed and water, can be mortality. Day-old chicks' mortality during transport can be monitored through DOA (Mitchell and Kettlewell, 2004a,b) described in Section 4.9.

4.7.4. Heat stress

4.7.4.1. Description

Heat stress is when the chicken experiences stress and/or negative affective states such as discomfort and/or distress due to difficulties to maintain the body temperature in the thermal comfort zone when exposed to high effective temperatures. Body temperature of a day-old chick is normally between 40°C and 40.5°C (Mujahid and Furuse, 2009; Maman et al., 2019). Although, in some studies submitting chicks to so called 'thermoneutral zone' (35°C), Vieira et al. (2016) cloacal temperature of chick can reach 41.2°C.

Mitchell et al. (1996) have employed physiological stress modelling, measurement of metabolic rate to determine optimum transport thermal environments for newly hatched chicks. Chicks were placed in commercial transport containers in calorimeter chambers housed in controlled climate rooms. Temperatures of 20–35°C with relative humidity (RH) of 50–65% and durations of exposure from 3–12 h were investigated. The results showed an optimal temperature–humidity range of 24.5–25.0°C and 60–63% RH based on the observation of minimal changes in body temperature, basal metabolic rate, hydration state, electrolyte balance, body weight loss and plasma metabolite concentrations. It was highlighted that these conditions are very similar to those currently employed by breeders and producers. In commercial practice, the recommended temperature for chick transport is 24–26°C

(Meijerhof, 1997; Aviagen, 2018; Weeks and Nicol, 2000) and the breeder companies recommend to control humidity as well (75% at 24°C). The studies also demonstrated that, with a controlled thermal environment (30–32°C in the vicinity of the chicks), journey durations of 12 h may be undertaken with no detrimental effect due to cold stress. It is concluded that both productivity and welfare of newly hatched chicks in transit can be maintained by careful regulation of the temperature and water vapour density to these prescribed limits inside the transport boxes. The optimum temperature for chicks at normal stocking density in transport containers was reported to be 24–26°C with relative humidity in the range of 60–63% (Meijerhof, 1997; Mitchell, 2009). However, Xin and Harmon (1996) found that 30–32°C was optimal at humidities below 40%, while de Lange (2012) suggested that 32–35°C maintains chicks in their thermoneutral zone and, importantly, ensures that yolk sac protein reserves are conserved for the development of the immune and digestive systems. However, with the advent of new vehicle designs and operation, the thermal specifications may have to be adjusted from the parameters of temperature and humidity described above (Meijerhof, 1997; Mitchell, 2009) to match the new higher ventilation rates and improved internal air mixing.

Vieira et al. (2019) ran an experiment in climate chambers submitting day-old-chicks to cold and heat stress for different exposure time. In the heat treatment (38°C, 75% humidity) there was impact on respiratory rate (> 100/min), average cloacal temperature (44.7°C) and increase gene expression of HSP70, especially after the three first hours of exposure. Mortality also raises above 6% in this group.

Vieira et al. (2016) in another experiment used to monitor both cold and heat stress during 1-h duration: mean surface and cloacal temperature, respiration frequency, body weight and mortality. The most affected ABM was mean surface temperature, which was 35.1°C in thermoneutral conditions (35°C) and 38.1°C in heat condition (40°C). It has to be noted that cold stress is, in this experiment, 20°C below neutral condition and heat stress only 5°C more. The cloacal temperatures associated were, respectively, from 40.9 to 41.2°C in neutral condition and 42.4–42.6°C in heat condition. Respiratory frequency showed important increase in case of thermal stress, from 60 to 66 respiration/min to 225–252 in heat stress conditions, this is probably due to stress reaction, rather than mechanism of thermoregulation. It has to be noted that in this experiment a temperature of 35°C was considered as thermoneutral conditions.

4.7.4.2. ABMs

In case of heat stress, chicks can be seen with an increase of respiration frequency. Indeed, due to low capacity in thermal regulation, chicks do not really pant but rather show increase respiratory rate due to heat stress.

Vieira et al. (2016, 2019) used mean body surface temperature as an ABM of heat stress. Considering the different heat challenges in these experiment (40°C, 1 h in Vieira et al., 2016 and 38°C 0, 3 and 6 h in Vieira et al., 2019) we can propose that chicks start to experience heat stress when mean body surface temperature is above 38°C. Cloacal temperature was also used by these authors and TCZ was expected to correspond to a cloacal temperature of chicks below 41.2°C. In breeding guides of fast-growing chicken genotypes, the cloacal temperature above which heat stress starts is 41°C.

ABMs that are considered feasible for assessing heat stress in day-old chicks during transport are given in Table 21.

Table 21: List of ABMs for heat stress in day-old chicks

ABM (animal categories)	Definition and interpretation of the ABM
Respiration frequency	**Definition**: Domestic Birds: frequency of breathing. A frequency of breathing that increases above 100/min might be a sign of heath stress (Vieira et al., 2019). **Measurement:** Observation of the proportion of chickens showing this behaviour in a representative sample of chickens. **Sensitivity** is high, as chickens will show increased respiration frequency when exposed to heat stress. **Specificity** is moderate, as increase of respiration rate may occur due to other welfare consequences (e.g. handling stress)

ABM (animal categories)	Definition and interpretation of the ABM
Mean surface body temperature	**Definition:** Mean surface body temperature measured at the surface of the chicken (Vieira et al., 2016)
	Measurement: Measuring surface temperature using an infra-red thermometer on: wing, head, pad and back of each chick to calculate an average temperature per chick (equation for calculation is available in Vieira et al., 2016).
	Sensitivity is high. When chickens are too hot the mean surface temperature increases higher than 38°C.
	Specificity is high. When chickens are not too hot, they do not have a mean surface temperature above 38°C.
Cloacal temperature	**Definition:** Temperature measured in the cloaca of the chicken (Mujahid and Furuse, 2009). It is a proxy ABM of the core body temperature. A cloacal temperature higher than 41°C indicates that chickens are experiencing heat stress (Mujahid and Furuse, 2009; Maman et al., 2019)
	Measurement: Measuring cloacal temperature using a thermometer in a representative sample of chickens
	Specificity is high. When day-old chicks are too hot, the cloacal temperature increase. For older chickens that can regulate their body temperature, the measure has moderate sensitivity, as these birds may not experience a drop in cloacal temperature before prolonged exposure to cold stress.
	Sensitivity is high. When chickens are not too hot, they do not have a cloacal temperature above 41°C.

ABMs to assess heat stress such as 'respiration frequency', 'mean surface body temperature' and 'cloacal temperature' are sensitive and specific for assessment. Currently, none of these ABMs are measured during the journey. They are more feasible when the vehicle stops or at unloading. For the ABMs that can be measured visually, they can be assessed on the animals of the periphery of the boxes. In the future, video recording devices may be elaborated for constant monitoring.

4.7.4.3. Hazards

Too high effective temperature

Too high effective temperature can be due to too high environmental temperature and humidity, lack of ventilation, exposure to the sun. This can happen during loading, during the journey and at arrival when animals are left outdoor with no protection from the sun or ventilation (on a tarmac, a courtyard, etc.). It is considered too high temperature when it is above 35°C in the vicinity of the birds.

The effect of the climatic conditions, the addition of heat and water vapour and the ventilation rate and distribution have been extensively addressed in relation to the transport of broiler chickens at slaughter age (e.g. Hoxey et al., 1996; Kettlewell et al., 2000) but the corresponding characteristics of chick transporters have received less detailed study. Quinn and Baker (1997) assessed the ventilation characteristics of commercial chick vehicles, with a full-scale experimental determination of ventilation patterns and the prediction of same by Computational Fluid Dynamics (CFD). They concluded that the load of stacked chick boxes had a channelling effect upon the air flow through the load space with significant amounts of air by-passing the chick boxes and being re-circulated. The ventilation regime for air flow in the chick containers affected the temperature distributions with peak temperatures occurring in the front central boxes and cooler air by-passing the load. In addition, cooler air entered from beneath the vehicle in the fully loaded configuration and reduced the flow through the load as well as potentially introducing exhaust fumes into the load space.

4.7.4.4. Preventive measures

Controlled environment or air-conditioned vehicles can regulate or modify internal thermal conditions by appropriate heating or cooling. They have the major advantage that the internal environment may be controlled precisely regardless of external weather or thermal conditions and does not rely upon vehicle movement.

The temperature in the vicinity of chicks should be monitored and action (e.g. decreasing heating, provide ventilation, using air conditioning) should be taken in order that it does not exceed 35°C in the boxes. For example, this temperature in the boxes could be reached by setting the equipment at 25°C dry bulb, 60–63% humidity in classical trailer. But it has to be noted that settlement of temperature and humidity could be different in a modern vehicle, according to their ventilation characteristics.

During loading, arrival and unloading, when chicks are submitted to outdoor temperature in hot climatic conditions, they should be protected from direct sun and provided ventilation.

When loading in the vehicle, boxes should be spaced out, to allow air circulation and good ventilation.

In a survey in Belgium by Lambrecht et al. (2020) during warm weather, responsible personnel indicated that they reduce the number of chicks per crate from 90 to a maximum of 80, increasing the space per chick from 26.7 to 30.0 cm^2.

4.7.4.5. Corrective and mitigative measures

During journey, the temperature inside the boxes should not exceed 35°C. This can be achieved, e.g. providing more or better quality (high flow) ventilation or colder air conditioning. At arrival, boxes must be spaced out and chicks removed from boxes immediately.

4.7.5. Cold stress

4.7.5.1. Description

Cold stress is when the chicken experiences stress and/or negative affective states such as discomfort and/or distress due to difficulties to maintain the core temperature above 41°C when exposed too low temperatures. Cold stress is present during the stages of loading/unloading, journey and arrival. As the effective temperature goes down, the cold stress will increase and when chicks are outside their thermoneutral zone, they might become lethargic and even die.

Vieira et al. (2019) ran an experiment in climate chambers submitting day-old chicks to a cold stress treatment (20°C and 75% humidity) for different exposure times. There was no mortality, but animals showed hypothermia from 3 h onwards, with a cloacal temperature of 39.6°C and mean surface body temperature of 28°C and 27.5°C after 3 h or 6 h, respectively, exposed to the cold treatment.

Vieira et al. (2016) exposed day-old chicks to cold (temperature of 15°C), thermoneutral (temperature of 35°C) and heat (temperature of 40°C) in control chambers for one hour. Body weight, respiratory frequency, mean body surface and cloacal temperature and mortality were monitored. Mean surface (mean for infrared thermometer measure of four zones of the body) and cloacal temperature responses were markedly affected by cold treatment, suggesting a decrease in animal welfare. The mean surface body temperature was 35.1°C in thermoneutral conditions (temperature of 35°C) and 21.3°C in cold conditions (temperature of 15°C). The cloacal temperatures associated ranged from 40.9°C to 41.2°C in neutral condition and from 37.6°C to 37.8°C in the cold treatment. Respiratory frequency increased considerably from 60 to 66 respiration/min to 124–130 in cold stress (probably due to stress).

If the cloacal temperature of a newly hatched chick drops below 40°C, it will become inactive and lie down, which further accelerates the undercooling (Mujahid and Furuse, 2009). These chicks will not eat and drink, will be more susceptible to infections and have a higher first-week mortality.

Based on the limited available evidence, the AHAW Panel concluded that chicks will experience cold stress if their cloacal temperature drops below 40°C. The lower limit of the comfort zone of day-old chicks is estimated to be at a temperature of 30°C. If the effective temperature (at the level of the chicks) remains above this threshold, domestic birds are unlikely to experience cold stress during transport. If the effective temperature is below this threshold, the chicks will have a risk of experiencing cold stress.

Vieira et al. (2016, 2019) used mean body surface temperature as an ABM of heat stress. In both experiments the mean surface body temperature remain above 34.7°C when chicks are kept in the thermoneutral zone. Considering the different cold challenges in the experiments of Vieira et al (2016, 2019) (15°C, 1 h in Vieira et al., 2016 and 20°C 0, 3 and 6 h in Vieira, 2019) it can be proposed that the risk of cold stress starts when mean surface body temperature drops below 34°C.

4.7.5.2. ABMs

Chicks that are too cold show huddling behaviour or huddle under the brooders (on farm). The sound level they produce increases due to an increase in distress calls (Mujahid and Furuse, 2009; Herborn et al., 2020). Distress calls can be scored manually or by automated sound recording devices (Herborn et al., 2020). Lethargy (Mujahid and Furuse, 2009) can be scored by observation of chicken behaviour, either manually or by automated analysis of video recordings, but automated recording is not yet in place at hatchery. Further, cold stress can increase first-week mortality (Heier et al., 2002); (Deaton et al., 1996); (Renwick and Washburn, 1982)).

The ABMs presented in Table 22 can be assessed before and after the journey but not during the journey. Temperature inside of the box can be used as a proxy during the journey.

Table 22: List of ABMs for cold stress in day-old chicks

ABM (animal categories)	Definition and interpretation of the ABM
Huddling	**Definition:** Chickens are grouping together into tight groups to reduce heat loss, sitting closely alongside each other, often in 'clumps' with areas of empty space in between, which is distinct from normal 'loose grouping' that chickens show when resting (Welfare Quality, 2009).
	Measurement: Observation of the proportion of chickens showing huddling behaviour (Welfare Quality, 2009).
	Sensitivity is high. Chickens will clump together as a response to low environmental temperatures; persistent huddling indicates cold stress (Welfare Quality, 2009).
	Specificity is high. When the chickens are not experiencing cold stress, they may also group but these groups are looser with empty spaces in between and differs in that sense from huddling behaviour (Welfare Quality, 2009).
Distress calls	See detail under Section 4.7.1
Mean surface body temperature	**Definition:** Surface body temperature measured at the surface of the chicken (Vieira et al, 2016)
	Measurement: Measuring surface temperature using a infra-red thermometer on: wing, head, pad and back of each chick to calculate an average temperature per chick (equation for calculation is available in Vieira et al, 2016).
	Specificity is high. When chickens are too cold the mean surface temperature is below 34°C.
	Sensitivity is highly. If the mean surface temperature is at or above 34°C, chicks are not too cold.
Cloacal temperature	**Definition:** Core body temperature measured in the cloaca of the chicken (Mujahid and Furuse, 2009). A body temperature lower than 40°C indicates that chickens are too cold (Mujahid and Furuse, 2009); (Maman et al., 2019))
	Measurement: Measuring cloaca temperature using a thermometer in a representative sample of chicks.
	Specificity is high. When day-old chicks are too cold, the cloacal temperature decreases. For older chickens that can regulate their body temperature, the measure has moderate sensitivity, as these birds may not experience a drop in cloacal temperature before prolonged exposure to cold stress.
	Sensitivity is high. When chickens are not too cold, they do not have a cloacal temperature below 40°C.
Respiration frequency	**Definition**: Breathing with increased frequency
	Measurement: Observation of the proportion of chickens showing this behaviour in a representative sample of chickens.
	Sensitivity is high, as chickens will show high respiration frequency when exposed to cold stress.

ABM (animal categories)	Definition and interpretation of the ABM
	Specificity is moderate, as in the absence of cold stress, increase of respiration rate may occur for other reasons
Lethargy	**Definition:** Sitting motionless with head drooped or standing with eyes closed, not responding to any stimuli (Mujahid and Furuse, 2009). If chickens are lethargic they do not move to feed and water, leading to dehydration and starvation which eventually leads to more lethargy and even mortality (Heier et al., 2002).
	Measurement: Observation of the proportion of chickens showing this behaviour.
	Sensitivity is high. If the welfare consequence is severe, the animals are likely to become lethargic
	Specificity is moderate. Chicks may become lethargic due to other welfare consequences (e.g. prolonged hunger and thirst).

The most sensitive and specific ABMs to measure cold stress in day-old chicks are huddling, distress calls, mean surface temperature, cloacal temperature, respiration frequency and lethargy.

4.7.5.3. Hazards

Too low effective temperature

The temperature as perceived by the chick, which is a combination of environmental temperature, ventilation, radiation and humidity in the in the vicinity of day-old chick is the main hazard responsible for cold stress (Maman et al., 2019; Viera et al., 2019). It is considered too low effective temperature when dry-bulb temperature is below 30°.

During loading (at the hatchery) and unloading (at the farm), day-old chicks can suffer cold stress if they are submitted to too low effective temperatures. This can be the case if the loading is taking place or takes place, even briefly, in areas where temperature is not controlled (e.g. outside). Then cold or/and rain/high humidity and/or wind/rapid air movements might place day-old chicks in too cold environments. Effective temperature might be too low because of chicks taken outside, doors opening, or if the air speed is too high.

During journey, chicks might be suffering cold stress if the temperature inside the vehicle is not maintained in their TCZ.

In addition, chicks could get cold during loading when they were not properly dried after spray vaccination, but no scientific literature on this has been reported (Lambrecht et al., 2020). In this hypothesis, chicks will then get wet and get cold in case they are submitted to high air speed and/or low temperature.

4.7.5.4. Preventive measures

At loading, ensure proper drying of the chicks before crating. Provide heat in the loading area and ensure continuity of suitable climatic conditions until chick boxes are placed in the vehicle.

During the journey, temperature, humidity and ventilation should be actively monitored and controlled to maintain the temperature of day-old chicks at 40°C. Heating and ventilation systems should be adjusted to ensure an effective temperature of above 30°C. This can be provided with a dry-bulb temperature of 24.5–25°C at 60–63% relative humidity, settled in the vehicle during the journey (FAWC opinion, 2019). Doors should be closed as soon as possible and remain shut. If they have to be opened, it should be for a brief period or in a temperate area.

At arrival, same climatic conditions should be ensured. Vehicles may park in protected areas, with the engine still on to provide temperature regulation.

During unloading, the vehicle should get as close as possible to the barn, unloading should take place in a protected area, that will ideally have the same climatic conditions as in the vehicle. Day-old chicks should be unloaded as soon as possible, avoiding windy areas and the coldest hours of the day if animals have to be transported outside.

4.7.5.5. Corrective and mitigative measures

Effective temperature in the vicinity of chicks should be monitored, and when the temperature is below 30°C, chicken should be moved to a warmer place.

4.7.6. Prolonged hunger and thirst

4.7.6.1. Description

In the other chapters of this opinion prolonged hunger and prolonged thirst are discussed separately since they might have different hazards. In day-old chicks, the unique hazard is the delay access to both feed and water, that are always provided together (hydrogel or feed + water). For this reason, both welfare consequences are presented together.

Prolonged hunger is defined as craving or urgent need for food or a specific nutrient, accompanied by a negative affective state, and eventually leading to a weakened condition as metabolic requirements are not met, resulting in loss of body weight due to starvation (see definition) and finally to death. Prolonged thirst is defined as craving or urgent need for water, accompanied by an uneasy sensation (a negative affective state) and eventually leading to dehydration as metabolic requirements are not met. Prolonged hunger and thirst are potentially present for all stages of transport from loading to uncrating.

Day-old chicks hatched in a hatchery do not have immediate access to feed. Early hatched chicks are at risk of prolonged hunger and thirst (Van de Ven et al., 2011) as they need to stay in the hatcher until the remaining chicks hatch. Chicks hatch over a period of 24–48 h (Careghi et al., 2005; Willemsen et al., 2010; Bergoug et al., 2013). Chicks are then removed from the incubator, processed (e.g. selection, vaccination), subjected to handling and transportation before they can access feed and water after placement on the farm. This post-hatch holding period may last on average 48 h (between 36–60 h and up till 72 h in case of long-term transport) (Careghi et al., 2005; Willemsen et al., 2010; Bergoug et al., 2013). This can result in increased mortality and impaired performance (de Jong et al., 2017). Effects on organ development and physiological indicators appear to be mainly short-term (de Jong et al., 2017).

Old studies (MacLeod, 1980; Freeman and Manning, 1984) proposed that yolk stores in the newly hatched chick constitute 18% of total body weight and contain approximately 2 g of lipid and 2.5 mL of water. In the absence of excessive thermoregulatory demands (see heat stress Section 4.7.4), this represents energy (75–80 kJ) and water supplies sufficient for 3 days without further provision of food and water. More recent studies have proposed that in modern newly hatched chicks high metabolic rate and rapid utilisation of resources in the first 24 h post-hatch coupled to delays in transit and placement result in poorer performance and health status throughout flock life (Hackl and Kaleta, 1997; Tanaka and Xin, 1997; Xin and Lee, 1997; Vieira and Moran, 1999; Bigot et al., 2001). Major causes of in transit and post-transport mortality and morbidity are dehydration and undernutrition (Xin and Lee, 1997).

Chicks are believed to survive 72 h after hatching without feed or water by using yolk sac reserves. However, it was assumed in EFSA AHAW Panel (2011) that modern genetic lines may deplete their reserves more quickly, due to the higher metabolic rates associated with faster growth. A higher metabolic rate during incubation will lead to a lower residual yolk weight and energy reserve for the hatchling that might affect post hatch development and performance. Van der Wagt et al. (2020) concluded that residual yolk weight and the total solid amount of the residual yolk at hatch have been decreased in the recent decades. Although the genetic progress and improved management and incubation conditions, effects on yolk utilisation efficiency and embryonic metabolic heat production are in fact limited. Egg size and incubation temperature have a major effect on residual yolk weight at hatch.

A delay of 48–72 h to feed and water access resulted in higher residual yolk weights at 96 h after hatch than in immediately fed chickens (Noy et al., 1996; El-Husseiny et al., 2008). The higher yolk utilisation of immediately fed chickens in comparison to delayed-fed chickens might be related to a higher intestinal activity, probably due to peristaltic movements. On the contrary, other studies did not find differences in yolk utilisation or residual yolk weights between immediate and delayed post-hatch feed intake up to 72 h (Gonzales et al., 2003; Van den Brand et al., 2010). Furthermore, De Jong et al. (2016), reported that 48 h (36–60 h) post-hatch food and water deprivation leads to lower body weights and higher total mortality in chickens up to 6 weeks of age. Van de Ven et al. (2013) concluded that early feed and water access enables early hatching chicks to compensate for their apparent disadvantage in development at hatching, whereas chicks subjected to fasting show metabolic adaptations to preserve nutrients.

Discrepancies in the results of different papers about the effect of delayed access to feed and water on chicks can be due to differences in incubation temperature (Ipek et al., 2014), brooding

temperature (Akşit et al., 2010) or breeder age (Van der Pol et al., 2013), but much is unknown about how these factors affect post-hatch yolk utilisation.

There is also limited knowledge about whether the delayed development of post-hatch feed and water deprived chicks has any on long-term welfare consequences (including health, production and development) compared to early fed chicks.

Bergoug et al. (2013) studied the effect of transport duration on body weight. Chicks not transported had a significantly higher body weight than chicks transported for 4 and 10 h during the following 21 days of age.

Besides the fact that long-term effects were not often demonstrated, short-term effects are more regularly present, but the exact nature of welfare impact on chicks is unknown. To be on the safe side of chick's welfare, we assume that delayed access to feed and water will lead to prolonged hunger and thirst, despite the fact that the yolk sac reserves exist and that animals have never 'eaten' nor 'drunk' before. Most duration of delay in access to water and feed that give temporary effects are between 36 and 48 h (up to 60 h). Shorter transportation duration (e.g. 14 h) also affects, but in this case, it is a combination of both feed/water deprivation and transportation duration, so it is difficult to conclude on feed deprivation *per se*.

All broiler chicks hatched in a hatchery without a system providing direct access to feed and water will experience prolonged hunger and thirst to some degree. Some of them will experience prolonged hunger/thirst for a duration that has been shown to impose negative effects on welfare (de Jong et al., 2017). Prevalence is high since all chicks will be submitted to it and the severity of hunger and thirst will depend on the time between hatching and access to feed and water. That is why this welfare consequence has been retained as highly relevant for transported chicks.

4.7.6.2. ABMs

ABMs that are considered feasible for assessing hunger in day-old chicks are given in Table 23. Reduced body weight of day-old chicks upon arrival at the farm may indicate prolonged hunger and thirst (de Jong et al., 2017). In addition, increased first-week and total mortality may be related to prolonged hunger and thirst post-hatch (de Jong et al., 2017).

A positive relationship between time without feed and weight loss was found, where chicks showed increased relative growth up to 7 days when they had immediate access to feed compared to those with delayed access (Careghi et al., 2005). Conversely, early hatched chicks (fasted at least 7 h longer) were heavier than late-hatched chicks from day 1 up until day 9 of age, possibly due to late hatchers being of lower quality and then showing lower performances (Bergoug et al., 2015).

Table 23: List of ABMs for prolonged hunger

ABMs	Definition and interpretation of the ABM
Body weight loss	**Definition:** A reduction in body weight. Chickens that have delayed access to feed and water have a reduced body weight upon placement on the farm, (de Jong et al., 2017).
	Measurement: The difference between chicken body weight measured using a weighing scale immediate post-hatch and upon placement.
	Specificity is moderate. Prolonged hunger and thirst due to feed and water deprivation will lead to first exhaustion of the yolk reserve followed by a loss of body weight.
	Sensitivity is low. Even in the absence of prolonged hunger and thirst, body weight loss can occur for various reasons, e.g. chicks are exposed to heat stress.

It has to be noted that behaviour trials to assess hunger and thirst have not been studied per se during deprivation of feed and water for day-old chicks, but it is rather the impact of delayed access to feed and water on animal health and performances that have been assessed. As in most of the studies, this delay has negative impact, at least in early health and performances of animals. Although the effect on the affective state of the chicks has not been assessed, it has been assumed that prolonged hunger and thirst and their physiological implications are present and have negative consequences on day-old chicks.

Newly hatched male grandparent breeder chicks in commercial containers (80 chicks per container) were exposed for 50 h to different temperature fluctuation regimens in the TCZ (Xin, 1997). Chicks with feed and Aqua-Gel supply had significantly lower ($p < 0.001$) body weight loss ($2.7 \pm 0.7\%$) than the chicks without feed and Aqua-Jel ($15.0 \pm 0.5\%$).

4.7.6.3. Hazards

Too long feed and water deprivation

Newly hatched chicks may be held for more than 48 h prior to road transportation. The chicks will therefore experience long periods of fasting in commercial hatcheries. There are few publications describing in detail the physiological status of chicks in such situations. Tong et al. (2015) measured a range of stress responses and other biomarkers and observed that chicks may experience significant levels of stress during the holding period. Therefore, as newly hatched chicks are often subjected to other concurrent stressors (heat, dehydration, dust and deprivation of feed) in the first 24 h after hatching, extended holding may exacerbate the associated detrimental effects. As a consequence, the adverse effects during the critical early hours following hatch can result in an increased percentage of early mortality. It was concluded that shortening the hatch window and minimising the number of chicks that experience a long holding period before pulling may improve chick quality and physiological status, which may be due to unfavourable environmental conditions that include feed and water deprivation.

The following factors will delay chicks to access feed and water after hatching is a risk factor for prolonged hunger:

- Early hatching in combination with a long hatch window (Van de Ven et al., 2011),
- No access to feed post-hatch due to hatching in a hatchery, especially if this period exceeds 48 h (de Jong et al., 2017),

The latter might be caused by no provision of feed and water immediately after hatching, long post-hatch processing and/or long duration of day-old chick transport.

Thus whilst, it has been considered that the utilisation rate of yolk sac substrates and reserves in newly hatched chicks from modern commercial broiler lines might impact upon acceptable journey times for newly hatched chicks. A review of all the available and reliable evidence (Van der Wagt et al., 2020; Aviagen, 2021) suggests that this may not be the case, and that journeys no longer than 24 h, without food and water are provided, are adequate to protect the chicks.

A positive relationship between time without feed and weight loss was found, where chicks showed increased relative growth up to 7 days when they had immediate access to feed compared to those with delayed access (Careghi et al., 2005). Conversely, early hatched chicks (fasted at least 7 h longer) were heavier than late-hatched chicks from day 1 up until day 9 of age, possibly due to late hatchers being of lower quality and then showing lower performances (Bergoug et al., 2015).

4.7.6.4. Preventive measures

Prolonged hunger and thirst could be prevented with the provision of feed and water in the hatchery immediately after hatching, which has been studied and commercially applied in some hatcheries (Lambrecht et al., 2020). When chicks were fed immediately after hatch, an increase in chick weight, yolk free body mass, heart weight, liver weight and intestine length was found compared to chicks that had not been fed until their arrival at the farm (Molenaar and Reijrink, 2011).

A study of Cardeal et al. (2021) suggested that the use of preplacement feed (and water) inside transport boxes is recommended for chicks from young broiler breeders that will be submitted to delayed feed access (in this experiment 48 h compared to 24 h) in order to improve gastrointestinal tract development, mainly that of the duodenum mucosa.

A suggested strategy to reduce metabolic depletion during extended transport is the exploitation of the reduction in metabolic rate in crated chicks in the dark (Tanaka and Xin, 1997; Mitchell, 2009). The quantity and rate of use of metabolic reserves by basal metabolism, however, is clearly not the only factor which will influence chick survival during transportation.

The negative impact of prolonged hunger and thirst post-hatch on the welfare of day-old chicks can be prevented by minimising the time between hatch and access to feed and water. This can be achieved by:

- Applying on-farm hatching with immediate access to feed and water (Van de Ven et al., 2009; de Jong et al., 2019, 2020; Souza da Silva et al., 2021; Jessen et al., 2021a).
- Shortening the duration of the hatch window (Bergoug et al., 2013) and minimising the number of chicks that experience a long holding period before pulling (Tong et al., 2015).
- Provision of nutritious gels in the hatchery (Van der Pol et al., 2015; Souza da Silva et al., 2021) or preplacement feed in the transport boxes (Cardeal et al., 2021).

- Minimising post-hatch handling and transport time to limit the risk of suffering from prolonged hunger and thirst (Careghi et al., 2005; Willemsen et al., 2010)

The literature presented above shows that the total time before access to feed and water of 48 h or more has detrimental effects on day-old chicks. Therefore, chicks should not wait for more than 48 h between the time they hatch and the time they access feed and water.

4.7.6.5. Corrective and mitigative measures

When chicks experience prolonged hunger and thirst, the only corrective/mitigative measure is to unload them as soon as possible and provide them with access to feed and water without delay.

4.8. Transport duration

Day-old chicks being of low weight and not fully able to regulate their temperature, the longer the journey duration, the more severe the impact of the welfare consequences e.g. prolonged hunger and thirst will be on the animals. Some research results show different negative impacts of length (in km) (Khosravinia, 2015) or duration of transportation on chicks' welfare and health (Bergoug et al., 2013). Other results were either inconclusive, particularly about long terms effects, or not showing any negative impact of long transport (Jacobs et al., 2016 comparing 11 h and 1 h30; Valros et al., 2008, 14 h vs. 4 h). But in all cases, the impact of transport duration is confounded with the impact of delayed access to feed and water (time before access).

Therefore, no recommendation can be made about maximum time of transportation, but rather about maximum time without access to feed and water. Regarding the arguments developed above, the maximum time between hatching and first access to feed and water (including time spent in the hatchery, holding time, loading, transport and unloading time) must not exceed 48 h. Therefore, the maximum time of transportation (including loading, journey, unloading) is equal to 48 h, counted from the first chicks to hatch until the last chick to access feed and water.

4.9. Iceberg indicators

Like in domestic birds (see Section 3.9.1 for details), DOA is also a relevant iceberg indicator for the assessment of chick welfare during transport (Mitchell and Kettlewell, 2004a,b).

To be a valid iceberg indicator, DOA should be collected and monitored for each individual transport. Day-old chicks might die during transport due to factors that are not attributable directly to the transport conditions. Therefore, the cause of DOA might not need to be investigated for each individual transport, but if it exceeds a certain level, that might be attributed to the transport conditions. In a study from Yerpes et al. (2021) in Spain, including a total of 66 journeys made in commercial conditions between May and November 2017, the mean chick mortality during transport was 0.055% (SD = 0.043). The literature about specific DOA of day-old chicks is very scarce, so from this recent paper, it can be assumed that DOA is not exceeding 0.1%.

5. Assessment of scenario 3: Road transport of rabbits

5.1. Introduction

Recent information on the production of rabbits in the EU has not been identified, but an overview report on the commercial farming of rabbits in the EU (European Commission, 2017d) provides relevant information based on MSs' replies to specific requests. The report states that there were around 180 million farmed rabbits reared for meat consumption in the EU in 2016. Around 119 million (66%) were kept in commercial farms and transported for slaughter. The other 61 million (34%) were reared, sold and consumed via back-yard farms' direct and local sales. These figures indicate that in the EU, rabbits are in the sixth position regarding the numbers of farmed animals killed for human consumption (after poultry, laying hens, trout, salmon and pigs) and eighth in terms of meat volume with 168,000 t (after ovine meat but before caprine meat) in 2016. Rabbit farming is mainly concentrated in three countries representing 83% of total EU production. Spain was the main producer with 48.5 million rabbits slaughtered in 2016, followed by France with 29 million and Italy, with an estimated 24.5 million. Five MSs (Hungary, Portugal, Germany, Belgium and Poland) produce the 14% and another 10 MSs the remaining 3%. Ten MSs do not farm and slaughter rabbits.

There are no reports on the numbers of rabbits transported within EU MSs, although such transport constitutes the majority of the journeys.

5.1.1. Means of transport between Member States as reported in TRACES

Transport of animals between MSs is registered in TRACES. Extractions from TRACES (Table 24) show that road transport constituted 99% of total transports of rabbits between MS reported in 2018 and 2019. Transport within MSs is very likely by road. Small numbers were reported for aeroplane and roll-on–roll-off transport.

Table 24: Number of rabbits* transported between EU MSs by different means of transport in 2018 and 2019

Means of transport	Year			
	2018		**2019**	
Aeroplane	435	0.02%	654	0.03%
Other	262	0.01%	638	0.03%
Road vehicle	2,095,608	99.86%	1,991,606	99.87%
Ship/Road vehicle	2,202	0.1%	1,313	0.07
Total	2,098,508	100%	1,994,211	100

*: In addition to rabbits kept for meat production, other categories of rabbits, such as laboratory animals and pets were reported (EFSA extraction from TRACES).

5.1.2. Journey times as reported in TRACES

Extractions from TRACES (Figure 27) show that 45% of the journeys reported had a duration of less than 4 h, and that 82% had a duration of less than 8 h. Eleven and seven per cent of the transports were between 8 and 12 h and longer than 12 h, respectively. This is based on estimated journey times (excluding loading and unloading) as reported in journey plans.
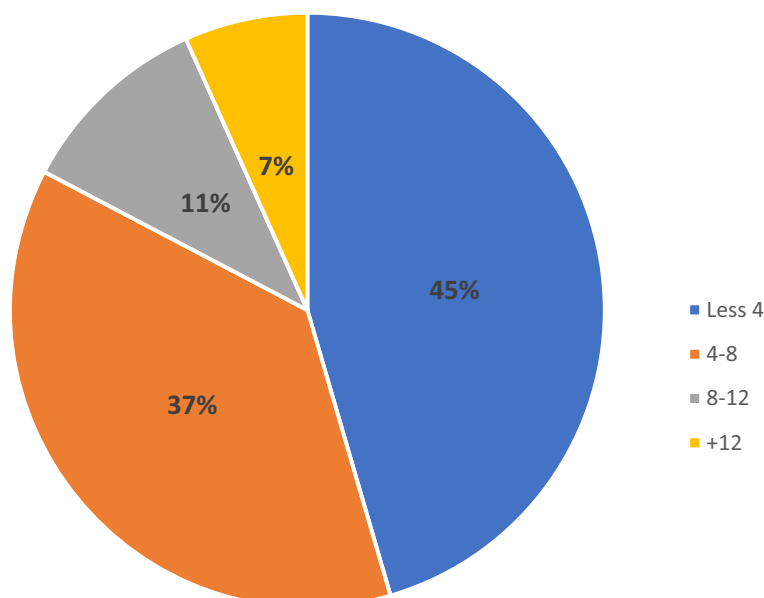
Figure 27: Distribution of journey times for rabbits transported between EU MSs (EFSA extraction from TRACES)

5.1.3. Stages of transport

Transport of rabbits as described in this scientific opinion consists of five stages:

1) *Preparation* includes planning of the journey and preparation of the animals by the removal of feed and assessment of fitness for transport.

2) *Loading* includes catching the animals, placing them in containers (crating) and loading of containers onto the vehicle.
3) *Journey* includes the movement of animals by vehicle and any intermediate stops (e.g. to change drivers) along the way until the place of destination is reached.
4) *Arrival* includes the period from arrival of the vehicle, unloading of the containers from the vehicle and waiting period up to the start of the uncrating. In the slaughterhouse, this stage includes the lairage period (on vehicle or in the lairage area).
5) *Uncrating* includes removing the animals from the containers.

5.2. Stage 1: Preparation of rabbits for transport

5.2.1. Description

Preparation for transport is not considered to be a part of transport, but is an important stage as it can prevent or mitigate the welfare consequences that rabbits might experience during transport, such as thermal stress, injuries and prolonged thirst and hunger. If rabbits are not correctly prepared for transport, this will increase the risk of them experiencing severe welfare consequences during the whole transportation process until uncrating. In this section, the practices relevant for this stage, namely the planning of the transport (loading, journey, arrival and uncrating), the removal of feed and assessment of fitness, are considered.

5.2.2. Planning of the transport

Transport of rabbits is usually planned in advance from the stage of preparation to arrival and unloading. This includes the coordination of the different stages of the transport (itinerary for loading and unloading, location of any driver resting places/stops during the journey), estimation of their duration and time of arrival. Effective planning includes communication with live rabbit catching teams and hauliers and with the slaughterhouse or destination farm for planning and coordinating the arrival of live rabbits.

Identification of the potential welfare consequences and their possible hazards (such as analysis of weather forecasts), as well as implementation of preventive and mitigating measures, are not always carried out in a structured way. Some companies have developed contingency plans with the purpose of helping the driver and the transport company to ensure the welfare of the animals in case of emergency.

5.2.3. Feed withdrawal

For rabbits transported to the slaughterhouse, the feed can be removed on the farm to allow time for the evacuation of the intestinal contents. It generally occurs 4–6 h prior to loading (NFACC, 2021), but on farm feed withdrawal is a less common practice for rabbits than for poultry (Verga et al., 2009). Water is offered until catching commences.

5.2.4. Fitness for transport

5.2.5. Introduction

Please refer to Section 3.2.4 for domestic birds.

5.2.6. Conditions leading to rabbits being unfit for transport

The principal conditions which will make rabbits unfit for transport are:

- Evident signs of illness.
- Emaciation and cachexia.
- Severe lameness: Unable to stand or walk more than a few steps.
- Female rabbits in the last third of gestation.
- Female rabbits and young up to 7 days after parturition.
- Un-weaned rabbits.
- Open wounds, prolapse and abscesses.
- Fractures (legs, etc.) and dislocations.
- Wet fur in low effective temperatures.

The list is not exhaustive, but is based on the limited published evidence and the expert opinion of the AHAW Panel members.

Evident signs of illness

Ill animals are at a high risk of compromised welfare during transport. Evident signs of illness include distressed breathing (breathing faster than usual or noisy breathing), discharge from the eyes or saliva around the mouth/chin, facial swelling, dull or missing fur, diarrhoea or unformed cecotropes (freshly soiled fur beneath the tail), neurological symptoms (e.g. head withdrawn towards body (hunched), and lethargy (Hart, 1988; Aubert, 1999; Millman, 2007).

Emaciation and cachexia

Emaciation refers to animals that are underweight compared with other animals in the group and which look thin. While a small amount of weight loss can be normal, when it is more severe, accompanied by muscle degradation, cachexia is a signal of a serious underlying medical condition. Emaciation is a serious, usually chronic and progressive condition characterised by significant (> 20%) body weight loss. Cachexia is the term used to describe the end stage of emaciation. Significant weight loss, associated with emaciation or cachexia, typically results from catabolism of body fat and protein in excess of caloric intake (Ford and Mazzaferro, 2012). Conficoni et al. (2020) found cachexia to be highly prevalent in condemned carcasses in the Italian slaughterhouse. In the period from 2003 until 2017, cachexia was the third most frequent finding recorded in young rabbits (29.7%) and the most frequent cause of carcass condemnation for breeding females (38.9%). Similarly, Tantiñá et al. (2000) reported cachexia as one of the main causes of condemnations of the inspected carcasses in Spain and in Poland (Drozd et al., 2019). Animals which would be rejected as unfit for human consumption should not be transported to slaughter.

Severe lameness

Any animal unable to stand is unfit to be transported. Rabbits which cannot move for more than a few steps are also unfit for transport as the underlying causes are likely to be painful. A hopping test proposed by Stauffacher (2000) can be used to score if a rabbit still has the ability of proper movement but this needs further research and development. The gait can be scored to identify problems with locomotion and lame rabbits if enough space is available. Gait scoring can be done by either taking the rabbit out of the cage and letting it run over a certain distance, or by gently pushing the rabbits in the cage with a hand and scoring if any lame rabbits are observed (if the cage is large enough) (de Jong et al., 2011). Gait score considers three possibilities: 'no problem' if the animal does not have any difficulty moving; 'moderate problem' if the animal has any difficulty in moving and 'severe problem' if the animal has several difficulties (no use of one leg or minimum weight-bearing) (Botelho et al., 2020). Gait score 3 (severe problem) represents severe lameness and leads to the animal being unfit for transport.

Female rabbits in the last third of gestation

Pregnant female rabbits shall not be transported near the time of parturition. This is to ensure that they are not put at risk of abortion or that parturition does not commence during transport. Rabbits are more likely to abort under the stress of transport during the last third of gestation (Swallow et al., 2005). Rabbits are particularly prone to fetal loss at 23 days when the fetuses are susceptible to dislodgement by rough handling (Harkness and Wagner, 1995). Therefore, as a precautionary principle, the AHAW Panel concluded that rabbits should not be transported during the last third of gestation. The typical period of rabbits is 30–32 days, and thus, they can be shipped up to 22 days into the pregnancy (Swallow et al., 2005).

Female rabbits with young till 7 days after parturition

Nursing rabbits with young can only be transported following an appropriate period after parturition (e.g. after a minimum of 7 days based on expert opinion). After this period and until weaning, they can be transported, but they will require additional care including adequate additional bedding and nesting material (Swallow et al., 2005).

Un-weaned rabbits

Rabbits should not be transported before weaning. Weaning is usually carried out between 28 and 35 days of age (Kovács et al., 2012). If early transport is necessary, they should be transported with

their mothers. Rabbits are born with no or only little hair cover and are deaf and blind. At birth, neonates are very sensitive to cold, but as their fur grows within a couple of days, they become less sensitive (Batchelor, 1999). Neonates should not be transported until their navels have healed (Swallow et al., 2005) and since neonates cannot maintain their body temperatures until they are 7 days old, they must be kept in a warm environment (Wolfensohn and Lloyd, 2003) and must not be transported.

Open wounds, prolapse and abscesses

The opinion of the AHAW Panel is that wounds that penetrate all skin layers, or which also damage the underlying tissue, are a cause for concern. Minor abrasions or scratches that are healed do not constitute a reason for declaring rabbits unfit for transport. More severe skin tears and lacerations involving all skin layers that are unhealed or such wounds that are infected and inflamed (e.g. abscesses) will result in pain and discomfort and should be regarded as reasons for exclusion from transport.

In rabbits, any rectal, vaginal or uterine prolapse is essentially similar to wounds comprising unprotected, protruding flesh that is potentially painful and susceptible to further injury and infection, thus rendering the animal unfit for transport.

Fractures (legs, etc.) and dislocations

While no data are available for rabbits, several studies have associated fractures and dislocations with an increased risk of DOA in poultry (e.g. Gregory and Wilkins, 1990; Warriss et al., 1992; Nijdam et al., 2004; Caffrey et al., 2017). These conditions would make any animal unfit for transport.

Wet fur in low effective temperature

Animals should not be allowed to become wet before and during transport if the effective temperatures are low and there is a risk of cold stress during transport. There is a shift in the lower limit of the thermo-neutral zone in wetted rabbits (Seltmann et al., 2009); thus, the wet rabbits will experience cold stress at temperatures still within thermo-neutral zone for dry rabbits (see Section 5.7.8 on cold stress). Not only wetness and ambient temperature but also interactions with body mass are involved in shaping an animal's thermal balance. Particularly younger animals with lower body mass are susceptible to cold stress when wet (Seltmann et al., 2009). In conclusion, wet integument only makes animals unfit to travel if they will be transported in cold weather without the application of preventive and corrective measures discussed in Section 5.7.8.

5.3. Stage 2: Loading of rabbits

5.3.1. Description

The loading stage includes preparation for catching, catching the rabbits, placing them in containers or crates (crating) and loading of containers onto the vehicle.

5.3.2. Types of containers used for transport of rabbits

Rabbits are transported in containers, also referred to as crates or cages. Types of containers vary globally, ranging from traditional loose chicken crates to purpose-built rabbit transport modules. In the EU, rabbits are usually placed in transport crates originally designed for poultry. Two different systems are established for the transport:

  a) Loose crate system: Stackable container systems transported to the vehicle in multi-floor crate stands. Crates can be of such a size that a person can lift one of them (a and b of Figure 28) or be modular units that need to be lifted with a forklift vehicle (c of Figure 28) (Verga et al., 2009).
  b) Modular units that need to be lifted with a forklift vehicle (c of Figure 28) (Verga et al., 2009).
  c) Fixed cages on the vehicle, whereby the rabbits are carried to the vehicle.

Standard rabbit crate dimensions (length × width × height) vary from 100–110 × 50–60 × 22–30 cm (Verga et al., 2009) to 74–78 × 5–58 × 22–30 cm. Buil et al. (2004), in a survey of Spanish rabbit abattoirs, found that container size differed widely among farms (ranging from 1,430 to 10,000 cm^2).
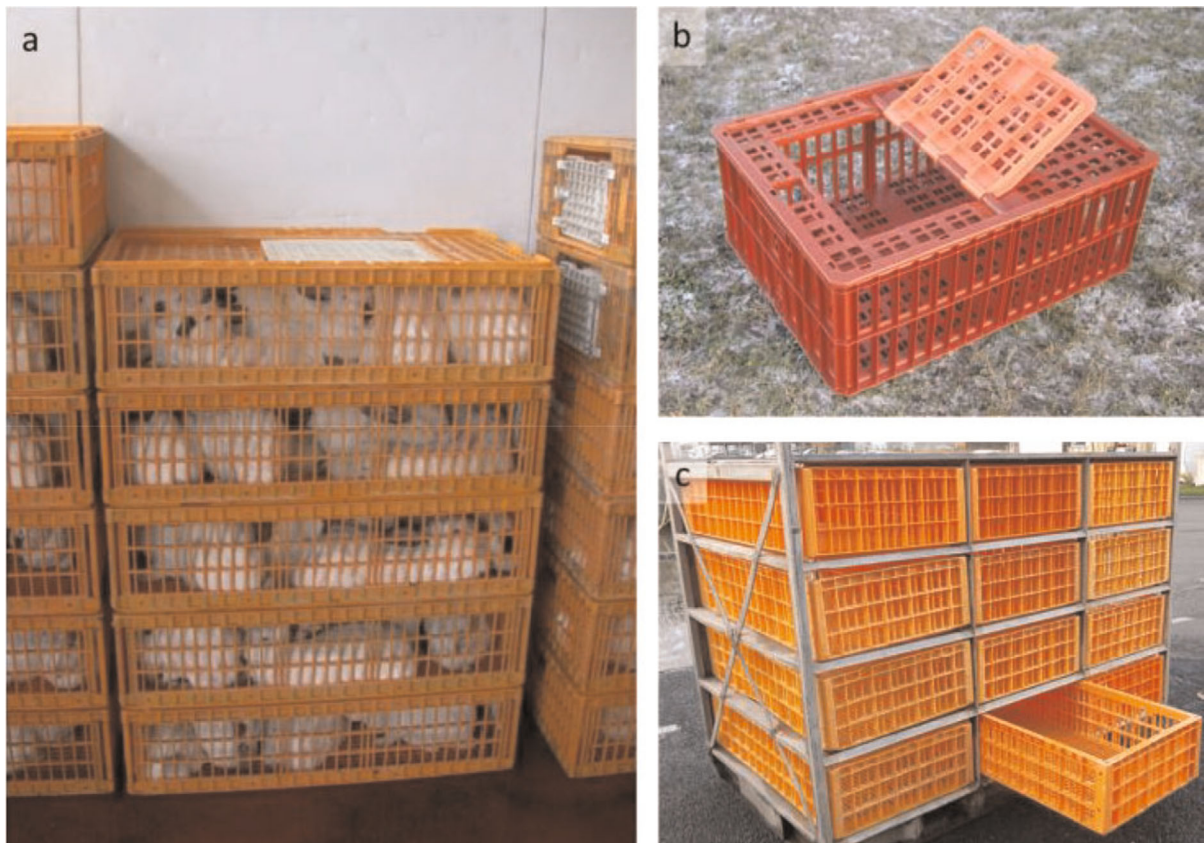
Figure 28: Common crates and containers used for the transport of rabbits: (a) Plastic crates with a sliding top and side opening, (b) plastic crate with top opening or top and side opening, depending on model, (c) modules with plastic drawers that slide (Source Credit Avipôle Formation) (EFSA AHAW Panel, 2020a)

5.3.3. Catching and crating

Rabbits are removed from the on-farm cages individually by holding and lifting by the neck skin (scruff) by one hand, with or without support of the body with the other hand (European Commission, 2017d). Once outside the cage, their body is supported with the other hand. Grasping rabbits by the hind legs or by the ears may also occur. Buil et al. (2004), in a survey of Spanish rabbit abattoirs, found that handling procedures differed widely among farms. Loading facilities were adequate but only a few hauliers had received specific training courses.

The EU transport regulation does not specify minimum space requirements for rabbits during transport. Current practice varies, apparently within a range from 130 to 160 cm^2/kg (Buil et al., 2004; De la Fuente et al., 2004b; Lambertini et al., 2006; Verga et al., 2009).

5.3.4. Loading of containers onto the vehicle

Once containers are filled, they are loaded either manually or mechanically by forklift onto the vehicle.

5.4. Stage 3: The journey by road of rabbits

5.4.1. Description

This stage includes the movement of animals by vehicle and intermediate stops along the way until the place of destination is reached.

The transport of rabbits from farms to slaughterhouses is carried out by authorised transporters by means of road transport. The vast majority of transport vehicles are open naturally ventilated vehicles. To protect the rabbits from adverse weather conditions most vehicles are fitted with side curtains that can be opened or closed depending on the wind and weather conditions.

Regarding the duration of transport to slaughterhouse, Buil et al. (2004) found that in Spain, in a sample of 21 hauliers, the mean time was 154 min (Range: 20–600 min), 14% of them lasted less than 30 min and 19% lasted more than 180 min. Most journeys (67%) were in the morning. At the abattoir, animals were unloaded almost immediately (the average time of the container to be unloaded was 4.5 min) (Buil et al., 2004).

In Europe, a common transport procedure is to load one vehicle with animals from different farms (Buil et al., 2004), before arriving at the slaughterhouse, hence not all rabbits are transported for the same amount of time. The logistics of loading commercial rabbits (including several stops) can greatly increase the journey time. The waiting time on farm can sometimes be longer than the transport itself (Buil et al., 2004).

5.5.   Stage 4: Arrival of rabbits

5.5.1.   Description

This stage includes the period from arrival of the vehicle, unloading of the containers from the vehicle and lairage period (on vehicle or in lairage area) up to the start of the uncrating.

Following arrival of the vehicle at the slaughterhouse, rabbits are either kept in lairage on the vehicle or unloaded and moved to designated areas. In commercial slaughterhouses, the containers are unloaded either manually or mechanically using forklifts and placed on top of each other in a protected area to await slaughter. Usually, this lairage area is placed outside, and very few slaughterhouses provide an enclosed area equipped with forced ventilation and water-misting sprays as a control strategy for adverse environmental conditions (Verga et al., 2009). The containers are arranged in spaced rows allowing human passage and observation of the rabbits. This arrangement also allows air to circulate between the containers.

Commercial practices vary from unloading of animals and moving them straight to the point of stunning without any time in the lairage to holding them in lairage for some time. Rabbits may wait for an indeterminate length of time in the containers at the slaughterhouse holding area (Buil et al., 2004). The study from Buil et al. (2004) in Spain indicated that 55% of the rabbits included in the survey were unloaded in the morning and the unloading procedure lasted on average 23 ± 15 min. Cages were mainly (85%) unloaded in groups or in cage stands. In most of the cases (80%), the unloading zone was covered and protected from the wind. Veterinary inspections took place at the unloading stage in all 21 slaughterhouses included in the study.

5.6.   Stage 5: Uncrating of rabbits

5.6.1.   Description

At the slaughterhouse, rabbits are removed individually from the cages by hand (Buil et al., 2004) for the purpose of restraining, stunning and slaughtering. At this stage of the process, rabbits that are dead on arrival (DOA) are removed.

5.7.   The highly relevant welfare consequences identified for transport of rabbits

As explained in Section 2.2.2, an exercise based on expert knowledge elicitation was performed to identify the highly relevant welfare consequences for transport of rabbits. Welfare consequences were not identified for the preparation stage, as they will mainly appear at later stages of transport. An overview of the results is presented in Table 25 below. The description and assessment of the welfare consequences are provided in this chapter.

Table 25: Welfare consequences identified as highly relevant for road transport of rabbits to slaughterhouses or production sites. (n.a. = non-applicable, x = highly relevant, − = not relevant)

Transport scenario 3: Road transport of rabbits to slaughterhouses or production sites					
Welfare consequence	*Preparation*	*Loading*	*Journey*	*Arrival*	*Uncrating*
Handling stress	*n.a.*	x	−	−	x
Bone lesions[1]	*n.a.*	x	−	x	x
Soft tissue lesions and integument damage[1]	n.a.	x	−	x	x
Restriction of movement	*n.a.*	x	x	x	x
Sensory overstimulation	*n.a.*	x	x	x	x
Motion stress	*n.a.*	x	x	x	−
Heat stress	*n.a.*	−	x	x	−
Cold stress	*n.a.*	−	x	x	−
Prolonged hunger	*n.a.*	−	x	x	−
Prolonged thirst	*n.a.*	−	x	x	−

The welfare consequences i. bone lesions and ii. soft tissue lesions and integument damage apply to all stages and will have the same preventive and corrective measures. For this reason, to avoid repetition of text, they have been presented in the same section under the common term 'Injuries' which will therefore referred to as from here onwards to indicate these two welfare consequences.

5.7.1. Handling stress

5.7.1.1. Description

As rabbits are a terrestrial prey species, being picked up is likely an innately stressful experience for them (McBride, 2017). The human–animal relationship plays a key role in handling commercial rabbits, due to their shyness towards man (Trocino and Xiccato, 2006). Rabbits that have been scared by humans, or have not been handled, try to flee and may injure themselves (Lidfors et al., 2007).

In response to a stressful event (Manteca, 1998), a fight or flight response occurs, during which catecholamines are released into the bloodstream (Kuchel, 1991), which makes the body temperature rise (Greco and Stabenfeldt, 1997). An increase in infrared temperature (measured in the eye, inner ear and outer ear) between the initial state without previous handling and after handling the animal was observed in other studies (Jaen-Tellez et al., 2020). Gascón and Verde (1987) observed increased corticosterone levels from 60 s after the start of handling. Finally, Olivas and Villagrá (2013) used rectal temperature to evaluate stress in rabbits.

Positive interactions with humans at an early stage in life could reduce their level of fearfulness. It has been shown that, in an apparent sensitive period, even minimal human contact is effective in reducing avoidance of the caretaker, and thus, handling might be a useful tool to reduce stress and improve welfare even under intensive farming conditions (Csatádi et al., 2005; Zucca et al., 2012). It has also been shown that handled rabbits approached humans significantly sooner than non-handled ones (Csatádi et al., 2007) and that frequent handling of young rabbits not only changes their behaviour by reducing fear of humans but also positively influences the growth rate and reduces the mortality rate (Jezierski and Konecka, 1996). Swennes et al. (2011) documented that regular handling during 3 weeks when rabbits were hand-carried resulted in habituation of rabbits to human contact and reduced fear. More compliant rabbits might be less likely to experience injury, particularly lumbar vertebral fractures, due to kicking while being carried or restrained. In contrast, rabbits that have not been handled by humans during the lactation period, do not become accustomed to handling in the fattening phase and stress occurs, as evidenced by body temperature variations (Jaen-Tellez et al., 2020).

The effects of the loading method on some stress indicators in commercial rabbits transported to the slaughterhouse were studied (Vignola et al., 2008). A total of 192 animals were transported on 100 min journeys to the slaughterhouse. Rabbits were loaded either in a smooth way (taken from the farm crates, placed in a wide trolley and carried gently into the transport cage) or in a rough way (rabbits from four crates were carried all together in the same trolley and loaded hurriedly).

Corticosterone significantly increased only when using the rough loading method. Rough rabbit handling has been reported to increase preslaughter mortality and main carcass defects, such as haemorrhages, bruises and broken bones (Verga et al., 2009). Mazzone et al. (2010) investigated the impact of handling during loading on rabbit meat quality. A total of 192 rabbits were loaded onto the vehicle smoothly (each subject carefully placed into the transport crate) and 192 rabbits were loaded roughly (loading was hurried and carelessly executed by the transport operator, throwing each animal into the crates fixed on the vehicle) and then transported (100 min, 12 animal per cage, 57.7 kg/m^2) to the slaughterhouse. Transport, independently of loading method, significantly increased neutrophilia, lymphocytopaenia, serum AST, alanine transaminase (ALT) and CK activities and serum corticosterone concentration. PCV after transport did not differ from values detected at the farm, in agreement with Liste et al. (2008), who did not find any variation of PCV in commercial rabbits transported to the slaughterhouse. No adverse effects of loading method on carcass traits and meat quality were highlighted. On the basis of these findings, Vignola et al. (2008) and Mazzone et al. (2010) concluded that the stress parameters analysed were more influenced by transport (including handling) rather than by the different loading methods or crate position in the vehicle. Liste et al. (2006) also found that different loading methods did not exert significant differences on the stress indicators. They concluded that, in order to have an effect on meat quality, the threshold for stress was probably higher than the threshold needed to have an effect on welfare indicators.

Considering the available evidence, the AHAW Panel concluded that handling stress is a welfare consequence inherent to transport, and all rabbits are likely to experience this welfare consequence, mainly during catching and crating of the loading stage and during the uncrating stage.

5.7.1.2. ABMs

ABMs that are considered feasible for assessing handling stress in rabbits during transport are given in Table 26.

Table 26: ABMs for handling stress in rabbits

ABM (animal categories)	Definition and interpretation of the ABM
Escape attempts (or startling behaviour)	**Definition**: Attempts to move or run away from the situation (Graml et al., 2008) (from EFSA AHAW Panel, 2020a). Pain and fear due to incorrect handling can be assessed through escape attempts and vocalisations (EFSA AHAW Panel, 2020a). **Measurement:** Number of escape attempts or number of rabbits showing escape behaviours **Sensitivity** is high since in case of handling stress, rabbits will try to escape. **Specificity** is moderate as the absence of handling stress does not mean that escape behaviour cannot be seen due to other welfare consequences (e.g. sensory overstimulation).
Piling up	**Definition:** Rabbits crowding against and on top of each other (from EFSA, 2020a). Escape behaviours during the catching process can lead to the animals accumulating in a corner of the cage or barn. **Measurement:** Number of animals or events. **Sensitivity** is moderate since not all the animals experiencing handling stress will show this behaviour. **Specificity** is moderate as the absence of handling stress does not prevent piling up due to other reasons.
Vocalisations	**Definition:** Single or repeated short and loud shrieking (screaming) at high frequencies (Manteuffel et al., 2004) as a measure of pain (from EFSA, 2020a). **Measurement:** Number of calls or proportion of rabbits vocalising. **Sensitivity** is moderate, as only a proportion of animals experiencing handling stress present vocalisation. **Specificity** is moderate because the absence of handling stress does not prevent vocalisation for other reasons.

The ABMs escape attempts, piling up and vocalisations are highly to moderately sensitive and specific to assess handling stress in a group of rabbits during catching and crating of the loading stage, during unloading at the arrival stage and during the uncrating stage. However, they are not feasible during the journey.

5.7.1.3. Hazards

Staff and equipment

Humans are the main source of handling stress since they are perceived as predators. Depending on the quantity and quality of previous human contact and handling (calm vs. rough), the level of handling stress will vary from low to very high.

Rough handling

Rough handling during catching, carrying and putting or removing rabbits from the containers causes pain and fear and distress. Catching a rabbit that is located away from the openings can be difficult. The operator may resort to catching and handling the animal roughly, by the head, ears, neck or legs, increasing the risk of pain, fear and distress. This hazard has staff as origin, attributed to lack of the appropriate skill sets needed to perform tasks or due to fatigue.

5.7.1.4. Preventive measures

Staff and equipment

Handling stress due to staff and equipment cannot be prevented, only mitigated since catching, crating and loading are necessary procedures when moving rabbits from the husbandry system to containers for transport.

Rough handling

Handling during catching, crating and uncrating should be performed smoothly. This can be achieved by a good training of the staff to acquire the knowledge and skills required to perform their allocated tasks efficiently, letting them know that animals are sentient beings that can suffer pain and fear and therefore should be treated correctly to avoid negative welfare consequences (EFSA AHAW Panel, 2020a,b).

5.7.1.5. Corrective and mitigative measures

Rough handling

Rabbits should be removed from the husbandry cages individually by holding and lifting by the neck (scruff) by one hand, with support of the body with the other hand (European Union, 2017d). Once outside the cages, their body should be supported with the other hand (EFSA AHAW Panel, 2020a,b) (Figure 29).
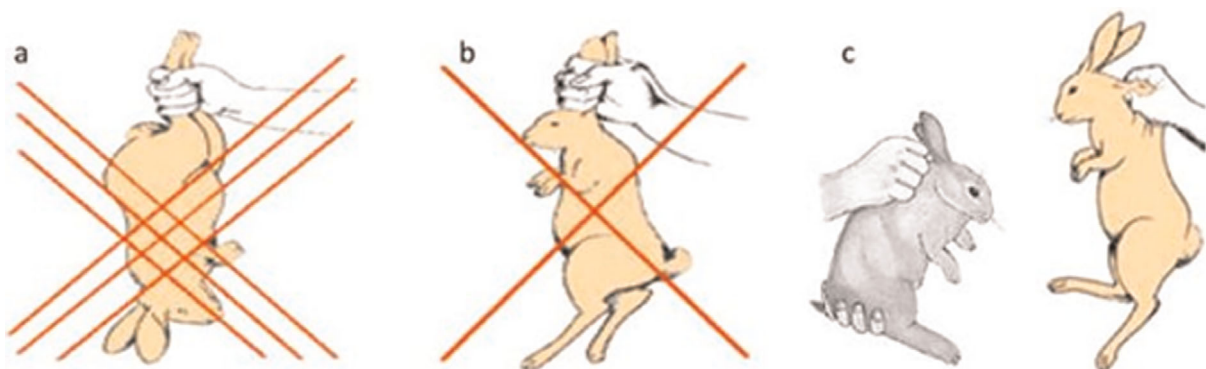
Figure 29: Poor practice of grasping rabbits by the hind legs (a) or by the ears (b), and good practice of grasping rabbits by the skin of the neck (c) (Source: Federation of French Poultry Industries)

Therefore, staff training and rotation, use of appropriate wide opening container are the most effective preventive measure. If the hazard still occurs, the line speed should be reduced.

EFSA (2005) recognised every practice producing a negative experience of rabbits towards humans as a hazard and recommended to adopt the following measures: a progressive approach to kits; quiet and slow movements during handling and catching; rabbits must never be picked up or held by ears; rabbits should be caught with a minimum of chasing. To improve human–rabbit relationship and to reduce fearful reaction, the regular daily handling of lactating kits recommended by EFSA (2005) has been confirmed to be useful by later studies (Csatádi et al., 2005; Verga et al., 2007; Verwer et al., 2009; Zucca et al., 2012). Indeed, kit exposure to only human smell reduces fear towards man and improves their welfare (Dúcs et al., 2009).

5.7.2. Injuries

As explained in Section 5.7, the term 'Injuries' refers to two welfare consequences which are here grouped together i. bone lesions (including fractures and dislocations) and ii. soft tissue lesions and integument damage.

5.7.2.1. Description

For a description of this welfare consequence, please refer to Sections 3.7.1 and 3.7.2.

The handling of rabbits during loading, arrival and uncrating from transport containers can be considered as risk factors in terms of injury or even death. Post-mortem examination records of a study carried out in the Czech Republic over a decade showed that among animals transported to slaughter in containers, the occurrence of traumatic injury was highest in laying hens (2.80%) and in rabbits (1.52%) while the overall incidence of trauma was below 0.5% in other species (Valkova et al., 2021). Traumatic injuries to the limbs occurred significantly more often than traumatic injuries to the trunk (Valkova et al., 2021).

This risk of injuries increases with rough handling, and when large numbers of animals are handled (Buil et al., 2004; Petracci et al., 2010) as staff caution decreases (Petracci et al., 2010). Rough handling leads to traumatic lesions such as bruises, abrasions, contusions and/or fractures (Verga et al., 2009). Injuries can be also caused by fighting due to mixing of older rabbits culled from breeding. However, this will not be a problem in fattening rabbits transported to slaughter while still prepubertal (De la Fuente et al., 2007).

5.7.2.2. ABMs

Measures have been validated for pain assessment in laboratory rabbits, based on changes in behaviour, facial expressions and body temperature (Leach et al., 2009; Farnworth et al., 2011; Keating et al., 2012). These could also be utilised for farmed rabbits. Behavioural changes include reduced feeding and drinking, tight huddle posture (sitting with their back arched and fore and hind limbs drawn in tightly), locomotory changes including shuffle (walking at a very slow pace) and partial hop movements (forward extension of forelimbs as if to hop, without movement of hind limbs) (Farnworth et al., 2011). General grooming is also reduced, although sites of injury may receive increased grooming (Farnworth et al., 2011). EFSA (2005) noted that although rabbits are normally silent animals, they may squeal loudly if in severe pain or distress; they may also grind the teeth in cases of more chronic pain. The use of facial expression as an indicator of pain has been validated in rabbits (Keating et al., 2012). The Rabbit Grimace Scale assesses five different facial action units (orbital tightening, cheek flattening, nose shape, whisker position and ear position) to create an overall score that increases when rabbits experience pain.

ABMs that are considered feasible for assessing injuries in rabbits during transport are given in Table 27. They include physical damages to the muscles and skin (e.g. scratches and open wounds, bruises). Traumatic injuries during post-mortem inspection have been considered (Grilli et al., 2015) to be an indicator for bad transport, loading/unloading and handling causing negative welfare consequences, without possibility to distinguish the phase(s) where the injuries appeared.

Table 27: ABMs for injuries in rabbits

ABM	Definition and interpretation of the ABM
Wounds	**Definition**: A skin lesion will be considered a scratch or open lesion bigger than 2 cm in any part of the animal (Botelho et al., 2020) occurring due to rough handling
	Measurement: Number and size of wounds per rabbit. Score: if the animal does not have any lesion bigger than 2 cm will be a score 0; the presence of one or more lesions bigger than 2 cm will be a score 1; and the presence of one or more lesions bigger than 5 cm will be a score 2 (Botelho et al., 2020).
	Sensitivity is moderate, as rabbits might be injured without having wounds.
	Specificity is high since non-injured rabbits will not have wounds.
Bruises	**Definition:** Bruising is an injury that occurs after trauma, it results from a haematoma and is often without rupture of the skin. The most frequently bruised areas on the rabbit are the thoracic region, legs and the inner loin. Unfortunately, the bruises become visible after the skin is removed only (Verga et al., 2009).
	Measurement: Number of rabbits showing bruises.
	Sensitivity is moderate, as rabbits might be injured without having bruises.
	Specificity is high since non-injured birds will not have bruises.

ABMs of injuries, such as wounds and bruises, are moderately sensitive and highly specific during catching and crating, loading and unloading of rabbits.

5.7.2.3. Hazards

Rough handling

Rough rabbit handling has been reported to increase preslaughter mortality and main carcass defects, such as haemorrhages, bruises and broken bones (Verga et al., 2009).

Narrow crate openings will increase the risk of injury when handling the animal. Similarly, catching a rabbit that is located away from the openings can be difficult. The operator may resort to catching the animal roughly, by the head, neck or legs, increasing the risk of injury. Animals colliding with the surfaces will increase the risk of injuries as well.

Rabbits placed on upper vehicle levels are often subjected to a greater number of falls than those on lower levels (EFSA AHAW Panel, 2011).

5.7.2.4. Preventive measures

Rough handling

Avoiding rough handling of rabbits will reduce the risk of injuries. Training of staff to acquire the knowledge and skills required to perform their allocated tasks efficiently, but without damaging the rabbits, is identified as the most important preventive measure. Proper supervision of the catchers can result also in improved handling and low levels of injuries.

Injuries due to hitting or pushing rabbits against the edges of the crate or container entrance can be prevented by using crates or containers with large doors or openings.

Containers (crates) should be placed as close as possible to the rabbits to minimise the distance rabbits are carried. This will also reduce the risk of staff fatigue. Furthermore, staff rotation is important to reduce fatigue, which can lead to poor rabbit handling practices.

Loading should be performed smoothly and in a horizontal position to prevent tilting of containers that causes rabbits to pile up or bunch.

Containers must be in good condition without broken plastic or metal parts protruding inwards, which might cause injuries and bruises.

5.7.2.5. Corrective and mitigative measures

There are no corrective measures once rabbits are injured. The welfare consequences of injured rabbits can be mitigated by identifying injured rabbits before the processes of catching and crating and prevent them from travelling.

5.7.3. Restriction of movement

5.7.3.1. Description

Restriction of movement is a welfare consequence inherent to the transport of rabbits, as they are confined in containers. During transport, most rabbits will experience the welfare consequence restriction of movement, as space allowance in all dimensions (floor space and height) is limited in transport containers. In addition to high prevalence, the welfare consequence is present from crating to emptying of the container. The severity depends on the space available for the animal to adapt natural postures, rest comfortably and move. Severity is regarded as high when the animal cannot adopt natural resting or sitting postures with the head and ears extended in a comfortable position, and moderate when it is unable to change posture within the container.

Under farming conditions, rabbits rest for 12–18 h a day and will likely have a need to do so during transport. Animals which are not able to adopt relaxed postures, or are forced to lie on inadequate or dirty surfaces, can suffer from physical discomfort (e.g. cold stress, lesions, pain) as well as an impaired affective state (EFSA AHAW Panel, 2020b). While resting, they adopt a crouched position (lying alert) (position 1 – Figure 31) or lying postures characterised by stretching of the front legs (position 2 – Figure 32) and of the hind legs and of the body, including full lateral lying on the side (Figure 36), that are thought to be associated with relaxation (EFSA, 2005). During transport, in order to avoid welfare consequences from restriction of movement, rabbits need to be able to rest in resting position 1. Applying the allometric equation, with a k-value of 270, rabbits will have more space than required to take position 1 for all animals, and therefore, they will also have space to move around and to change positions. There might also be a need to adapt a stretched out recumbent position to thermoregulate during heat stress, in which case more space would be required as described for position 3 (Figure 33) or 4 (Figure 34) (Giersberg et al., 2015). The welfare consequence will be severe if rabbits cannot sit at least simultaneously without overlapping. However, no scientific evidence is available to document whether rabbits adopt a fully stretched out position during transport. Further research is needed to ascertain the preferred posture of rabbits during transport under various thermal conditions.

Accorsi et al. (2017) reported that different space allowance in transport containers (307 cm^2/rabbit, 373 cm^2/rabbit and 475 cm^2/rabbit corresponding to 102–123 cm^2/kg, 124–149 cm^2/kg and 158–190 cm^2/kg, respectively) was associated with different levels of serum cortisol and glucose (measured at the time of slaughtering in all groups). A higher space allowance reduced cortisol and glucose levels that tended to increase with space reduction. They suggested not reducing the space allowance for rabbits of 2.6–2.7 kg below 350 cm^2/rabbit (approx. 130 cm^2/kg) for journeys lasting more than 2 h. De la Fuente et al. (2004a,b) did not find any difference in physiological parameters related to stress (cortisol, creatine kinase, lactate dehydrogenase, lactate, glucose, packed cell volume, osmolarity and albumin and globulin concentrations) when comparing rabbits averaging 1.85 $\pm$ 0.21 kg live weight and transported at low (0.05 m^2/animal; equivalent to a space allowance of ca. 279 cm^2/kg) or high stocking densities (0.03 m^2/animal; equivalent to a space allowance of ca. 186 cm^2/kg). The authors interpreted that the different space allowances were insufficient to have an effect on rabbit welfare. In contrast, Caucci et al. (2018) observed significantly increased DOAs at a stocking density higher than 29.3 kg/crate (< 196 cm^2/kg).

Evidence in literature on the effect of cage height on rabbit welfare is limited and concerns housing conditions as opposed to transport. Further research is needed to ascertain the position of the rabbits during transport. The size of rabbits of up to 2.5 kg in a sitting position with ears vertically extended is about 20 cm plus 10 cm (Figure 30). Between 4.5 kg and 6 kg, the size adopting the sitting position is 30 cm with 10 cm for the ears vertically extended (Michel, personal communication).

Figure 30: Rabbit in sitting position. Source: Shutterstock

5.7.3.2. ABMs

Recognising that some degree of restriction of movement is inevitable when transporting rabbits, the ABMs have been defined to indicate the minimum space allowance required to prevent severe welfare consequences regarding the restriction of movement.

'Sitting posture', 'lying posture' and 'ear position when sitting' are the suggested ABMs to assess restriction of movements in rabbits as they are sensitive and specific during crating and before loading for transport (Table 28).

Table 28: ABMs for restriction of movement in rabbits

ABM (animal categories)	Definition and interpretation of the ABM
Sitting posture	**Definition:** Rabbits are adopting a sitting position (Hocks in contact with the ground, front legs stretched and touching the floor; chest not touching the floor; Position 1) for resting without overlapping.
	Measurement: The body posture of rabbits can be visually inspected at loading, unloading and during stops. Nevertheless, it is difficult to assess the body posture of rabbits while they are inside the containers, as only the rabbits in the periphery of the outer containers can be assessed.
	Sensitivity is high. If movement is severely restricted, not all rabbits will sit in a natural position and some of them will overlap.
	Specificity is moderate as without severe restriction of movement, rabbits might prefer to be in a lying position.
Lying posture	**Definition:** Rabbits are adopting a lying posture in ventral recumbency (front legs extended and hind legs bent to the body; Position 2) without overlapping.
	Measurement: The body posture of rabbits can be visually inspected at loading, unloading and during stops. Nevertheless, it is difficult to assess the body posture of rabbits while they are inside the containers, as only the rabbits in the periphery of the outer containers can be assessed.
	Sensitivity is high. If movement is restricted, not all the rabbits will adopt lying posture and some of them will overlap.
	Specificity is moderate as even when there is enough space to rest, rabbits might prefer to be in a sitting position.
Ear position when sitting	**Definition:** The rabbits adopt a sitting position (hocks in contact with the ground, front legs stretched and touching the floor; chest not touching the floor) with ears upright without touching the ceiling of the container.

ABM (animal categories)	Definition and interpretation of the ABM
	Measurement: The position of rabbits can be visually inspected at loading, unloading and during stops.
	Sensitivity is high. If vertical movement is severely restricted, the rabbits in a sitting position will touch the ceiling of the container with the ears.
	Specificity is high. If vertical movement is not severely restricted, the rabbits will not touch the ceiling of the containers while adopting a sitting position**.**

5.7.3.3. Hazards

The principal hazards are containers with insufficient floor space allowance and insufficient height.

Insufficient floor space allowance

The space allowance is the space per rabbit in the container. The more it is reduced, the fewer the possibilities that rabbits have to sit or lie simultaneously and change position.

Giersberg et al. (2015) measured the floor space that a rabbit occupies due to its physical size and shape in one sitting and three recumbent positions, described below in Figures 31–34:
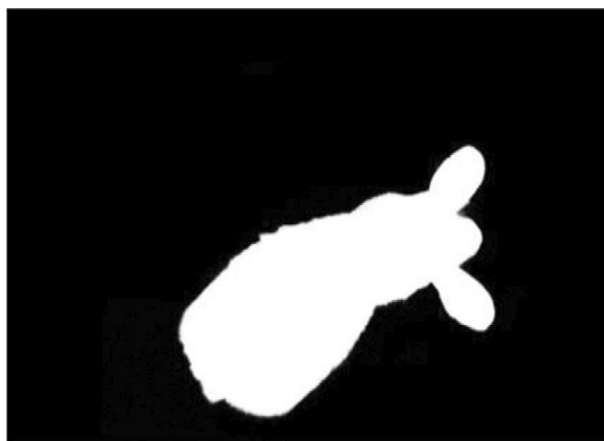
Figure 31: Position 1 (Sitting): Hocks in contact with the ground, front legs stretched and touching the floor; chest not touching the floor
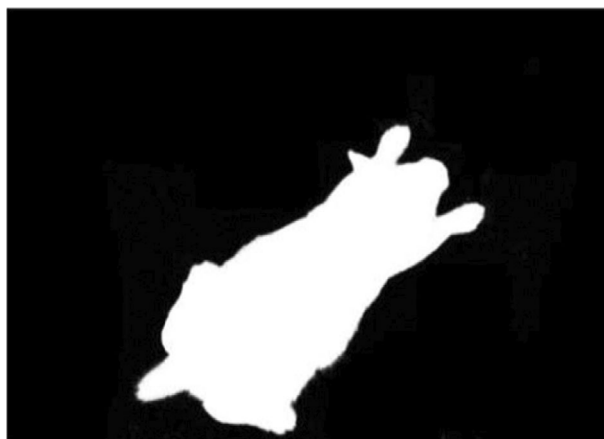
Figure 32: Position 2 (Recumbent): Ventral recumbency, front legs extended and hind legs bent to the body
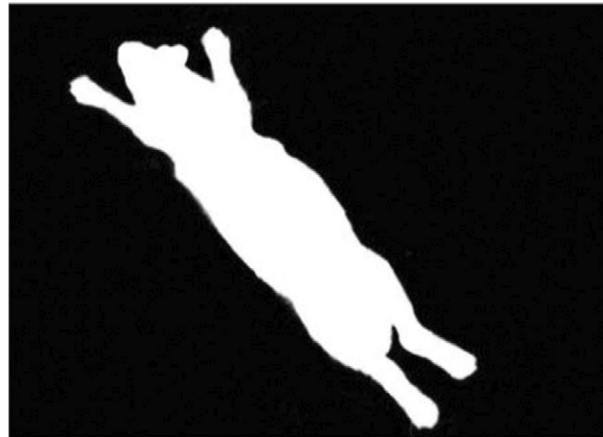
Figure 33: Position 3 (Recumbent): Ventral recumbency, legs fully outstretched

Figure 34: Position 4 (Recumbent): Lateral recumbency; hind legs fully stretched out to the right or left and front legs more or less stretched

Allometric equations have been developed and used to determine what floor space animals require based on their body weight. Petherick and Phillips (2009), suggested the following equation to calculate the minimum floor space allowance to enable mammals to adopt a resting position (position 1) simultaneously during transport: $(m^2) = 0.027 \times$ live weight $(kg^{2/3})$, corresponding to $cm^2 = 270 \times$ live weight $(kg^{2/3})$.

The floor space required to adopt Positions 1 and 2, reported in Giersberg et al. (2015), are converted into corresponding space allowances for rabbits of different weights and compared to the Petherick and Phillips (2009) equation with a constant k value of 270 in Table 30. Considering the available evidence, the AHAW Panel, based on expert opinion, concluded that the Petherick and Phillips (2009) allometric equation is a good proxy for the space needed for rabbits to adopt a sitting position (Position 1), change posture within the container and to provide some extra space between animals to ensure proper ventilation. However, if rabbits should be allowed to adopt recumbent position (Position 2), more space is needed (Table 29). This position might be adopted when rabbits are transported at high effective temperature.

Table 29 indicates the different space requirements based on planimetric measurements – depending on rabbit size and position – as well as allometric measurements and related stocking density.

Table 29: Space allowance cm^2/kg based on planimetric depending on rabbit size and position, allometric measurements and related stocking density (kg/m^2)

Weight class $\pm$ 0.25 kg	Space allowance for Position 1 (Sitting) cm^2/kg Based on planimetric measurement	Space allowance for Position 2 (Recumbent) cm^2/kg Based on planimetric measurement	Space allowance cm^2/kg (calculated for position 1) Based on Petherick and Phillips (2009) allometric equation	Stocking density kg/m^2 Based on space allowance calculated with the allometric equation
1.00	246	294	270	37
1.50	217	269	236	42
2.00	205	260	215	47
2.50	183	238	200	50
3.00	170	232	188	53
3.50	158	214	179	56
4.00	151	198	171	59
4.50	146	184	164	61
5.00	140	177	159	63

Current practice varies among farmers/transporters, apparently within a range from 130 to 160 cm^2/kg (Verga et al., 2009; De la Fuente et al., 2004a,b; Lambertini et al., 2006; Buil et al., 2004). As shown in Table 29 based on planimetric measurements (Giersberg et al., 2015), for a rabbit of 2.5 kg (normal slaughter weight for rabbits), 183 cm^2 are required to sit (Position 1 – Figure 31) and 238 cm^2 are needed to adapt resting posture (Position 2 – Figure 32). The allometric equation provides a space allowance of 200 cm^2.

Insufficient height of the container

The cage height enabling the rabbits to perform look-out sitting position is 40 cm for rabbits up to 3 kg of body weight (Council of Europe, 2006). Martrenchar et al. (2001) observed that growing rabbits cannot sit up on their hind legs in a 30-cm high cage. However, even in higher or open top cages, this behaviour was rarely observed on farm (Martrenchar et al., 2001; Princz et al., 2008). Under farming conditions, on the basis of preference test results, Princz et al. (2008) concluded that the 30–35 cm high cages are adequate for the growing rabbits up to 11 weeks of age. However, EFSA (2005) concluded that 50 cm high cages could favour rabbits on farm to sit and stand with their ears erect, and occasionally to rear up, and to thermoregulate. EFSA AHAW Panel (2020b) concluded that conventional cages, with a typical height of 28 cm and total surface of 1,200 cm^2, should not be used on farm as they lead to restriction of movement.

Transport is of a limited duration compared to time spent in housing, and the behavioural needs of rabbits during transport are poorly understood. Thus, evidence from housing conditions might not be applicable for transport conditions. Although more research is needed, it is reasonable to assume that a rabbit will experience restriction of movement if the ears touch the ceiling of the container when in a sitting position, possibly leading to discomfort and negative affective states such as frustration.

The height will be insufficient when the rabbits cannot sit in a natural position without the ears touching the ceiling of the container. It is not known if rabbits would sit up on their hind legs during transport if given the possibility. If so, they might feel restricted when the container is too low. The AHAW Panel agreed that when rabbits' ears touch the ceiling of the crate for a prolonged time they will be experiencing restriction of movement. Furthermore, their inability to stretch their ears can reduce their ability to cope with heat stress (Figure 35).

Figure 35: Rabbits destined for slaughter (2.5 kg) in a transport container. The height of the container is 25 cm (Courtesy of Leonardo J Vinco)

5.7.3.4. Preventive measures

No preventive measures exist as restriction of movement is a welfare consequence inherent to the transport of rabbits, as they are confined in containers.

5.7.3.5. Corrective and mitigative measures

Insufficient floor space allowance

Even if it cannot be avoided, the severity of restriction of movement can be mitigated by providing enough floor space per animal in the container to ensure they can all adopt a recumbent position at the same time. Furthermore, severe restriction of movement can be mitigated if rabbits can all adopt a sitting posture and are able to change posture within the container. The minimum space allowances recommended by the AHAW Panel to mitigate restriction of movement are given in Table 30 and are based on the planimetric measures when the rabbits are in ventral recumbency, front legs extended and hind legs bent to the body, and on the allometric Petherick and Philips (2009) equation [Space allowance (cm^2/rabbit) = 270 × live weight (kg$^{2/3}$)] for various categories of rabbits. Related stocking density is also provided. The equation is used to calculate the space (cm^2/rabbit) for a rabbit of a certain live weight. For practical use the results of the equation are transformed to kg/cm^2 by dividing the outcome of the equation by the average live weight of the rabbits.

Table 30: Minimum required space allowance (cm^2/kg) or maximum stocking density (kg/m^2) for rabbits at different body weights

Weight class ± 0.25 kg	Space allowance cm^2/kg (position 1) Based on Petherick and Phillips (2009) allometric equation*	Stocking density kg/m^2 Based on space allowance calculated with Petherick and Phillips (2009) allometric equation
1.00	270	37
1.50	236	42
2.00	215	47
2.50	200	50
3.00	188	53
3.50	179	56
4.00	171	59
4.50	164	61
5.00	159	63

*: Space allowance (cm^2/kg) = (270 × live weight (kg$^{2/3}$).

Insufficient height of the container

To mitigate restriction of movement (and heat stress) during transport, the height of the container should allow the rabbit to sit in a natural position, referred to as position 1 (Figure 33) in the study of Giersberg et al. (2015), with their ears erect and without touching the ceiling of the container. The height required depends on the size and category of rabbit. Considering the available evidence, the AHAW Panel, based on expert opinion, suggested, for rabbits up to 3 kg, a height of 35 cm is needed, and for rabbits between 4.5 kg and 6 kg, a height of 40 cm is required.

5.7.4. Sensory overstimulation

5.7.4.1. Description

Sensory overstimulation occurs when the rabbit experiences stress and/or negative affective states such as fear or discomfort due to visual, auditory or olfactory overstimulation by the physical environment. Sensory overstimulation happens due to the forces exerted on the animals, aversive or unfamiliar odours, noise or changes in lightning that take place during their placement in the transport containers and loading on lorries manually or with forklift.

De la Fuente et al. (2007) investigated the effect of noise for 4.5 h prior to slaughter on certain physiological and meat quality parameters of rabbits. Exposure to noise in the context of transport leads to muscular damage as demonstrated by increased levels of CK and lactate dehydrogenase (LDH) activity in the blood and a high final pH in meat. However, rabbits exposed to noise showed physiological responses to the potential stressor to a lesser degree than rabbits exposed to heat stress.

Loud sound/noise cause adverse effects including nervous and behavioural abnormalities (Peterson, 1980). Sudden noises can also cause a startled response and traumatic injuries to limbs and back. Particularly, intensity rather than audio frequency is the most damaging aspect. The sensitive hearing sensitive range of rabbits lies between 0 and 20 dB sound pressure. Nevertheless, they are also sensitive to high sound frequencies (ultrasound) detected by humans. The effect of sound levels of high intensity on rabbits is however unknown. Nayfield and Besch (1981) reported levels of 112 dB to be stressful.

The exposure to a daily 3–4 h noise regime of 65 dB for 18, 28, 40 and 50 days caused a general increase in biochemical parameters such as urea, uric acid, creatinine, cholesterol and triglycerides, while glucose level decreased significantly in domestic male rabbits weighting 1,000–1,200 g (Elwasife et al., 2015). The authors concluded that noise exposure may also enhance gluconeogenesis and glucose mobilisation to the blood.

As rabbits are crepuscular animals, their vision is not suited for either bright day-time or dark night-times. However, Harkness (1988) showed that rabbits exposed to varying light intensity, did not make an effort to avoid brighter lights. Bright light is a concern especially in albino rabbits with no pigmentation in their eyes.

The rabbit nose has a very complex structure (Xi et al., 2019) showing the high olfaction acuity (Negus, 1954). Rabbits rely upon their olfactory system (e.g. for detecting danger, finding food, attracting mates, identifying territories) and they can sense predictors miles away, or smell food below the ground (Schalken, 1976). Even newborn rabbits have superior olfaction, which guides them even with closed eyes to their mothers' nipples (Schaal et al., 2003). Rabbits often sniff or wiggle their noses, to capture or distinguish tiny traces of pheromones or chemical molecules.

The odour of a predator in the enclosure can induce physiological stress as shown by an increase in faecal corticosterone metabolites concentrations (Monclús et al., 2006). However, the odour did not affect all the animals in the same way. In particular, males experienced a higher increase than females, though the overall response was similar for both sexes.

On the other hand, the olfactory region can be susceptible to infectious diseases and toxic damage (Xi et al., 2019). The exposure to the automotive exhaust fumes can cause ultrastructural-changes of the brain (Yoshino et al., 1981).

5.7.4.2. ABMs

In the context of transport, no feasible ABMs for sensory overstimulation in rabbits were found.

5.7.4.3. Hazards

Novel visual, auditory or chemical stimuli may evoke a potential avoidance response in the animals simply as a result of caution related to any threatening stimulus not previously experienced. Other

stimuli that are sudden, repeated or of high intensity may also be perceived as threatening or potentially dangerous.

Unexpected loud sound/noise

Sudden alterations in sound, will all constitute a threat to the animals' well-being and will induce fear to the rabbits. The catching and crating process as well as the journey in the vehicle are stages with loud noises originating mainly from machines, the manipulation or movement of containers, or people who enter the house without precaution (e.g. shouting while catching the animals).

Sudden light changes

Excessive light or darkness can have significant negative effects as well as sudden changes in light intensity.

Unfamiliar odours, aversive levels of aerial dust and gases

Rabbits have superior olfaction, unfamiliar smells perceived during transport may elicit stress in rabbits. Furthermore, given its sensitivity, olfactory region itself can be susceptible to toxic damage. Exposure to exhaust fumes and other general pollutants can have harmful effects (Hue-Beauvais et al., 2019).

5.7.4.4. Preventive measures

Unexpected loud sound/noise

Excessive sound or noise should be avoided or minimised in all stages of animal production, handling and transport. Therefore, strategies should be employed which reduce the risk of loud and/or unexpected sound and noise.

It is important to limit unexpected loud noises because they lead to fear and decrease coping capacities of animals. The preventive measures will consist in staff education and training (i) to make them aware that noise at the animals' level should be avoided and (ii) to make them avoid shouting and making noise with the equipment, and identify and eliminate the sources of noise. In addition, the machine should be setup correctly to avoid excessive noise.

Entering the rabbit house and approaching the animals calmly with a minimum of noise to minimise disturbance. Limiting sudden and extreme noise of equipment as much as possible. Careful selection of people with appropriate skills and the right attitude or training to acquire skills appropriate to the tasks would help to minimise fear when handling animals.

The noise produced by the vehicle through engine operation, air movement around and through the load and road surface contact can be minimised (although not entirely prevented) by the use of improved suspension systems and vehicle enclosure or insulation (use of curtain or solid sides with high quality ventilation). In the future the use of electric vehicles may reduce the engine sounds significantly.

Visual stimuli

Excessive visual overstimulation should be avoided or minimised in all stages of animal production, handling and transport. Therefore, strategies should be employed which reduce the risk of visual overload. The preventive measures will consist in staff education and training, in the choice of proper vehicle design and use of curtains.

Careful selection of people with appropriate skills and the right attitude or training to acquire skills appropriate to the tasks would help to minimise fear when handling animals.

Sudden switching on of lights without prior warning should be avoided during the dark period as it may cause consternation and even spontaneous ovulation and occasional self-injury (CCAC, 2020).

Unfamiliar odours, aversive levels of aerial dust and gases

Excessive olfactory overstimulation should be avoided or minimised in all stages of animal production, handling and transport. Therefore, strategies should be employed which reduce the risk of olfactory overload. The preventive measures will consist in staff education and training, in the choice of proper vehicle design and materials, proper maintenance to avoid excessive exhaust fumes.

In transit noxious gases may enter the load from vehicle exhaust fumes on busy roads when the vehicle is moving slowly or is stationary. This risk may be reduced by vehicle enclosure (closed curtain sides) and ensuring adequate ventilation (including mechanical ventilation.

5.7.5. Motion stress

5.7.5.1. Description

An overall description of motion stress is given in Section 3.7.5.1.

Rabbits have been used as model animals in anatomic and physiologic research studying acute vestibular dysfunction and motion sickness (e.g. Balaban, 1996), research focused on testing of safety of drugs against motion sickness (e.g. Al-Ghananeem et al., 2007; Cao et al., 2007) and even space motion sickness (e.g. Yang et al., 2002). However, they have only rarely been used to study effects of velocity, acceleration and vibration on their welfare.

Reséndiz Cruz (2016) evaluated changes in metabolites, blood gases and electrolytes in rabbits exposed to vehicular transport (group 1) and movement simulations (group 2) for 30 min and 60 min (group 3). In group 1, animals were placed in plastic cages on the rear platform of a vehicle during the travel time of 30 or 60 min. A vehicle was driven at a speed of 70 km/h, in a circuit of dirt roads (unpaved roads) of 1.6 km diameter, located 100 m away from the rabbit production unit. The topography of the road had a maximum transverse slope of 13%, allowed an average maximum speed of 30 km/h, had vertical curves at 4 m/%, to an overelevation maximum of 10%, and a maximum crown width of 4 m. In group 2, a vibration platform Bio- Compact Pro-form was used with an area of 70 cm × 57 cm × 27 cm. Acceleration movements were programmed at a frequency of 60 Hz during 30 or 60 min. The results show that the movement of rabbits on the vibration platform induced stress but at a lower level than that induced by the transportation itself where more factors such as slow or sudden movements, drafts, dust and environmental changes were involved.

5.7.5.2. ABMs

In the context of transport there are no feasible ABMs for motion stress in rabbits.

5.7.5.3. Hazards

During journey, animals experience motion stress due to the forces exerted as a result of acceleration, braking, stopping, cornering, gear changing, vibrations and uneven road surface.

Vibration and acceleration

Vibrations, accelerations, decelerations, impacts may result in poor postural stability, muscle fatigue, exhaustion and in some cases motion sickness. Hazards affecting motion stress in rabbits include those related to the driver (careless driving – driver accelerating and braking too fast or too sudden), those related to the vehicle (type of vehicle, suspension) and those related to the road (inappropriate roads, uneven road surface, bumps).

5.7.5.4. Preventive measures

Driving style appropriate for the carriage of livestock should be ensured by the necessary training and education. Excessive acceleration, deceleration, braking and cornering should be avoided.

Appropriate type of vehicle should be chosen for animal transport. For any vehicle, good vehicle maintenance will reduce the risks associated with motion and vibration. Particular attention should be paid to tyre pressures and maintenance of the vehicle suspension.

When planning animal transport, the appropriate route should be chosen to avoid inappropriate roads.

5.7.5.5. Corrective and mitigative measures

Adjust driving style to the road surface.

5.7.6. Thermal stress

Thermal stress means that the animal experiences stress and/or negative affective states such as discomfort and/or distress, when exposed to effective temperatures outside its thermal comfort zone. The comfort zone temperature in rabbits is around 18–21°C (Marai and Habeeb, 1994; Habeeb et al., 1998, Abd El-Monem, 2001). Optimal climatic conditions for rabbits are air temperature 13–20°C (average 15°C) and relative humidity 55–65% (average 60%) (Marai and Rashwan, 2004).

The thermoneutral zone varies from 20°C to 30°C. Research on physiological parameters such as respiration rate, panting and body temperature estimated the upper critical temperature to be below 30°C (Gonzalez et al., 1971; Kluger et al., 1973; McEwen and Heath, 1973; Scheele et al., 1985;

Jin et al., 1990). There is lack of information for intermediate values from 20°C to 30°C increasing the uncertainty.

Based on significant differences in heat production between 10°C and 20°C, most studies suggested the lower critical temperature should be between these values. Cervera and Carmona (2010) estimated the lower critical temperature to be at approximately 15°C.

5.7.7. Heat stress

5.7.7.1. Description

The animal experiences stress and/or negative affective states such as discomfort and/or distress when exposed to high effective temperature.

Rabbits are very sensitive to high temperature since they have limited ability in eliminating excess body heat through evaporation, e.g. by sweating or panting. Heat stress induces behavioural (e.g. depression in feed intake) and physiological changes (e.g. disturbances in water, protein, energy and mineral metabolism balances, enzymatic reactions, hormonal secretions and blood metabolites (cited in: Marai and Rashwan, 2004; Johnson, 1980; Wittroff et al., 1988; Habeeb, 1993; Kasa and Thwaites, 1992). Dalmau et al. (2015) described an increase in faecal cortisol metabolites and behavioural changes in rabbits subjected to mean temperatures of 27°C for 6 h a day, when compared to animals subjected to 20°C. Blood metabolites such as glucose, serum total protein, serum total lipids and cholesterol decrease in rabbits exposed to heat stress conditions, which may be correlated to the decrease in energy metabolism during heat exposure (Cervera and Carmona, 2010). Heat stress is also related to changes in blood cortisol, lactate and glucose, creatine kinase and lactate dehydrogenase enzymes (De la Fuente et al., 2004a,b). The plasma concentrations of cortisol, lactate and glucose, CK and LDH activity and osmolarity, as well as liver and muscle glycogen concentrations were higher in rabbits transported in summer than in winter. Stocking density had no effect on the analysed parameters (De la Fuente et al., 2004a,b).

The rabbit's normal body temperature ranges from 38.3°C to 39.4°C. The average lethal body temperature is considered to be 42.8°C.

Exposure to high ambient temperature induces rabbits to try to balance the excessive heat load by using different means to dissipate, as much as possible, their latent heat. When the ambient temperature increases above 30°C, rabbits show a significantly higher respiration rate, more than 32–60 breaths per minute in does which is probably age dependent (Harcourt-Brown, 2002). Johnson et al. (1957) reported that respiration rate increased rapidly from 69 to 190 breaths/min in NZW rabbits when environmental temperature increased from 10°C to 30°C. Brody (1945) estimated that respiration rate goes up by 5–6 breaths /min for each 1°C increase in air temperature.

Marai and Habeeb (1994) indicated that between 0°C and 30°C, latent heat evacuation is only controlled by altering the breathing rate. Moreover, the latter is very inefficient when the environmental temperature is in excess of 30°C (Fayez et al., 1994). Since rabbits do not sweat, at temperatures above 30°C they stretch out to lose as much heat as possible by conduction, radiation and convection, raise their ear temperature, stretch the ear pinnae and spread them far from the body to expose the surface to the surroundings in order to increase heat dissipation (Marai and Rashwan, 2004). Gonzalez et al. (1971) reported that ear temperature increased by 14°C, when environmental temperature was elevated from 20°C to 30°C. Breeding rabbits under heat stress increase the time lying in a stretched out position, ventrally, laterally or dorsally (Rafel et al., 2012), and decrease their activity levels (Marai and Rashwan, 2004). According to Lebas et al. (1986), rabbits can no longer regulate their internal temperature above 35°C and heat prostration sets in, while at 40°C, considerable panting and salivation occurred.

The effective temperature (i.e. the temperature that animals actually experience) may differ by several degrees from that measured in the surroundings and depends on several factors, such as air speed and temperature, relative humidity, flooring and cage type, stocking density and the animal's stage of production and health status (NFACC, 2021).

Since there is a close relationship between the ambient temperature and humidity, the relative humidity has to be taken into consideration when measuring the severity of thermal discomfort (e.g. heat stress). The feeling of warmth under hot ambient temperature increases with high relative humidity.

5.7.7.2. Index based on temperature and humidity to predict heat stress (THI)

For the above reasons, the application of a 'temperature–humidity index' (THI) was proposed by LPHSI (1990), which was modified and adopted for rabbits by Marai et al. (2001) as follows:

$$THI = db°C - [(0.31 - 0.31RH)(db°C - 14.4)],$$

where db^{o}C is the dry-bulb temperature in degrees Celsius and RH is the relative humidity.

The thermal comfort limits for rabbits (Marai et al., 2002) are defined as: temperature–humidity index (THI) < 27.8, absence of heat stress; 27.8–28.9, moderate heat stress; 28.9–30, severe heat stress; THI > 30, very severe heat stress.

When comparing such classification with that used for larger animals, such as sheep and cattle (< 22.2 = absence of heat stress, 22.2 − < 23.3 = moderate heat stress, 23.3 − < 25.6 = severe heat stress and ≥ 25.6 very severe heat stress; Marai et al., 2000), it is evident that rabbits tolerate higher climatic stress than do large mammals. This may be due to rabbits' higher body temperature.

During commercial transport, rabbits are often exposed to factors that may act as stressors and/or limit the possibility of the animals to thermoregulate using methods present in non-transported control conditions. In addition, most of the classical studies on thermal physiology have allowed animals a certain level of feed and water intake, as well as stable or resting conditions. In contrast, transport often includes feed and water deprivation, exposure to vibration and motion forces, low space allowances and highly variable ventilation rates.

Effective temperature can be divided into a safe, alert or danger zones based upon thermoregulatory demands and physiological responses. In the safe zone, animals require minimal thermoregulatory effort. In the alert zone, animals need increasing thermoregulatory efforts. In the danger zone, animals will require serious thermoregulatory efforts that might be insufficient and will lead to heat stress.

Based on the available evidence, the AHAW Panel concluded that if THI remains below 27.8, rabbits are unlikely to experience heat stress during transport (safe zone). Between THI values of 27.8 and 28.9, there will be an increasing risk of heat stress (alert zone) and above THI of 28.9, the rabbit's mechanisms to cope with heat stress will become less effective and the rabbits will experience heat stress (danger zone). The combinations of relative humidity and dry-bulb temperature giving rise THI values for the safe, alert and danger zones were calculated and can be found in Figure 36.
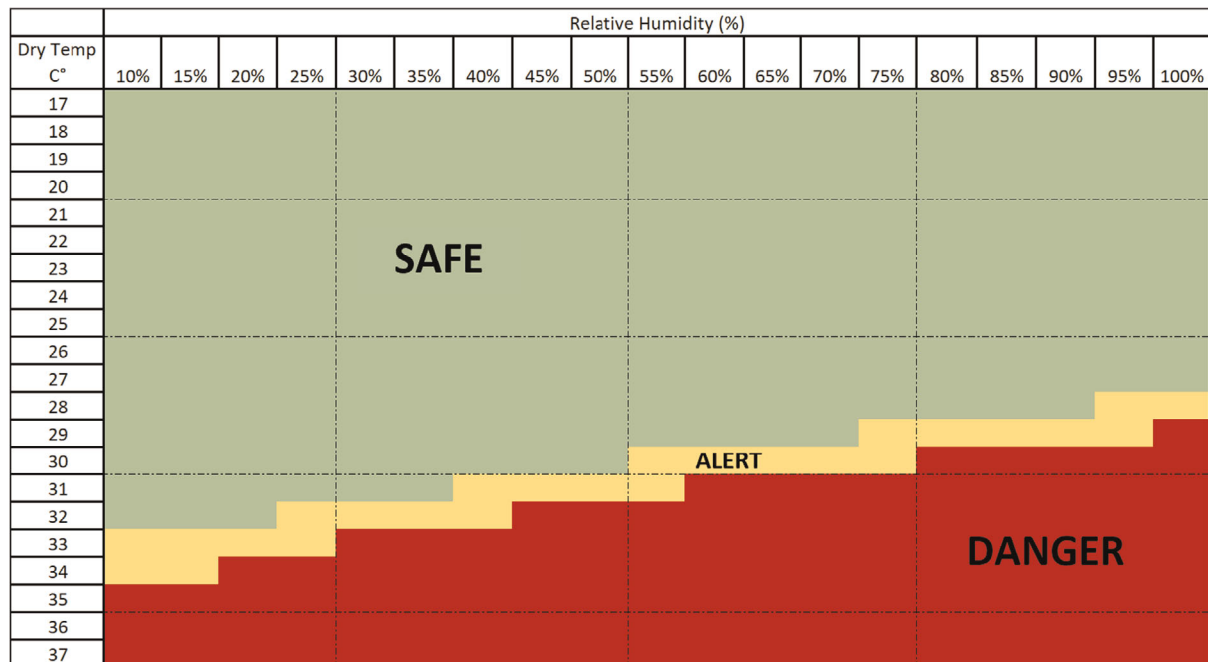
Figure 36: Thermal Comfort Zones for rabbit transport defined by temperature humidity index (THI)

5.7.7.3. ABMs

ABMs that are considered feasible for assessing injuries in rabbits during transport are given in Table 31.

Table 31: ABMs of heat stress in rabbits

ABM	Definition and interpretation of the ABM
Panting	**Definition**: An animal is considered panting when it is breathing with short and quick breaths and with the mouth open (Dalmau et al., 2020). The head is extended backwards (Lebas et al., 1997). Respiration rate goes up by 5–6 breaths/min for each 1°C increase in air temperature (Brody, 1945).
	Measurement: Observation of the proportion of rabbits showing this behaviour in a representative sample of animals, especially checking containers located on top and in centre of load. Manual inspection of these sections is feasible at unloading. Containers on the edge of load can be inspected also at loading and during stops.
	Sensitivity is high as panting occurs only during heat stress.
	Specificity is high, as the absence of heat stress is associated with normal respiration rates (absence of panting).
Position and colour of the ears	**Definition**: The rabbits stretch the ear pinnae and spread them far from the body to expose the surface to the surroundings, while the ear temperature increases (Marai and Rashwan, 2004). The ears with prominent blood vessels (Lebas et al., 1997) turn from pink to red (Peeters, 1989) (from EFSA AHAW Panel, 2020a).
	Measurement: Observation of the proportion of rabbits showing this behaviour in a representative sample of animals, especially checking containers located on top and in centre of load. Manual inspection of these sections is feasible at unloading. Containers on the edge of load can be inspected also at loading and during stops.
	Sensitivity is low, as not all the rabbits experiencing heat stress will stretch the ear pinnae and turn the ears from pink to red. Furthermore, some animals might not stretch the ears due to limited space.
	Specificity is low, as in the absence of heat stress the rabbits might have also reddening of the ears due to a different reason (inflammation, parasitic infestation).
Salivation	**Definition:** An abnormally abundant flow of saliva (from EFSA AHAW Panel, 2020a). If there is a drool on the rabbit's chin, it is a sign of excessive salivation
	Measurement: Observation of the proportion of rabbits salivating in a representative sample of animals, especially checking containers located on top and in centre of load. Manual inspection of these sections is feasible at unloading. Containers on the edge of load can be inspected also at loading and during stops.
	Sensitivity is moderate as not all the rabbits experiencing heat stress will salivate.
	Specificity is low, as in the absence of heat stress, rabbits might salivate due to other reasons.

Among these ABMs, only panting is considered to be sensitive and specific for the assessment of heat stress.

5.7.7.4. Hazards

Too high effective temperature

The effective temperature perceived by a rabbit is a combination of the temperature, the humidity and the ventilation or speed of wind. In hot and humid environmental conditions, poor ventilation will exacerbate the perceived temperature. When the effective temperature is too high (above their thermoneutral zone ($< 27.8°C$), the thermoregulatory capacities of the rabbits for homoeothermy are exceeded and they show difficulty achieving a balance between body heat production and body heat loss that leads to heat stress.

Heat exchange via radiation is often overlooked but can be a major contributor to heat stress.

Space allowance also has an important impact on thermoregulation. Since rabbits do not sweat, at temperatures above 25–30°C they stretch out to lose as much heat as possible by radiation and

convection, raise their ear temperature, stretch the ear pinnae and spread them far from the body to expose the surface to the surroundings in order to increase heat dissipation. Inability to do so when the stocking density is too high may increase the heat stress. In contrast, the reduction of space availability does not protect against low temperatures as Caucci et al. (2018) found batches with high stocking density showing a higher DOA even in winter.

The position on the transport vehicle has an effect on blood corticosterone levels in rabbits (Liste et al., 2006, 2008). The studies suggest that animals could better adapt to the conditions in the transport if placed on the top level because of better ventilation, more visibility and less dirt. Bottom or middle positions showed increased corticosterone levels (Liste et al., 2008). According to Liste et al. (2006), in hot weather, the position on the transport vehicle had a greater effect on rabbit welfare than the duration of the journey.

In hot and humid environmental conditions, poor ventilation will exacerbate the perceived effective temperature. Ventilation plays a key role in the dissipation of heat and humidity as produced by animals during transport (Schrama et al., 1996; Consortium of the Animal Transport Guides Project, 2017). A higher level of humidity in the air will worsen the effect of high temperatures. A higher air speed (in practice: active or fan ventilation) will reduce this effect.

Insufficient space allowance

Space allowance has an important effect on thermoregulation. Insufficient space allowance is detrimental in hot weather as the animals cannot space and stretch out to lose heat from the sides of their body – rather they are gaining conductive heat from the body warmth of conspecifics in close contact.

5.7.7.5. Preventive and corrective measures

Preventive and corrective measures for too high effective temperature

To prevent rabbits to experience heat stress, they should travel in their safe zone corresponding to THI values below 27.8, so they require no or minimal thermoregulatory efforts during the journey. Travelling in the alert zone will result in minimal or moderate physiological responses reaction to heat stress, similarly to domestic birds. Rabbits could be transported when in the alert zone, however evidence is lacking about the maximum journey duration without detrimental effects on welfare. Rabbits should never travel in the danger zone in order to avoid heat stress (THI above 28.9). The combinations of relative humidity and dry-bulb temperature giving rise to AET values for the safe, alert and danger zones were calculated and can be found in Figure 38.

To monitor effective temperature, vehicles should be equipped with sensors recording dry-bulb temperature and relative humidity inside the containers. Although sensors recording dry-bulb temperature have most commonly been used in road transport of animals in the past, it would be a significant refinement to use improved sensors which take account of humidity effects. Temperature and humidity sensors should be fitted as close to the rabbits as possible at several locations to include the top 'hot spot' at the front, the 'cold spot' at the outside rear and bottom of the load and representative points in between. Both the driver and the supervisor should monitor the microclimate (combination of T and RH) of the load and adjust the ventilation if the THI exceed the comfort zone levels.

Convective heat loss (including increased air movement within the containers) is the most effective means of managing heat stress for animals in transit. This depends on good ventilation providing effective air distribution throughout the load with the capacity to alter air speeds within each container. At too high effective temperatures increased ventilation of the load is required to remove excess heat and moisture production from the rabbits (thus reducing the effective thermal load with particular attention to regulating the in-crate humidity or water vapour density). In passive ventilated vehicles, the removal of the curtains will increase ventilation and may allow the rabbits to stay in their thermoneutral zone when the vehicle is in transit but not when it is stationary.

To aid passive ventilation, vehicles should have solid roofs which can be raised when stationary in hot weather, including during loading except in the case of containers fixed to the trailer with a central longitudinal passage (also called liners). In this case the roof should be open above this passage. There should be vents in the headboards and tailgate, with sliding covers to enable air flow to be adjusted according to weather and bird conditions.

New designs of modules and crates have been developed by commercial companies to increase load ventilation and specifically to increase air movement in and around the crates and animals.

Modified crate structure (e.g. increased size or number of perforations) increases efficacy of passive ventilation regimes. The passive ventilation of all vehicles has been improved by attention to pressure profiles over the moving vehicle, location of natural air inlets and outlets and internal air flow mixing. These developments are particularly effective in minimising the risk of heat stress in relatively low external temperature conditions where, in fully curtained/sheeted vehicles, internal heat and moisture production can result in elevations of internal bio-load temperature and humidity above the recommended limits.

Other preventive measures to avoid too high effective temperature can be put in place at the time of transportation. Like for broilers, hottest hours of the day should be avoided (Warriss et al., 2005). Reducing the number of rabbits per crate or drawer will reduce heat production and improve air flow around the animals. During loading and at arrival, when there is a too high effective temperature, rabbits should be protected from the sun and ventilation should be provided. At arrival, rabbits should be unloaded immediately from the vehicle. Containers should be spaced out and fans directed at the rabbits if panting is observed.

If there are delays upon arrival at a slaughterhouse in warm or hot weather naturally ventilated vehicles should keep moving (go out on the road again) until rabbits can be unloaded into a lairage with appropriate environmental conditions as detailed in the Slaughter Opinion (EFSA AHAW Panel, 2020a).

In order to reduce the risk of thermal stress all slaughterhouses should be equipped with holding facilities with banks of fans to ventilate the stationary vehicle or trailers prior to lairage or unloading.

Nevertheless, the modular and container structure of vehicles for rabbit transport makes it difficult to achieve good ventilation of all rabbits in the bio-load using only passive ventilation systems.

Air circulation and ventilation rate are better controlled in fan ventilated vehicles, where the movement of air into, within and out of the vehicle container is controlled by a combination of suitably positioned mechanical fans of sufficient capacity, and natural apertures which should enhance established natural air pressure gradients. A major advantage of mechanical ventilation is that air flow may be adjusted in response to external conditions, number of rabbits transported and their physiological requirements. These vehicles have the major advantage that the ventilation is not dependent upon vehicle movement and improve the distribution of airflow in the actual load thus reducing the risk of thermal stress. Therefore, to achieve thermal comfort for all rabbits in transit, controlled and uniform mechanical ventilation is essential. Mechanically ventilated vehicles and trailers have been designed and are successfully operated in other sectors of livestock transportation (e.g. cattle, pigs and sheep).

Furthermore, controlled environment or air-conditioned vehicles or trailers can regulate or modify internal thermal conditions by appropriate heating or cooling. They have the major advantage that the internal environment may be controlled precisely regardless of external weather or thermal conditions and does not rely upon vehicle movement. Controlled environment vehicles are not common in the EU for the transport of rabbits for slaughter but should be employed as designs are available.

Preventive and corrective measures for insufficient space allowance

Increased space may facilitate penetration of air movement into and between the containers and thus enhance convective cooling from the surface of the birds and evaporative cooling of the exchange surfaces including the respiratory tract. Common practice from industry is to increase space allowance of 10–25%. However, it is not possible currently to make precise recommendations for quantitative strategies as no published research data are available.

In order to mitigate heat stress during loading in hot weather, the vehicle can be loaded from back to front and preferably completing first the whole lower layer (where 2-tier module systems are used). This is because heat rises and accumulates behind the front headboard so loading potential hot-spots last delays the potential onset of heat stress during the journey. Where fans are fitted to the vehicle, turn them on during loading and if rabbits are observed to be panting direct free-standing fans at the loaded rabbits.

5.7.8. Cold stress

5.7.8.1. Description

The animal experiences stress and/or negative affective states such as discomfort and/or distress when exposed to low effective temperature.

The thermal comfort zone temperature in rabbits is around 18–21°C (Marai and Habeeb, 1994; Habeeb et al., 1998). Below 18°C, the animal has to expend energy to maintain its body temperature (Marai et al., 2002).

Vasoconstriction of peripheral blood vessels helps the rabbit to keep warm in cold conditions, so that metabolism does not need to increase to offset heat loss. In particular, the amount of heat conserved in the ears (with an area depending on breed, approximately 0.026 m^2 in adults) is considerable. McEwen and Heath (1973) reported that heat loss was 0.2 W °C − 1 linear over the range of 0–30°C ambient temperature, about half of the theoretical loss if the ears were maintained at core body temperature. Responses to cold stress rely on more carbohydrates being utilised and fat tissue being easily mobilised. Long-term adaptation to cold involves increased insulation (Cervera and Carmona, 2010).

The rabbits exposed to cold showed physiological responses to the potential stressor, although to a lesser degree than rabbits exposed to heat (De la Fuente et al., 2007). Luzi et al. (1992) observed lower liver and muscle glycogen concentration as evidence of increased muscle activity during winter transport. De la Fuente et al. (2007) assessed cold stress in rabbits transported for 4.5 h with a maximum and minimum temperature recorded of 2.1°C and − 1.1°C, respectively, for the first trial, and 2.1°C and − 0.5°C, respectively, for the second trial with a humidity ranging between 55 and 60%. They reported increased levels of creatine kinase and a higher PCV as well as decreased muscle glycogen concentration compared to the control.

In an experiment reported by Nagasaka (1974), rabbits´ rectal temperature which was 38.94°C ± 0.1 at ambient temperature 25°C, fell considerably in the cold environments. At 5°C, rectal temperature was 37.39°C ± 0.18. However, in cold acclimated rabbits, rectal temperature was regulated within the range of 38.6–38.3°C at ambient temperature ranging from 5°C to 25°C.

Most studies have found appreciable differences in heat production between 10°C and 20°C and, therefore, the lower critical temperature should be between these values. From a survey of the literature, McEwen and Heath (1973) calculated a value of approximately 15°C for the lower critical temperature (Cervera and Carmona, 2010).

Based on the limited available evidence, the AHAW Panel concluded that the lower limit of the comfort zone is estimated to be at 18°C. If the effective temperature in the containers remains above this threshold, rabbits are unlikely to experience cold stress during transport. T° values between 18°C and 10°C (estimated to be critical temperature of the thermal neutral zone) will increase the risk of cold stress. Below 10°C, the rabbit's mechanisms to cope with cold stress will become less effective and the rabbits will experience cold stress.

5.7.8.2. ABMs

ABMs that are considered feasible for assessing cold stress in rabbits during transport are given in Table 32.

Table 32: ABMs for cold stress in rabbits

ABM	Definition and interpretation of the ABM
Huddling	**Definition**: Sitting close together in tight groups or clumps often with open space in between (from EFSA AHAW Panel, 2020a)
	Measurement: Observation of the proportion of rabbits showing huddling behaviour.
	Sensitivity is high as rabbits will clump together as a response to low environmental temperatures.
	Specificity is low as in the absence of cold stress, rabbits might also huddle voluntarily (de Jong et al., 2011). If rabbits are kept in close proximity of each other, this does not necessarily mean that they experience cold.
Piloerection	**Definition:** The rabbit's fur is raised from the body, giving a rounded, fluffy appearance. This increases the thickness of the insulating fur.
	Measurement: Observation of the proportion of rabbits showing piloerection. Especially checking containers located at the bottom and on the side of load. Manual inspection of these sections is feasible at loading, unloading and during stops.
	Sensitivity is high, as piloerection always occurs in severe cold stress (autonomic response).

ABM	Definition and interpretation of the ABM
	Specificity is high as the absence of piloerection is a sign of the absence of cold stress.
Shivering	**Definition:** Shaking slightly and uncontrollably (Strawford et al., 2011). Slow and irregular vibration of any body part, or of the body as a whole.
	Measurement: Observation of the proportion of rabbits showing this behaviour in a representative sample, especially checking containers located at the bottom and on the side of load. Manual inspection of these sections is feasible at loading, unloading and during stops.
	Sensitivity is medium. Rabbits with cold stress might not show shivering as individual rabbits vary in their capacity to use shivering for thermoregulation.
	Specificity is moderate as in the absence of cold stress, rabbits might shiver as signal of fear or nervousness.
Core body temperature	**Definition:** Core body temperature is the temperature of the internal organs. A body temperature lower than 38°C indicates that rabbits are too cold.
	Measurement: Measuring rectal temperature by inserting a thermometer into the animal's anus. Accurate measurement of core body temperature involves invasive medical procedures but rectal temperature is believed to provide the best estimation of the core body temperature.
	Sensitivity is high. When rabbits are cold, rectal temperature decreases.
	Specificity is high. When rectal temperature is 38°C or above, rabbits are not too cold.
Dropping of the ears	**Definition:** When the ambient temperature is below 10°C, rabbits curl up to minimise their body's surface area and drop the ears to bring them closer to the body (Marai and Rashwan, 2004).
	Measurement: Observation of the proportion of rabbits showing this behaviour in a representative sample, especially checking containers located at the bottom and on the side of load. Manual inspection of these sections is feasible at loading, unloading and during stops.
	Sensitivity: is high. When rabbits are cold, they will drop the ears.
	Specificity: is low. In the absence of cold stress, droopy ears might be result of stress in general or of lack of space.

Among these ABMs, piloerection, shivering and core body temperature are considered to be sensitive and specific for the assessment of cold stress.

5.7.8.3. Hazards

Too low effective temperature

The effective temperature perceived by an animal is a combination of the temperature, the humidity and the ventilation or wind speed. In this case, humidity is not so relevant as for high effective temperature.

Loading into wet containers in cold weather may cause initial chilling of the animals. During transport in cold and humid environmental conditions, lack of curtains to protect the rabbits and lack of heating system in the vehicle might result in rapid decrease of the perceived temperature.

On the majority of passively ventilated vehicles the air flow when moving tends to be from back to front due to the pressure profile when curtains or sides are closed. Adequate ventilation, regardless of ambient temperature, means that even with minimal ventilation vents minimised, there is still a risk of water entry from both precipitation and road spray. Such wetting is accompanied by increased convective cooling in the load region close to the air inlet. The combined effects of disruption of the fur insulation by wetting, enhanced convective cooling by air movement and increased evaporative cooling from the wetted surface. Seltmann et al. (2009) found a shift in the lower limit of the thermo-neutral zone in wetted rabbits. Not only wetness and ambient temperature but also interactions with body mass were involved in shaping an animal's thermal balance. A higher body mass was beneficial under cold conditions when the thermal conductance of the fur was increased by wetting. These thermal challenges combined with lower air temperatures greatly increase the risk of cold stress.

Cold stress has a less significant effect on rabbit welfare compared to heat stress because, unless extreme, it can be tolerated for longer with fewer lasting effects. Voslarova et al. (2018) did not find a negative impact on mortality in rabbits during transit transported within a wide range of environmental temperatures from −5°C to 19.9°C. However, journeys carried out at temperatures below −5°C were associated with increased mortality (0.17%). Petracci et al. (2010) reported a significantly increased DoA risk associated with environmental temperatures below 7.3°C. The number of rabbits that died between catching and the moment of slaughter was 0.091%.

5.7.8.4. Preventive and corrective measures

Under cold climatic conditions, vehicles and/or modules should have curtains which can be used according to weather conditions. The design may need to vary according to weather and may need a combination of mesh and solid panels. The use of curtains would be merited in order to prevent the rabbits freezing at the rear of the vehicle. Some vehicles in colder countries may have solid sides with defined inlet and outlet apertures which may function in a similar manner to modified curtained vehicles but give more thermal protection during cold weather.

In mechanically ventilated vehicles, the lower temperature limit can be maintained or exceeded by management of the ventilation rate and air distribution by adjusting vents and inlet outlet area ratios. At low external temperature the heat and moisture production of the animals within the load will allow elevation of in-crate temperature by reducing crate ventilation rate and mixing rate of on-board air with incoming cold air from outside the vehicle.

It is recommended that any wetting of crates and drawers is avoided (e.g. washing of containers with inadequate drying or rain ingress during the journey to the farm). As the risk is greatest at the air inlet it may be necessary to avoid loading of these modules and crates with rabbits (leave empty).

During loading and unloading, the vehicle should park in warm and sheltered areas, avoiding wind in cool or cold weather. Avoiding coldest hours of the day for transportation of animals will allow them to avoid extreme climatic conditions, which are especially problematic when vehicles are not equipped with curtains (European Commission, 2017b). At arrival and lairage, if effective temperature is still below thermoneutral zone, then adequate shelter should be provided to protect rabbits from the wind.

At lairage, in cold weather, closed shelter with heating should be provided. It is also advisable to reduce the gap between the rows of containers to limit draughts without preventing the movement of employees between crates. It is also advisable to ensure that the doors of the waiting area are properly closed.

5.7.9. Prolonged hunger

5.7.9.1. Description

Rabbits transported in containers might be deprived of feed for a period of time before being caught and crated and until they are slaughtered or released into new housing. This practice may contribute to reducing the incidence of faecal contamination of the carcass during slaughter even if it is a less common practice compared to poultry (Verga et al., 2009). Due to caecotrophy, rabbits are usually considered to be very resistant to hunger (Lebas et al., 1986), although crating and transportation can also cause the rupture of caecotrophy practice which leads to higher spillage and rabbit contamination (Jolley, 1990).

Prolonged hunger occurs when rabbits experience craving or urgent need for food or a specific nutrient, accompanied by an uneasy sensation (a negative affective state), and eventually leading to a weakened condition as metabolic requirements are not met. Depending on total time without feed, this welfare consequence might affect all rabbits transported in containers, and prevalence is thus regarded as high. The duration of the welfare consequence will depend on onset and journey time, but, unless the rabbit ingests sufficient feed while in the containers, prolonged hunger will continue with increased severity until it is slaughtered or unloaded and provided with feed. Severity of this welfare consequence is regarded as moderate to high.

Behavioural responses in rabbits to 24 h food deprivation include an alternation of short periods of search behaviour (4–10 min) and longer periods of rest (20–70 min) (Kromin et al., 2016).

Malikova and Petrova (1998) used thermoencephaloscopy to study the distribution of brain thermal fields during thirst and hunger. Thermal asymmetry was observed in the states of thirst and hunger with the temperature of the left hemisphere being higher. Motivational states of hunger or thirst in rabbits measured with this method were produced by the relevant deprivation of different duration (24 and 48 h). Generalisation of the brain temperature reaction depended on the level of motivational

excitation. The interhemispheric brain asymmetry with higher temperature in the left hemisphere in the states of feed and water deprivation had a pronounced trace effect, i.e. persisted for some time after satisfaction of the corresponding need.

Feed deprivation affects carcass yield and live weight losses. Lambertini et al. (2006) reported weight loss due to emptying of the guts over the first 4–6 h. If feed deprivation persists beyond 6 h, there will be a reduction in water content and body tissues with a corresponding effect on carcass yield (Trocino et al., 2003). According to Cavani and Petracci (2004), rabbits lose 3–6% of body weight during the first 12 h of fasting. This increases to about 8–12% at 36–48 h. Generally, weight loss is slightly lower if fasted rabbits are allowed access to water before crating (Ouhayoun and Lebas, 1994). Kromin et al. (2016) also documented behavioural responses to 24 h food deprivation showing that rabbits experience negative affective state.

Considering the available evidence, the AHAW Panel, based on expert opinion, concluded with 50–100% certainty (more likely than not) that rabbits subjected to feed withdrawal periods longer than 6 h will experience prolonged hunger, and with 66–100% certainty (from likely to almost certain) that rabbits subjected to feed withdrawal periods longer than 12 h will results in weight loss and will experience prolonged hunger which is detrimental to their welfare.

5.7.9.2. ABMs

There is no feasible animal-based measure during transport to estimate prolonged hunger in rabbits.

5.7.9.3. Hazards

Feed deprivation too long

As presented above, the hazard for prolonged hunger is the feed deprivation that is performed routinely as a hygienic measure. There is no benefit to rabbit welfare of feed withdrawal before crating.

Feed deprivation can be performed at different points in time on the farm before catching of the animals before transport.

5.7.9.4. Preventive measures

To prevent the risk of rabbits to experience prolonged hunger during transport, the total time without feed and water should not exceed 6 h.

To prevent prolonged hunger when rabbits are transported to slaughter, total feed withdrawal time should be minimised taking into consideration the duration of the journey and lairage time prior to slaughter, and by scheduling and prioritising the slaughter of the animals.

The longer the on-farm fasting time, the higher the risk of excessive feed withdrawal time. Therefore, to mitigate the prolonged hunger during transport, on-farm feed withdrawal should be avoided, and the total feed withdrawal time should always be considered when the duration of the journey is considered.

Communication with live animals catching teams and hauliers and with the slaughterhouses or destination farm for planning and coordinating the arrival of live animals, will reduce transport duration by minimising loading times and waiting time upon arrival of live animals and mitigating prolonged hunger.

5.7.9.5. Corrective and mitigative measures

Rabbits subjected to feed withdrawal periods longer than 6 h will start to experience prolonged hunger and a negative affective state, which will increase with time so that by 12 h or more the welfare consequence of prolonged thirst is likely. To mitigate it, the total feed withdrawal time should not exceed 12 h.

Provision of feed during transport might correct or mitigate prolonged hunger. However, if animals are to be fed in containers, the systems for feeding must be designed in a way enabling all animals access to sufficient amounts for feed. If this is not the case, the risk of prolonged hunger is not reduced. Unless all animals can access feed from their original position in the container, the space allowance, including height, must enable animals to move freely within the container to access feed. This is not common practice today, and, if it would be the case, it might also increase the risk of injuries (see Section 5.7.2).

The only feasible mitigative measure is to unload the animals and provide feed or slaughter them as soon as possible after arrival.

5.7.10. Prolonged thirst

5.7.10.1. Description

An animal is thirsty when it experiences craving or urgent need for water, accompanied by anuneasy sensation (a negative affective state), and eventually leading to dehydration as metabolic requirements are not met.

If drinking water is not provided to rabbits and the only feed available is dry with a moisture content of less than 14%, voluntary intake drops to nil within 24 h (Lebas et al., 1986).

Trocino et al. (2003) suggested that for transport longer than 6–8 h, a progressive dehydration of rabbits could be hypothesised, since dressing percentage decreased while gut content remained stable at around 16% live weight from 4 h of transport onward.

Considering the available evidence, the AHAW Panel, based on expert opinion, concluded with 66–100% certainty (from likely to almost certain) that rabbits subjected to water deprivation periods longer than 12 h will experience prolonged thirst.

5.7.10.2. ABMs

De Jong et al. (2011) suggested that there are no indicators available, which will measure prolonged thirst in a feasible way during transport. A potential indicator for dehydration is dry skin, which may be identified by lifting the rabbit's skin to examine its elasticity. If the skin tends to remain pinched and wrinkled before returning to its natural position, it means that the rabbit is dehydrated. However, this is practically impossible to examine on live rabbits during transport, and not easy to evaluate post-mortem either. Therefore, no feasible ABMs for prolonged thirst in rabbits exist.

5.7.10.3. Hazards

Water deprivation for too long

As presented above, the hazard for prolonged thirst is the water deprivation that is performed routinely as a hygienic measure. Water deprivation can be performed at different points in time on the farm before catching of the animals before transport.

Considering that rabbits might be spending a considerable time in the transport containers before the journey starts and in lairage after the journey ends, the total time without water potentially becomes very long, resulting in an increased risk of prolonged thirst. Furthermore, providing water during the journey is not feasible, as elaborated in the section on prolonged hunger.

Deprivation of water for periods longer than 12 h leads to prolonged thirst leading to dehydration in the rabbit.

Hot weather during transport of animals without access to water worsens the animal welfare consequences. There is a cumulative effect of deprivation of water with high temperature and high stocking densities.

5.7.10.4. Preventive measures

Rabbits should have free access to water until they are caught and placed in containers.

To prevent rabbits experiencing prolonged thirst during transport, the total time of water deprivation should not exceed 12 h. This time should be considerably reduced during heat stress.

Provision of water during transport might prevent, correct or mitigate prolonged thirst. However, if animals are to be watered in containers, the systems for watering must be designed in a way enabling all animals access and ingest sufficient amounts. If this is not the case, the risk of prolonged thirst is not reduced. Unless all animals can access water from their original position in the container, the space allowance, including height, must enable animals to move freely within the container to access water. This is not common practice today and, if it would be the case, it might also increase the risk of injuries (see Section 5.7.2).

Communication with live animals catching teams and hauliers and with the slaughterhouses or destination farm for planning and coordinating the arrival of live animals, will reduce transport duration by minimising loading times and waiting time upon arrival of live animals and mitigating prolonged hunger.

5.7.10.5. Corrective and mitigative measures

The only feasible mitigative measure is to unload the animals and provide water or slaughter them as soon as possible.

5.8. Transport duration

For the overall assessment of transport duration, it is not only the journey duration (period the vehicle is in transit), but also the time the animals are in the containers and eventually the feed withdrawal period on farm that should be considered. Regardless of how optimal the conditions of the transport provided are, rabbits can potentially be exposed to a number of hazards during transport that might, either on their own or in combination, result in impaired welfare consequences. The exposure to these hazards ends only when the transport ends and the animals are uncrated from the containers. Any aversive effects associated with exposure overstimulation or to restriction of water and feed are likely to increase with transport duration and could interact with other factors, such as temperature, that might also change during a journey. The recommended maximum transport time is based on an overall assessment across the highly relevant welfare consequences, and is based on the scientific evidence combined with expert opinion. The recommendation for transport duration is based on the assumption that recommendations on microclimatic conditions and space allowance are followed.

Sensory overstimulation and motion stress: As soon as rabbits are crated and loaded into the vehicle, and during all time when the vehicle is moving, all rabbits are to some extent exposed to motion stress and often also, at least periodically, to sensory overstimulation. As a consequence of the vehicle motion, animals experience stress potentially leading to fatigue and negative affective states such as fear and distress. The duration of the welfare consequence depends on transport duration and onset of vehicle motion. Because of the constant presence of motion stress, it is not possible to estimate a temporal cut-off for onset of this welfare consequence after initiation of the journey.

Prolonged hunger: The welfare consequence prolonged hunger is regarded as highly relevant in the transit stage. Prevalence is expected to be high, as no studies have documented the successful feeding of rabbits in the vehicle during journeys. Depending on factors such as time off feed before journey start, rabbits may not be hungry during the initial phase of the journey, but hunger will develop over time. The duration of prolonged hunger depends on time of on-farm withdrawal and transport duration until the rabbits are uncrated, and severity is expected to increase with increasing duration, as the need for feed becomes more and more problematic for the animals. Prolonged hunger may lead to exhaustion and a weakened condition. It is concluded that rabbits subjected to total feed withdrawal periods longer than 6 h will start to experience prolonged hunger and a negative affective state, which will increase with time so that by 12 h or more the welfare consequence of prolonged hunger is from likely to almost certain (66–100% certainty).

Prolonged thirst: The welfare consequence prolonged thirst is regarded as highly relevant in the transit stage. Prevalence may be high as water is not provided to the animals. So far, no documentation for proper intake of water, even in journeys on vehicles fitted with drinkers, are available. Depending on factors such as time off water before journey start and/or microclimatic conditions before and during the journey, rabbits may not be thirsty during the initial phase of the journey, but thirst will develop over time. The duration of prolonged thirst depends on transport duration, and severity is expected to increase with increasing duration, as the need for water becomes more and more problematic for the animals. The available data do not allow a detailed determination of the interval between journey start and initiation of thirst, especially due to the lack of repeated sampling. It is concluded that rabbits subjected to water withdrawal periods longer than 6 h will start to experience prolonged thirst and a negative affective state, which will increase with time so that by 12 h or more the welfare consequence of prolonged thirst is from likely to almost certain (66–100% certainty).

Other summarising considerations: In addition to the welfare consequences summarised above, the risk of animals experiencing pain and/or discomfort, as well as the severity of it, will also increase with transport time. This may happen if the animals had a pre-existing painful condition. Even though this should not happen, it is not always possible to identify pathological conditions in rabbits while they are on the farm, as they are known to not show overt signs of, e.g. discomfort.

In addition, animals which did not show a health condition before the journey may get injured during the transport due to e.g. catching and crating, and the pain and discomfort from such conditions will continue, and likely worsen, until the animal can be uncrated. In this weakened state,

rabbits are often less able to cope with the extra challenges associated with transport, and their condition is likely to deteriorate with time and transport duration.

The pain and/or discomfort from both types of the above-mentioned health conditions are not expected to be prevalent, but for the affected animals the consequences may be severe, and will develop over time. The duration of these negative affective states will depend on transport duration, as they cannot be terminated until the journey is stopped and the birds are uncrated. During a journey, such health conditions may lead to suffering. It is however, not possible to establish a temporal cut-off for when pain and/or discomfort may start.

Evidence on continuous welfare consequences involving stress and negative affective states suggests that limiting journey time would reduce the exposure to the hazards of the transit stage. Scientific evidence on how the progressively developing welfare consequences change with increasing journey duration is limited. Based on the presence of the continuous welfare consequences combined with the limited data on the progressively developing welfare consequences, it is not possible to scientifically define a maximum journey duration that will not impair animal welfare. In order to take into account all welfare consequences, continuous as well as progressively developing, 12 h can be suggested as a limit to journey duration (including on-farm feed and water withdrawal) until being uncrated when animals are kept under the suggested microclimatic and space allowance conditions, and no water or feed is available to the animals during that period. Importantly, this does not imply that the welfare consequences necessarily occur after a journey duration of 12 h, or that no welfare consequences occur before 12 h, as there are many factors other than journey duration that affect the risk of welfare consequences during a journey.

To reduce the transport duration transport should be planned in advance from the stage of loading to arrival and uncrating (see Section 5.2.2). This includes the coordination of the different stages of the transport (e.g. itinerary for loading and unloading, location of any driver resting places/stops during the journey), estimation of their duration and time of arrival. Effective planning includes communication with live bird catching teams and hauliers and with the slaughterhouse or destination farm (European Commission, 2017a, 2018) for planning and coordinating the arrival of rabbits. The potential welfare consequences and their possible hazards (such as analysis of weather forecast) should be identified, and preventive and mitigating measures implemented. Contingency plans will help the driver and the transport company to ensure the security and the welfare of the animals in case of emergency and reduce transport time.

5.9. Iceberg indicators

Dead-on-arrival rate (DOA) is routinely recorded at rabbit slaughterhouses, as it is considered an important indicator (i.e. iceberg indicator) providing information on transport conditions. According to EFSA (2004), rabbits' DOA rate ranges between 0.1% and 0.4%. In Italy, Petracci et al. (2010) reported mean transport-related mortality of 0.09%, ranging from 0% to 1.3%, in 2006. When compared with findings recorded in Italy in other species, preslaughter mortality of rabbits was slightly higher than that recorded in swine and bovine, while it was much lower compared to mortality rates reported in poultry which are transported using similar procedures and equipment. More recently Caucci et al. (2018) performed a retrospective analysis on rabbit batches transported in a 3-year period to a major abattoir of Northeast Italy. Average DOA was 0.08% for fattening rabbits, ranging from 0.0% to 2.0%. In transports with at least one dead rabbit the average DOA was 0.13%. Fourteen percent of transports of breeding rabbits (n = 65) showed at least one dead rabbit and a mean DOA value of 1.79%. Considering journeys with at least one dead rabbit, a higher DOA for breeding rabbits (1.79%) compared to fattening (0.13%) was observed (Caucci et al., 2018). In the Czech Republic, the overall transport-related mortality of rabbits (meat production) between 2009 and 2016 was 0.19% (Voslarova, 2018). Death losses in rabbits exceeded those in most other species of animals kept for meat production and transported for slaughter in the Czech Republic. In fact, higher mortality rates (0.37%) were only found in broiler chickens. Otherwise, the preslaughter mortality (during transport and at lairage) of rabbits was greater than that recorded in swine (0.07%) and bovine animals (0.02%), as well as in poultry with the exception of broiler chickens, that is ducks (0.08%) and turkeys (0.15%). We could speculate that lower DOAs reported in northeast Italy may be due to the enforcement of local legislation on the transport of rabbits. Nevertheless, a positive finding is that a significant negative correlation between mortality rates and monitored years in rabbits was found in the Czech study, indicating a general decreasing trend over time between 2009 and 2016 in rabbit losses.

Impact of heat stress on DOA

Petracci et al. (2010) identified temperatures higher than 22.6°C to be risk factors for DOA in fattening rabbits. Most importantly, temperatures above 22.6°C exhibited almost double the mortality risk in comparison with the reference category (12.4–17.5°C). These results concluded that heat stress is major cause of death in rabbits during transport, even if the majority of the batches were transported during the night or early morning (59.2%) and slaughtered during the morning, reducing the exposure of rabbits to the maximum temperatures during the hottest daytime hours (Petracci et al., 2010). High environmental temperature was also reported by Voslarova et al. (2018) as a risk factor in the Czech Republic. Journeys carried out at temperatures above 20°C were associated with higher death losses (0.15%). They were significantly higher than the mortality of rabbits related to transport under temperatures ranging from 10°C to 14.9°C (0.07%).

Impact of cold stress on DOA

Petracci et al. (2010) identified temperatures lower than 7.3°C as a risk for higher DOA rates in fattening rabbits. Their results showed that temperatures below 7.3°C can increase the risk of death by 28%. Voslarova et al. (2018) reported journeys carried out under temperatures below −5°C to be associated with significantly higher death losses (0.17%) than journeys carried out under temperatures ranging from 10°C to 14.9°C (0.07%), that was the temperature category associated with the lowest losses.

Impact of restriction of movement on DOA

Caucci et al. (2018) reported significantly increased DOAs of fattening rabbits at a stocking density higher than 29.3 kg/crate. The highest number of batches at high stocking density was found in winter (61% of all batches). Batches with high stocking density showed a higher DOA even in winter, suggesting that the reduction of space availability does not protect against low temperatures. In breeding rabbits no significant association between DOA and stocking density was detected. However, an increased DOA was observed in batches transporting more than $\approx$ 7 rabbits/crate.

Impact of transport duration on DOA

Voslarova et al. (2018) found significantly greater losses in rabbits transported over longer distances. Mortality rates ranged from 0.02% in rabbits transported over distances of less than 50 km to 0.29% in rabbits transported over distances exceeding 400 km. Despite less than half the rabbits being transported over distances of more than 100 km, such journeys accounted for 63.36% of death losses. According to Petracci et al. (2010), both increasing journey and lairage duration dramatically increase the risk of death during the preslaughter period. Rabbits belonging to batches either transported or laired for a long (above 308 min) time had a roughly threefold higher risk of death compared with reference category (< 103 min). Similarly, Caucci et al. (2018) found both journey and lairage durations to have a significant effect on DOA. Regarding travel duration, transports longer than 3 h increased significantly DOA by $\approx$ 40% compared to transports shorter than 1 h. A lairage longer than 7 h doubled the DOA compared to a lairage shorter than 2 h. The interaction between travel and lairage duration showed that a journey shorter than 1 h can significantly reduce DOA associated with a long lairage.

Batch size

Voslarova et al. (2018), found a significantly increased risk of DOA to be associated with shipments in which 500 and more rabbits were delivered per batch. Petracci et al. (2010) found not only a higher mortality risk but also an increased risk of bruising in very large batches (> 3,681 rabbits). They attribute this to the human factor, related to rough handling of catchers being less careful when dealing with a large load. However, Caucci et al. (2018) found higher values of DOA in both very small and very large batches in fattening rabbits, i.e. significantly increased DOAs were observed in batches with below 1,201 rabbits or more than 3,508 rabbits.

To be a valid iceberg indicator, DOA should be collected and monitored for each individual transport. Rabbits may die during transport due to factors that are not attributable directly to the transport conditions. Therefore, the cause of DOA might not need to be investigated for each individual transport, but if it exceeds a certain level, that might be attributed to the transport conditions. The AHAW Panel suggested that this level might be 0.1% for rabbits.

6. Uncertainty analysis results

Table 33 shows the statements contained in the well-defined questions of interest formulated for each key conclusion and for which experts were asked to provide their individual judgements, along with the consensus certainty range reflecting their collective uncertainty about the statement.

Table 33: Uncertainty assessment results for the conclusions of each welfare consequence

Animal category	Welfare consequence	Hazard	Statement	Certainty range
Domestic birds	Restriction of movement	Insufficient space allowance	90% or more of all domestic birds transported in 2022 will not experience restriction of movements when transported in cages of sizes equal to or larger than those presented in Table 11	90–100%
Domestic birds	Restriction of movement	Insufficient height	90% or more of all domestic birds transported in 2022 will not experience restriction of movements when transported in cages of heights equal to or larger than those presented in Table 12	90–100%
Rabbit	Restriction of movement	Insufficient height	90% or more of all slaughter rabbits up to 3 kg transported in 2022 will not experience restriction of movements when transported in a crate height of 35 cm	66–100%
Rabbit	Restriction of movement	Insufficient height	90% or more of all slaughter rabbits of 4.5–6 kg transported in 2022 will not experience restriction of movements when transported in a crate height of 40 cm	66–100%
Domestic birds	Heat stress	Too high apparent equivalent temperature (AET), enthalpy comfort index (ECI)	90% or more of all domestic birds transported in 2022 will not experience heat stress when the AET value is below 40 or the ECI is below 48 kJ/kg	90–100%
Domestic birds	Heat stress	Too high apparent equivalent temperature (AET), enthalpy comfort index (ECI)	90% or more of all domestic birds transported in 2022 will experience heat stress when the AET value is above 65 or the ECI is above 57.6 kJ/kg	90–100%
End-of-lay hens	Heat stress	Too high apparent equivalent temperature (AET), enthalpy comfort index (ECI)	The same conclusions extracted for domestic birds regarding heat stress above/below certain values for AET and ECI can be applied to EoL hens	66–100%
Domestic birds	Cold stress	Too low effective temperature	90% or more of all domestic birds transported in 2022 at temperatures below 10°C in the container will experience cold stress	66–100%
End-of-lay hens	Cold stress	Too low effective temperature	90% or more of all End of laying hens transported in 2022 at temperatures below 18°C will experience cold stress	66–100%
Day-old chicks	Heat stress	Too high body temperature	90% or more of all DOC transported in 2022 will experience heat stress when body temperature goes above 41°C	90–100%
Day-old chicks	Heat stress	Too high effective temperature	90% or more of all DOC transported in 2022 will experience heat stress when the effective temperatures in the container is above 35°C	66–100%

Animal category	Welfare consequence	Hazard	Statement	Certainty range
Day-old chicks	Cold stress	Too low body temperature	90% of more of all DOC transported in 2022 will experience cold stress when body temperature goes below 40°C	90–100%
Day-old chicks	Cold stress	Too low effective temperature	90% of more of all DOC transported in 2022 will experience cold stress when the effective temperature in the container is below 30°C	66–100%
Rabbit	Heat stress	Too high temperature–humidity index	90% or more of all rabbits transported in 2022 will experience heat stress when the THI is above 28.9	66–100%
Rabbit	Cold stress	Too low effective temperature	90% or more of all rabbits transported in 2022 will experience cold stress when the effective temperature is below 10°C	66–100%
Domestic birds	Prolonged hunger	Too long feed withdrawal period	90% or more of all domestic birds transported in 2022 will experience prolonged hunger when subjected to feed withdrawal periods longer than 6 h	50–100%
Domestic birds	Prolonged hunger	Too long feed withdrawal period	90% or more of all domestic birds transported in 2022 will experience prolonged hunger when subjected to feed withdrawal periods longer than 12 h	90–100%
End-of-lay hens	Prolonged hunger	Too long feed withdrawal period	90% or more of all End-of-lay hens transported in 2022 will experience prolonged hunger when subjected to feed withdrawal periods longer than 10 h	66–100%
Ducks, geese, quails, game birds	Prolonged hunger	Too long feed withdrawal period	90% or more of all ducks, geese, quails and game birds transported in 2022 will experience prolonged hunger when subjected to same periods as domestic birds	66–100%
Day-old chicks	Prolonged hunger and thirst	Too long feed withdrawal/water deprivation period	90% or more of all DOC transported in 2022 will experience prolonged hunger and thirst when subjected to feed and water withdrawal periods longer than 48 h	90–100%
Rabbit	Prolonged hunger	Too long feed withdrawal period	90% or more of all rabbits transported in 2022 will experience prolonged hunger when subjected to feed withdrawal periods longer than 6 h	50–100%
Rabbit	Prolonged hunger	Too long feed withdrawal period	90% or more of all rabbits transported in 2022 will experience prolonged hunger when subjected to feed withdrawal periods longer than 12 h	66–100%
Domestic birds	Prolonged thirst	Too long water deprivation period	90% or more of all domestic birds transported in 2022 will experience prolonged thirst when subjected to water deprivation for more than 6 h	66–100%
Domestic birds	Prolonged thirst	Too long water deprivation period	90% or more of all domestic birds transported in 2022 will experience prolonged thirst when subjected to water deprivation for more than 12 h	90–100%
End-of-lay hens	Prolonged thirst	Too long water deprivation period	90% or more of all end-of-lay hens transported in 2022 will experience prolonged thirst when subjected to water deprivation for more than 6 h	66–100%

Animal category	Welfare consequence	Hazard	Statement	Certainty range
End-of-lay hens	Prolonged thirst	Too long water deprivation period	90% or more of all end-of-lay hens transported in 2022 will experience prolonged thirst when subjected to water deprivation for more than 12 h	90–100%
Ducks, geese, quails, game birds	Prolonged thirst	Too long water deprivation period	90% or more of all ducks, geese, quails and game birds transported in 2022 will experience prolonged thirst when subjected to same periods as domestic birds	66–100%
Rabbit	Prolonged thirst	Too long water deprivation period	90% or more of all rabbits transported in 2022 will experience prolonged thirst when subjected to water deprivation for more than 6 h	66–100%
Rabbit	Prolonged thirst	Too long water deprivation period	90% or more of all rabbits transported in 2022 will experience prolonged thirst when subjected to water deprivation for more than 12 h	66–100%
Domestic birds	Handling stress	Rough handling	90% or more of all domestic birds transported in 2022 will experience handling stress in inversion compared to when handled in an upright position	90–100%
Domestic birds	Injuries	Rough handling	The number of domestic birds transported in 2022 in inversion that will experience injuries will be larger compared to when handled in an upright position	66–100%
Day-old chicks	Handling stress	Rough handling	90% or more of day-old chicks transported in 2022 will experience handling stress with changes of velocity exceeding 0.4 m/s, drop height above 280 mm and speed of belts over 27 m/min	66–100%
Domestic birds	NA	Iceberg indicator (Too high DoA)	90% or more of all transports of domestic birds with DoA higher than 0.1% is due to welfare consequences experienced during transport	66–100%
Day-old chicks	NA	Iceberg indicator (Too high DoA)	90% or more of all transports of day-old chicks with DoA higher than 0.1% is due to welfare consequences experienced during transport	66–100%
Rabbit	NA	Iceberg indicator (Too high DoA)	90% or more of all transports of rabbits with DoA higher than 0.1% is due to welfare consequences experienced during transport	66–100%

7.   Conclusions

7.1.   Conclusions for domestic birds

1) The transport of domestic birds consists of five stages: (1) Preparation, which includes: planning of the journey, preparation of the birds and assessment of fitness for transport; (2) Loading, which includes: catching the birds, placing them in containers (crating) and loading of containers onto the vehicle; (3) Journey, which includes: the movement of birds by vehicle until the place of destination is reached; (4) Arrival, which includes: the period from arrival of the vehicle, unloading of the containers from the vehicle and waiting period; and (5) Uncrating, which includes the removal of the birds from the containers.

2) The highly relevant welfare consequences for transport of domestic birds are handling stress, injuries, restriction of movement, sensory overstimulation, motion stress, heat stress, cold stress, prolonged hunger and prolonged thirst.

3) During the transport domestic birds are exposed to several hazards at the same time. The presence and severity of the welfare consequences they are exposed to mainly depend on the conditions of the transport and the way welfare consequences can be mitigated and/or the way hazards can be corrected/prevented.

7.1.1. Conclusions related to fitness for transport

1) Pre-existing conditions of poor health are more likely to increase the severity of the welfare consequences transported domestic birds are exposed to during the journey and the animals are more likely to die during the journey.

2) The main conditions rendering domestic birds unfit for transport are: evident signs of illness, emaciation, severe lameness (unable to stand or walk more than a few steps), open wounds and prolapse, fractures (legs, wings, etc.), dislocations and poor feather cover or wet plumage in low effective temperature. Wet plumage is not a risk for ducks and geese.

3) End-of-lay hens with poor feather cover are in any case unfit to travel if they are to be transported in cold weather without the application of preventive and corrective measures discussed in Sections 3.7.8.5 and 3.7.8.6.

7.1.2. Conclusions related to handling stress

1) Handling stress is a welfare consequence inherent to transport, and all domestic birds are likely to experience this welfare consequence, mainly during catching and crating of the loading stage, during unloading at the arrival stage and during the uncrating stage.

2) The ABMs escape attempts, piling up and distress calls are sensitive and specific to assess handling stress in a group of birds during catching and crating of the loading stage, during unloading at the arrival stage and during the uncrating stage.

3) Rough handling will increase the severity of handling stress compared to gentle handling.

4) The AHAW panel concluded with 90–100% certainty (from very likely to almost certain) that inversion increases the severity of handling stress compared to handling birds in an upright position.

5) Mechanical catching can reduce the severity of handling stress compared to manual catching and inversion, if it is operated correctly.

6) Handling stress cannot be prevented or corrected, only mitigated.

7.1.3. Conclusions related to Injuries

1) Injuries lead to negative affective states such as pain and discomfort in birds, and the severity of these welfare consequences can range from moderate to very high.

2) Injuries are usually inflicted during catching and crating and the welfare consequences prevail throughout the stages of transport.

3) Owing to the bone fragility and susceptibility to fracture, the catching, crating and uncrating of end-of-lay hens represent a particularly high risk of injuries.

4) ABMs of injuries, such as hanging, non-functional limbs, severe lameness, protruding bones, bruises and wounds, are highly to moderately sensitive and specific for the assessment during catching and crating of the loading stage, during unloading at the arrival stage and during the uncrating stage. Other injuries are mainly detected post-mortem at the slaughterhouse.

5) The prevalence and severity of injuries depend on many factors, but rough handling, due to lack of staff skills and training, is the main hazard.

6) Rough handling will increase the risk of injuries compared to gentle handling.

7) The AHAW panel concluded with 66–100% certainty (from likely to almost certain) that inversion and carrying birds to the container by the legs in inverted position will stimulate wing flapping and increase the risk of injuries (dislocated joints, fractures in legs or wings and bruises) compared to handling birds in an upright position.

8) Mitigating measures for handling stress will also reduce the risk of injuries.

9) Mechanical catching can potentially reduce the risk of injuries compared to manual catching and inversion, if it is operated correctly.

10) Design, operation and maintenance of equipment such as transport containers, catching machines, forklifts and conveyor belts will influence the risk of injuries to birds.

7.1.4. Conclusions related to Restriction of movement

1) Restriction of movement is a welfare consequence that is inherent to the transport situation, and all domestic birds will experience this welfare consequence as they are confined in containers.

2) Restriction of movement is considered severe when birds are not provided with enough floor space to sit all at the same time without overlapping, or are not provided with enough height to sit with their heads raised without touching the ceiling. Restriction of movement is less severe when birds have enough space to shuffle around within the container.

3) To assess restriction of movement in birds the ABMs 'sitting posture' and 'head posture' are the most specific and sensitive during loading and during unloading at arrival. However, assessment of these ABMs is not currently feasible during the journey stage.

4) Insufficient space allowance and insufficient height of container are the hazards for restriction of movement, also for other welfare consequences such as heat stress. However, high space allowance and height may increase the risk of injuries to the birds.

5) The generic allometric equation 'space allowance (cm^2/bird) = $290 \times$ live weight ($kg^{2/3}$) can be used to calculate the minimum required floor space allowance for most bird categories to adopt a sitting position and have the possibility to shuffle around and to provide some extra space between animals to ensure proper ventilation. For pullets and laying hens up to 2 kg, especially when well feathered, more floor space is needed than predicted using the allometric equation and therefore the planimetric measurements are preferable. Recommended minimum space allowances for the different animal categories are reported in Table 11 (Section 3.7.3.4). The AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) these space allowances will mitigate the welfare consequence of restriction of movement.

6) If domestic birds within the containers are able to keep their heads in a natural raised position when sitting and when changing their position, restriction of movement might be mitigated without increasing the risk of injuries to the birds. However, there is a lack of scientific knowledge on the height requirements of various domestic birds. Recommended minimum heights of the containers are provided in Table 12 (Section 3.7.3.4) for the different animal categories.

7.1.5. Conclusions related to Sensory overstimulation

1) Sensory overstimulation is a welfare consequence inherent to all stages of the transport and will affect all domestic birds.

2) During commercial transport, birds are subjected to stimulation of the visual, auditory, chemical, sensory somatosensory and vestibular systems. Overstimulation of any system or combination of systems will result in stress and/or negative affective states such as fear, and/or discomfort.

3) Escape attempts and distress calls are sensitive and specific ABMs to assess sensory overstimulation. However, these ABMs cannot be currently assessed during the journey, only during loading and at arrival.

4) Birds may be exposed to sudden unfamiliar changes in light or sound intensity and duration associated with vehicle movement and the travel environment.

5) Acute or sudden auditory or visual stimuli may become recurrent and continuous and will constitute a chronic over stimulation.

6) Olfactory and gustatory systems of the birds may be overstimulated by exposure to exhaust fumes and other general pollutants (e.g. ammonia).

7) In addition to the primary problem of sensory overstimulation and the induced aversion in the transported birds, this welfare consequence can exacerbate the effects of other welfare consequences such as handling stress, injuries, thermal stress, restriction of movement and prolonged hunger and thirst.

8) The prevention or mitigation of sensory overstimulation can only be achieved by planning of the transport, changes to loading equipment and vehicle design and structure or by attention to the catching and driving style and vehicle operation and maintenance and quality of the road.

7.1.6. Conclusions related to Motion Stress

1) The overstimulation of the somatosensory and vestibular systems will result in motion stress and may have a significant impact upon the welfare of domestic birds during the journey.
2) No feasible ABMs for assessing motion stress during transport in birds transported in containers have been identified.
3) The severity of the motion stress experienced will depend upon the frequency and intensity of acceleration, vibration and vehicle ride resulting from movement and operation of the vehicle.
4) Exposure to acceleration and vibration will induce postural instability, increase the requirement for muscle activity to restore stability and will increase the risk of injuries and fatigue.
5) These stimuli will be further dependent upon vehicle design and structure, container design and operation and factors such a vehicle speed, driving style and road type and surface.
6) The risk of motion stress may be reduced by attention to vehicle suspension, tire and vehicle maintenance, driving style and driver education and training.

7.1.7. Conclusions related to Heat Stress

1) Heat stress is a highly relevant welfare consequence when transporting domestic birds, with a higher prevalence in summer compared to other seasons. Domestic birds can experience heat stress during the entire journey or for parts of the journey. If continuously present, the severity will increase over time as animals experience distress and eventually fail to cope and die.
2) Panting is a highly specific and sensitive ABM of heat stress.
3) High effective temperature induces thermoregulatory responses in the animals and is the principal hazard for heat stress.
4) The effective temperature experienced by the animal is affected by many factors such as ambient temperature, humidity and radiation, and air movement. However, in the context of transport of animals in containers, effective temperature is considered to be primarily determined by dry-bulb temperature and humidity (water vapour content).
5) Effective temperature can be divided into safe, alert or danger zones based upon thermoregulatory demands and physiological responses. In the safe zone animals will not experience heat stress because they require no or minimal thermoregulatory effort. In the alert zone, animals are at risk of heat stress because they require increasing thermoregulatory efforts. In the danger zone, animals will experience heat stress because the mechanisms to cope with it will become less effective.
6) Several indices based on dry-bulb temperature and relative humidity have been developed to measure high effective temperature inside the transport containers. One of these is the Temperature-Humidity Index (THI) that is very commonly used for livestock. Two other indices have been validated for poultry: the Apparent Equivalent Temperature (AET) and the Enthalpy Comfort Index (ECI) that also combine dry-bulb temperature and relative humidity.
7) If AET is used, for the combinations of dry-bulb temperature and relative humidity resulting in an AET value below 40, the AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) that domestic birds will not experience heat stress during transport (safe zone). Between AET values of 40 and 65, there will be an increasing risk of heat stress (alert zone) and above an AET of 65, birds will experience heat stress (danger zone). In order to calculate AET the equation reported in the text in Section 3.7.7.2 can be used. The combinations of dry-bulb temperature and relative humidity giving rise to AET values for the safe, alert and danger zones are graphically displayed in Figure 23 in Section 3.7.7.3.
8) The enthalpy comfort index (ECI) combines the input variables of the dry-bulb temperature, the relative humidity of the air and the local barometric pressure. The equation is reported in the text in Section 3.7.7.2. If ECI remains below 48.0 kJ/kg, the AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) that domestic birds will not

experience heat stress during transport (comfort zone). If ECI exceeds this threshold, there will be an increasing risk of heat stress (warning zone). Above 57.6 kJ/kg the birds will experience heat stress (critical zone).

9) For end-of-lay hens, the AHAW panel concluded with 66–100% certainty (from likely to almost certain) that similar ranges of temperature and humidity of those recommended for domestic birds can be used.

10) In passively ventilated vehicles, there is an uneven distribution of thermal conditions within the load and the risk of heat stress will be highest close to the final air exit points (e.g. upper front and central areas).

11) Heat stress may also affect a proportion of birds in cold weather in passively ventilated vehicles especially when the vehicle is stationary and distribution of thermal conditions within the load is uneven with the upper locations being warmer.

12) Thermal comfort in all birds during the journey can be achieved in fan ventilated vehicles, where the movement of air into, within and out of the vehicle container is controlled by a combination of suitably positioned mechanical fans of sufficient capacity, and natural apertures which enhance established natural air pressure gradients.

13) Controlled environment or air-conditioned vehicles can regulate or modify internal thermal conditions by appropriate heating or cooling. They have the major advantage that the internal environment may be controlled regardless of external weather or thermal conditions and does not rely upon vehicle movement.

7.1.8. Conclusions related to Cold Stress

1) Cold stress is a highly relevant welfare consequence when transporting domestic birds, with prevalence being higher in the winter season. Domestic birds can experience cold stress during the entire journey or for parts of the journey. Severity will increase over time as animals experience distress and eventually fail to cope and die.

2) Huddling, fluffing up of feathers, shivering and cloacal temperature are highly sensitive ABMs of cold stress.

3) Low effective temperature induces thermoregulatory responses in the animals and is the principal hazard for cold stress.

4) Cold stress is mainly induced by low dry-bulb temperature, although it is also influenced by air movement. Humidity does not have a big impact on thermoregulation demands for cold stress. Therefore, conclusions for cold stress are based on dry-bulb temperature (and not on indexes combining temperature and humidity).

5) Domestic birds do not experience cold stress when exposed to effective temperatures within the comfort zone. Below the comfort zone, with small decreases in effective temperature, the birds can still cope and are most likely not experiencing cold stress. When temperatures decrease further, thermoregulatory demands will increase, and the risk of cold stress increases. This is a continuous process. At the critical temperature of the thermoneutral zone, thermoregulatory mechanisms become less effective and birds will experience cold stress.

6) The AHAW panel concluded with 66–100% certainty (from likely to almost certain) that at dry-bulb temperatures below 10°C, the bird's mechanisms to cope with cold stress will become less effective and the birds will experience cold stress.

7) Also, above 10°C, there might be a risk of cold stress in wet birds (following, e.g. rain/snow ingress, melted ice, road spray at air inlets, wetting during catching and loading) and in birds located at cold air inlets.

8) In passively ventilated vehicles, there is an uneven distribution of thermal conditions within the load and the risk of cold stress will be greatest close to air entry points.

9) For end-of-lay hens, since they have generally poorer feather cover and limited metabolic capacity and reserves, the lower limit for temperature is higher than for other categories of poultry and estimated to be at 18°C. The AHAW panel concluded with 66–100% certainty (from likely to almost certain) that if the temperature falls below this threshold, the hens' mechanisms to cope with cold stress will become less effective and the hens will experience cold stress. If the dry-bulb temperature in the containers remains above this threshold, end-of-lay hens will not experience cold stress during transport.

7.1.9. Conclusions related to Prolonged Hunger

1) The welfare consequence of prolonged hunger might affect all birds transported in containers due to the current practices of feed deprivation both on-farm, during the journey itself and upon arrival.
2) No feasible ABMs for assessing prolonged hunger during transport of birds in containers have been identified.
3) Deprivation of feed for too long is the main hazard for prolonged hunger. The length of the feed withdrawal time before catching and crating affects the risk of animals experiencing prolonged hunger during transport.
4) The total feed withdrawal duration starts when feed is removed on farm and ends when all animals are removed from containers following unloading from the vehicle and fed or slaughtered.
5) In broilers, the risk of experiencing hunger is likely to increase shortly after feed withdrawal, as they are bred to have a high motivation to feed and are used to freely available feed.
6) Domestic birds subjected to feed withdrawal periods longer than 6 h will exhaust crop feed stores and liver glycogen reserves and therefore the AHAW Panel concluded with 50–100% certainty (more likely than not) that they will experience prolonged hunger after this time period.
7) The AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) that domestic birds subject to feed withdrawal periods longer than 12 h will experience prolonged hunger and that these periods are also associated with intestinal cell breakdown, which is detrimental to their health and welfare.
8) End-of-lay hens are still productive birds laying eggs, which requires considerable metabolic effort and feed intake. Having laid almost one egg/day for a period of a year or more they are metabolically exhausted with few body reserves. Furthermore, the risk of experiencing hunger is higher compared to other bird categories.
9) For end-of-lay hens, the AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that, under thermoneutral conditions, they will experience prolonged hunger after feed withdrawal of 10 h.
10) As hens are at greater risk of experiencing cold stress, particularly when poorly feathered, a lack of feed will exacerbate this risk, and likewise exposure to cold will increase hunger.
11) For ducks, geese, quails and game birds, there is limited scientific evidence of the consequences of feed withdrawal. The AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that their hunger experience is similar to domestic birds.
12) There is no scientific evidence of a welfare benefit of fasting domestic birds before transport.

7.1.10. Conclusions related to Prolonged Thirst

1) The welfare consequence of prolonged thirst might affect all birds transported in containers due to the current practices of water deprivation during the journey time and upon arrival.
2) No feasible ABMs for assessing prolonged thirst during transport in birds transported in containers have been identified.
3) Deprivation of water for too long is the main hazard for prolonged thirst.
4) Birds require water in order to digest feed.
5) Considering the available evidence, the AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that domestic birds subjected to water deprivation periods longer than 6 h will experience prolonged thirst, demonstrated by the increase of plasma chloride as sign of dehydration.
6) Domestic birds subjected to water withdrawal periods longer than 12 h may show an increase in plasma creatinine; the AHAW Panel therefore concluded with 90–100% certainty (from very likely to almost certain) that they experience prolonged thirst.
7) Considering the available evidence, the AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that end-of-lay hens subjected to water deprivation periods longer than 6 h will experience prolonged thirst as demonstrated by changes in behaviour including an increase in levels of redirected aggression towards other hens.
8) End-of-lay hens subjected to water withdrawal periods longer than 12 h have increased levels of blood sodium and are highly motivated to drink and work hard to access water.

The AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) that they experience prolonged thirst.

9) If temperatures are above the thermal comfort zone, the evaporative heat loss through panting induces dehydration and advances the onset of prolonged thirst.

10) For ducks, geese, quails and game birds, there is limited scientific evidence of the consequences of water withdrawal; the AHAW panel therefore concluded with 66–100% (from likely to almost certain) that their thirst experience will be similar to other domestic poultry.

11) There is no scientific evidence of a welfare benefit of water deprivation of domestic birds before transport.

7.1.11. Conclusions related to maximum transport duration

1) The transport duration includes not only the journey duration itself (period the vehicle is in transit), but also the time the animals are in the containers and the prior feed withdrawal period on farm (if done).

2) With increasing journey time, and from the very start of the transport (catching and crating), animals are exposed for longer to a number of highly relevant welfare consequences (e.g. sensory overstimulation, motion stress, restriction of movement) that are stressful. These welfare consequences are present throughout the duration of the transport until birds are uncrated.

3) In addition, the risk of occurrence of new health conditions caused by events during transport (e.g. injuries, locomotory disorders) will increase with journey duration, as will the duration of potential suffering associated with these.

4) Evidence on continuous welfare consequences involving stress and negative affective states (e.g. fear) suggest that limiting transport time would reduce the exposure to the associated hazards.

5) Direct evidence on how the progressively developing welfare consequences change with increasing transport duration is limited.

6) Based on the presence of the continuous welfare consequences from the start of any transport, combined with the scarce data on the progressive welfare consequences, it can be concluded that any duration of transport will negatively affect animal welfare.

7) The severity of welfare consequences during the journey stage of transport will depend on the exact conditions pertaining to an individual journey (e.g. microclimatic conditions, space allowance and road conditions). These hazards potentially interact. The exposure to these hazards will continue at least as long as the journey continues.

8) Domestic birds cannot be fed and provided with water in the containers. After 12 h from food withdrawal, birds will experience prolonged hunger and thirst. Therefore, transport duration (including on-farm feed and water withdrawal until being uncrated and slaughtered or provided feed) exceeding 12 h will compromise animal welfare.

9) In end-of-lay hens, transport duration exceeding 10 h (including on-farm feed and water withdrawal until being uncrated and slaughtered) will compromise animal welfare, as end-of-lay hens will experience prolonged hunger and thirst.

10) The maximum transport duration times provided in 7 and 8 for domestic birds and end-of-lay hens do not imply that no welfare consequences occur before this time, as there are many factors other than transport duration that affect the risk of welfare consequences during a journey.

11) Related to thermal stress, birds will not experience heat stress if they travel in their safe zone (AET below 40 or ECI below 48.0 kJ/kg). If instead they travel in the alert zone (AET) or warning zone (ECI), the risk of compromised welfare is minimised for journey durations up to 4 h.

12) Planning, coordination and communication among the different actors involved with transport (e.g. live bird catching teams, hauliers, drivers, slaughterhouse or destination farm staff) is crucial to reduce transport duration by minimising loading times and waiting time upon arrival of live birds.

7.1.12. Conclusions related to iceberg indicators

1) Death on arrival (DOA) is an iceberg indicator for the assessment of welfare during transport and is shown to have a higher relationship with transport conditions than with farming situations.
2) There is a large variation in the average DOA between and within bird species and categories.
3) From the animal welfare point of view, the target DOA should be 0%. However, birds might die during transport due to factors that are not attributable directly to the transport conditions, e.g. breed.
4) The AHAW Panel concluded with 66–100% certainty (likely to almost certain) that DOA higher than 0.1% might be due to welfare consequences experienced during transport.
5) The main welfare consequences leading to DOA are heat stress, cold stress as well as the conditions of the animals (unfitness for transport).
6) The longer the transport duration the longer the animals are experiencing these welfare consequences and the higher the risk of DOA.

7.2. Conclusions for day-old chicks

1) The transport of day-old chicks (recently hatched chickens of less than 72 h) consists of five stages: (1) Preparation that includes planning of the journey, preparation of the chicks and assessment of fitness for transport; (2) Loading that includes catching the chicks, placing them in transport boxes and moving the boxes onto the vehicle; (3) Journey that includes the movement of birds by vehicle until the place of destination is reached; (4) Arrival that includes the period from arrival of the vehicle, unloading of the containers from the vehicle and waiting period; and (5) Uncrating that includes the removal of the birds from the transport containers.
2) The highly relevant welfare consequences for transport of day-old chicks are handling stress, sensory overstimulation and motion stress, heat stress, cold stress, prolonged hunger and prolonged thirst.
3) During the transport day-old chicks are exposed to several hazards at the same time. The presence and severity of the welfare consequences they are exposed to mainly depend on the conditions of the transport and the way welfare consequences can be mitigated and/or the way hazards can be corrected/prevented.

7.2.1. Conclusions related to fitness for transport

1) Pre-existing conditions of poor health are more likely to increase the severity of the welfare consequences transported day-old chicks are exposed to during the journey and the animals are more likely to die during the journey.
2) The main conditions making day-old chicks unfit for transport are: poor quality (this will occur when their activity is weak, when they are dirty and wet, present body with swallowed large yolk and rather hard to touch, closed eyes, infected legs, navel not closed and discoloured, large remaining membrane and very large remaining yolk), inability to stand, fractures and dislocations.

7.2.2. Conclusions related to handling stress

1) Handling stress is a welfare consequence inherent to transport, and all day-old chicks hatched in a hatchery are likely to experience this welfare consequence when put manually or automatically in boxes, loaded and unloaded from a vehicle.
2) ABMs for handling stress such as 'posture and orientation on conveyor belt', 'distress calls', 'escape attempts' and 'falling on the floor' are sensitive and specific for assessment while chicks are put in the boxes.
3) The main hazards for handling stress are 'Change in velocity, drop height acceleration and speed of conveyor belts', 'poor design of the system components' and 'rough manual handling'.
4) The AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that changes of velocity that exceed 0.4 m/s, a drop height above 280 mm and speed of belts

over 27 m/min will generate handling stress. There are no published data about specifications to completely avoid handling stress.

5) On-farm hatching will avoid handling stress as well as the other welfare consequences associated with transport.

7.2.3. Conclusions related to Sensory overstimulation and Motion stress

1) During commercial transport, day-old chicks are subjected to stimulation of the visual, auditory, chemical, sensory somatosensory and vestibular systems. Overstimulation of any system or combination of systems will result in stress and/or negative affective states such as fear, and/or discomfort.

2) 'Distress calls' and 'escape attempts' are ABMs that can be used to assess sensory overstimulation and motion stress. However, these ABMs cannot be assessed during the journey, only during loading and at arrival.

3) Chicks may be exposed to sudden changes in light or sound intensity and duration associated with handling, loading and vehicle movement leading to sensory overstimulation and motion stress.

4) The severity of the motion stress experienced will depend upon the frequency and intensity of acceleration, vibration and vehicle ride resulting from movement and operation of the vehicle.

5) The prevention or mitigation of sensory overstimulation and motion stress can only be achieved by planning of the transport, changes to loading equipment and vehicle design and structure or by attention to the catching and driving style and vehicle operation and maintenance and quality of the road.

7.2.4. Conclusions related to Heat Stress

1) Day-old chicks behave as poikilotherms and cannot regulate their body temperature by themselves. Appropriate effective temperature is therefore required to keep the body temperature between 40°C and 41°C during transport.

2) The AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) that day-old chicks will experience heat stress if the body temperature exceeds 41°C.

3) ABMs to assess heat stress such as 'respiration frequency', 'mean surface body temperature' and 'cloacal temperature' are sensitive and specific for assessment.

4) Too high temperature is the principal hazard for heat stress. In the context of transport of day-old chicks in containers, scientific information related to heat stress primarily referred to dry-bulb temperature (not accounting for humidity). Therefore, the conclusions related to temperatures refer to dry-bulb temperatures.

5) The upper limit of the comfort zone of day-old chicks is estimated to be 35°C (at the level of the chicks). The AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that, above a dry-bulb temperature of 35°C (near to the chicks), the bird's mechanisms to cope with heat stress will become less effective and the chicks will experience heat stress. Below this temperature, day-old chicks will not experience heat stress during transport (comfort zone).

7.2.5. Conclusions related to Cold Stress

1) The AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) that day-old chicks will experience cold stress if the body temperature drops below 40°C.

2) The most sensitive and specific ABMs to measure cold stress in day-old chicks are huddling, distress calls, increased respiration frequency, mean surface temperature, cloacal temperature and lethargy.

3) The lower limit of the comfort zone of day-old chicks is estimated to be at 30°C. The AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that if the effective temperature (at the level of the chicks) falls below a temperature of 30°C, the chick's mechanisms to cope with cold stress will become less effective and the chicks will experience cold stress. If it remains above this threshold, day-old chicks will not experience cold stress during transport.

4) As the effective temperature decreases below the lower limit of the comfort zone, the severity of the cold stress will increase and chicks might become lethargic and even die.

7.2.6. Conclusions related to Prolonged Hunger and thirst

1) In the case of day-old chicks, the welfare consequences 'prolonged hunger' and 'prolonged thirst' are dealt with together because the unique hazard is the delayed access to both feed and water.

2) All broiler chicks hatched in a hatchery without a system providing direct access to feed and water will experience prolonged hunger and thirst up to some degree. Prevalence is high since all chicks will be submitted to it and the severity of prolonged hunger and thirst will depend on the time between hatching and access to feed and water.

3) No sensitive and specific ABMs are available to assess prolonged hunger and prolonged thirst during transport of day-old-chicks.

4) The total time without feed and water starts when broiler chicks hatched in the hatchery and ends when all chicks access to feed and water. If provided during transport, they are given concurrently in the form of a nutritious hydrogel.

5) The AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) that day-old chicks subject to feed and water deprivation longer than 48 h (from hatching to access to feed and water at placement) will experience prolonged hunger and thirst which is detrimental to their welfare.

6) Transport of fertilised eggs and on farm hatching avoid the need to transport day-old chicks and prevent prolonged hunger and thirst due to delayed access to feed and water related to transport.

7) The provision of feed and water at the hatchery before the transport (in hatching tray or at hatchery) reduces the period without feed and water.

8) Provision of feed and water during transport (in the boxes) is also a way to prevent or mitigate prolonged hunger and thirst.

9) There is a gap of scientific evidence about the affective state associated with lack of feed and water provision, but these welfare consequences can be assumed.

7.2.7. Conclusions related to maximum transport duration

1) Transport duration includes not only the journey duration (period the vehicle is in transit), but also the time the day-old chicks are in the boxes and eventually the time they hatch in the hatchery.

2) With increasing journey time, and from the very start of the transport (putting chicks in transport boxes), day-old chicks are exposed for longer to a number of highly relevant welfare consequences (e.g. sensory overstimulation, heat and cold stress) that are stressful. These welfare consequences are present throughout the duration of the journey until day-old chicks are uncrated.

3) Maximum time for transport depends on the total time without access to feed and water from the moment they hatch. Therefore, if day-old chicks are not provided with feed and water during transport, the maximum time of transportation (including loading, journey, unloading) is equal to 48 h, counted from the first chicks to hatch until the last chick to access feed and water.

4) The maximum time between hatching and first access to feed and water (including time spent in the hatchery, holding time, loading, transport and unloading time) must not exceed 48 h.

5) Planning, coordination and communication among the different actors involved with transport (e.g. hatchery, drivers and destination farm staff) is crucial to reduce transport duration by minimising loading times and waiting time upon arrival of day-old chicks.

7.2.8. Conclusions related to iceberg indicators

1) Death on arrival (DOA) is an iceberg indicator for the assessment of welfare during transport.

2) From the animal welfare point of view, the target of DOA should be 0%. However, day-old chicks might die during transport due to factors that are not only attributable to the transport conditions.

3) The AHAW Panel concluded with 66–100% certainty (likely to almost certain) that DOA higher than 0.1% might be due to welfare consequences experienced during transport.

4) The main welfare consequences leading to DOA are heat stress, cold stress as well as the conditions of the animals (unfitness for transport).

5) The longer the transport duration the longer the time animals are experiencing these welfare consequences and higher the risk of DOA.

7.3. Conclusions for rabbits

1) The transport of rabbits consists of five stages: (1) Preparation that includes planning of the journey, preparation of the rabbits and assessment of fitness for transport; (2) Loading that includes catching the rabbits, placing them in containers (crating) and loading of containers onto the vehicle; (3) Journey that includes the movement of rabbits by vehicle until the place of destination is reached; (4) Arrival that includes the period from arrival of the vehicle, unloading of the containers from the vehicle, and waiting period; and (5) Uncrating that includes the removal of the rabbits from the containers.

2) The highly relevant welfare consequences for transport of rabbits are handling stress, injuries, restriction of movement, sensory overstimulation, motion stress, heat stress, cold stress, prolonged hunger and prolonged thirst.

3) During the transport, rabbits are exposed to several hazards at the same time. The presence and severity of the welfare consequences they are exposed to mainly depend on the conditions of the transport and the way welfare consequences can be mitigated and/or the way hazards can be corrected/prevented.

7.3.1. Conclusions related to fitness for transport

1) Pre-existing conditions of poor health are more likely to increase the severity of the welfare consequences transported rabbits are exposed to during the journey and the animals are more likely to die during the journey.

2) The main conditions making rabbits unfit for transport are: evident signs of illness, cachexia, severe lameness (unable to stand or walk more than a few steps), female rabbits in the last third of gestation, female rabbits till 7 days after parturition, un-weaned rabbits, open wounds, prolapses, abscesses, fractures, dislocations, wet fur in low effective temperature.

7.3.2. Conclusions related to handling stress

1) Handling stress is a welfare consequence inherent to transport, and all rabbits are likely to experience this welfare consequence, mainly during catching and crating of the loading stage, during unloading at the arrival stage and during the uncrating stage.

2) The ABMs escape behaviours, piling up and vocalisations are sensitive and specific to assess handling stress in a group of rabbits during catching and crating in the loading stage and uncrating.

3) Rough handling will increase the severity of handling stress compared to gentle handling.

4) Handling stress cannot be prevented or corrected, only mitigated.

7.3.3. Conclusions related to Injuries

1) Injuries lead to negative affective states such as pain and discomfort in rabbits, and the severity of these welfare consequences can range from moderate to very high.

2) Injuries are usually inflicted during loading and the welfare consequences prevail throughout the stages of transport.

3) ABMs of injuries, such as wounds and bruises, are sensitive and specific for the assessment during catching and crating of the loading stage, during unloading at the arrival stage and during the uncrating stage.

4) The prevalence and severity of injuries depend on many factors, but rough handling, due to lack of staff skills and training, is the main hazard.

5) Rough handling will increase the risk of injuries compared to gentle handling.

6) Design, operation and maintenance of equipment such as transport containers will also influence the risk of injuries to rabbits.

7) The mitigating measures for handling stress will also reduce the risk of injuries.

7.3.4. Conclusions related to Restriction of movement

1) Restriction of movement is a welfare consequence that is inherent to the transport situation, as rabbits are confined in containers.

2) Restriction of movement is considered severe when rabbits are not provided with enough floor space and height to adopt natural resting or sitting postures with the head and ears extended in a comfortable position, and moderate when it is unable to change posture within the container.

3) To assess restriction of movement in rabbits, the ABMs 'Sitting posture', 'lying posture' and 'ear position when sitting' are the most specific and sensitive ABMs used during loading and during unloading at arrival. However, assessment of these ABMs is not currently feasible during the journey stage.

4) Insufficient space allowance and insufficient height of container are the hazards for restriction of movement and also for other welfare consequences such as heat stress.

5) The generic allometric equation 'space allowance (cm^2/rabbit) = 270 × live weight (kg$^{2/3}$)' can be used to calculate the minimum required floor space allowance for rabbits to adopt a sitting position, change posture within the container and to provide some extra space between animals to ensure proper ventilation. Space allowances for the different rabbit live weights are reported in Table 30 of Section 5.7.3.4. The AHAW Panel concluded with 90–100% certainty (from very likely to almost certain) that these space allowances will mitigate restriction of movement.

6) However, if rabbits should be allowed to adopt recumbent position, more space is needed. This position might be adopted when rabbits are transported at high effective temperature or during long journeys. Position 2 of the planimetric measures provides the minimum required floor space for rabbits to rest in ventral recumbency, front legs extended and hind legs bent to the body (see Figure 34 and Table 30).

7) To prevent restriction of movement, the height of transport containers should allow the rabbits to keep their ears erected in a natural position while sitting.

8) The AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that a crate height of 35 cm will ensure slaughter rabbits (up to 3 kg) can sit with their ears erect.

9) The AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that a crate height of 40 cm will ensure breeding rabbits (between 4.5 kg and 6 kg) can sit with their ears erect.

7.3.5. Conclusions related to Sensory overstimulation

1) During commercial transport, rabbits are subjected to stimulation of the visual, auditory, chemical, sensory somatosensory and vestibular systems. Overstimulation of any system or combination of systems will result in stress and/or negative affective states such as fear, and/or discomfort.

2) No feasible ABMs have been identified to assess sensory overstimulation.

3) Rabbits may be exposed to sudden unfamiliar changes in light or sound intensity and duration associated with vehicle movement and the travel environment.

4) Acute or sudden auditory or visual stimuli may become recurrent and continuous and will constitute a chronic overstimulation.

5) Olfactory and gustatory systems of the rabbits may be over stimulated by exposure to exhaust fumes and other general pollutants (e.g. ammonia).

6) In addition to the primary problem of sensory overstimulation and the induction of aversion in the transported rabbits, this welfare consequence can exacerbate the effects of other welfare consequences such as handling stress, thermal stress, restriction of movement and prolonged hunger and thirst.

7) The prevention or mitigation of sensory overstimulation can only be achieved by planning of the transport, changes to loading equipment and vehicle design and structure or by attention to the catching and driving style and vehicle operation and maintenance and quality of the road.

7.3.6. Conclusions related to Motion Stress

1) The overstimulation of the somatosensory and vestibular systems will result in motion stress and may have a significant impact upon the welfare of rabbits during the journey.
2) No feasible ABMs for assessing motion stress during transport in rabbits transported in containers have been identified.
3) The severity of the motion stress experienced will depend upon the frequency and intensity of vibration and acceleration movement resulting from movement and operation of the vehicle.
4) Rabbits have been used to study the effects of motion sickness, but there is insufficient evidence that they become motion sick from road transportation.
5) Exposure to acceleration and vibration will induce postural instability, increase the requirement for muscle activity to restore stability and will increase the risk of injuries and fatigue.
6) These stimuli will be further dependent upon vehicle design and structure, container design and operation, and factors such as vehicle speed, driving style and road type and surface.
7) The risk of motion stress may be reduced by attention to vehicle suspension, tire and vehicle maintenance, driving style and driver education and training.

7.3.7. Conclusions related to Heat Stress

1) Heat stress is a highly relevant welfare consequence when transporting rabbits, with prevalence being higher in the summer season compared to other seasons. Rabbits can experience heat stress during the entire journey or for parts of the journey. With continued exposure, severity will increase over time as animals experience distress and eventually fail to cope and die.
2) Panting is a highly specific and sensitive ABM of heat stress.
3) High effective temperature induces thermoregulatory responses in the animals and is the principal hazard for heat stress.
4) The effective temperature experienced by the animal is affected by many factors such as ambient temperature, humidity and radiation, and air movement. However, in the context of transport of animals in containers, effective temperature is considered to be primarily determined by dry-bulb temperature and humidity.
5) Effective temperature can be divided into a safe, alert or danger zones based upon thermoregulatory demands and physiological responses. In the safe zone animals require minimal thermoregulatory effort. In the alert zone, animals need increasing thermoregulatory efforts. In the danger zone, animals will require serious thermoregulatory efforts that might be insufficient and will lead to heat stress.
6) Several indices based on dry-bulb temperature and relative humidity have been developed to measure high effective temperature inside the transport containers. One of this is the Temperature-Humidity Index (THI) that is very commonly used for livestock, including rabbits.
7) The AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that if THI is above 28.9, the rabbit's mechanisms to cope with heat stress will become less effective and the rabbits will experience heat stress (danger zone). If THI remains below 27.8, rabbits will not experience heat stress during transport (safe zone). Between THI values of 27.8 and 28.9, there will be an increasing risk of heat stress (alert zone). In order to calculate THI, the equation located in Section 5.7.7.2 can be used. The combinations of relative humidity and dry-bulb temperature giving rise THI values for the safe, alert and danger zones can be found in Figure 36 located in Section 5.7.7.2
8) In passively ventilated vehicles, there is an uneven distribution of thermal conditions within the load and the risk of heat stress will be highest close to the final air exit points.
9) Thermal comfort in all rabbits during the journey can be achieved in fan ventilated vehicles, where the movement of air into, within and out of the vehicle container is controlled by a combination of suitably positioned mechanical fans of sufficient capacity, and natural apertures which enhance established natural air pressure gradients.
10) Controlled environment or air-conditioned vehicles can regulate or modify internal thermal conditions by appropriate heating or cooling. They have the major advantage that the

internal environment may be controlled regardless of external weather or thermal conditions and does not rely upon vehicle movement.

7.3.8. Conclusions related to Cold Stress

1) Cold stress is a highly relevant welfare consequence when transporting rabbits, with prevalence being higher in the winter season. Rabbits can experience cold stress during the entire journey or for parts of the journey. Severity will increase over time as animals experience distress and eventually fail to cope and die.
2) Piloerection, shivering and core body temperature are considered to be sensitive and specific ABMs for the assessment of cold stress.
3) Too low effective temperature is the principal hazard for cold stress.
4) Cold stress is mainly induced by low dry-bulb temperature, although it is also influenced by air movement. Humidity does not have a big impact on thermoregulation demands for cold stress. Therefore, conclusions for cold stress are based on dry-bulb temperature (and are not based on indexes combining temperature and humidity).
5) Rabbits do not experience cold stress when exposed to effective temperatures within the comfort zone. Below the comfort zone, with smaller decreases in effective temperature, the animals can still cope and are most likely not experiencing cold stress. When temperatures decrease further, thermoregulatory demands will increase, and the risk of cold stress increases. This is a continuous process. At the critical temperature of the thermoneutral zone, rabbits will experience cold stress.
6) The AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that below 10°C, the rabbit's mechanisms to cope with cold stress will become less effective and the rabbits will experience cold stress.
7) In passively ventilated vehicles, there is an uneven distribution of thermal conditions within the load and the risk of cold stress will be the highest close to air entry points.

7.3.9. Conclusions related to Prolonged Hunger

1) Animals transported in containers in most cases will experience the welfare consequences of transport during the total time in the containers until they are removed from the containers. The total feed withdrawal starts when feed is removed on farm and ends when all animals are removed from containers following unloading from the vehicle and fed or slaughtered.
2) The welfare consequence of prolonged hunger might affect all rabbits transported in containers due to feed deprivation on-farm when applied, during the journey time and upon arrival.
3) No feasible ABMs for assessing prolonged hunger in rabbits whilst transported in containers have been identified.
4) Deprivation of feed for too long is the main hazard for prolonged hunger. The length of the feed withdrawal time before catching and crating affects the risk of animals experiencing prolonged hunger during transport.
5) Considering the available evidence, the AHAW Panel concluded with > 50–100% certainty (more likely than not) that rabbits subjected to feed withdrawal periods longer than 6 h will experience prolonged hunger.
6) The AHAW Panel concluded with 66–100% certainty (from likely to almost certain) that rabbits subjected to feed withdrawal periods longer than 12 h will experience prolonged hunger.
7) There is no scientific evidence of a welfare benefit of fasting rabbits before transport.

7.3.10. Conclusions related to Prolonged Thirst

1) The welfare consequence of prolonged thirst might affect all rabbits transported in containers due to the current practices of water deprivation during the journey time and upon arrival.
2) No feasible ABMs for assessing prolonged thirst during transport in rabbits transported in containers have been identified.
3) Deprivation of water for too long is the main hazard for prolonged thirst.

4) Considering the scarce available evidence, the AHAW panel concluded with 66–100% (from likely to almost certain) that rabbits subjected to water deprivation periods longer than 12 h will experience prolonged thirst.

5) If temperatures are above the thermal comfort zone, the evaporative heat loss through panting induces dehydration and advances the onset of prolonged thirst.

6) There is no scientific evidence of a welfare benefit of water deprivation in rabbits before transport.

7.3.11.   Conclusions related to maximum transport duration

1) The transport duration includes not only the journey duration itself (period the vehicle is in transit), but also the time the animals are in the containers and the prior feed withdrawal period on farm (if done).

2) With increasing journey time, and from the very start of the transport (catching and crating), animals are exposed for longer to a number of highly relevant welfare consequences (sensory overstimulation, motion stress, restriction of movement) that are stressful. These welfare consequences are present throughout the duration of the transport until rabbits are uncrated.

3) The risk of occurrence of new health conditions caused by events during transport (e.g. injuries) will increase with journey duration, as will the duration of potential suffering associated with these.

4) Evidence on continuous welfare consequences involving stress and negative affective states (e.g. fear) suggest that limiting transport time would reduce the exposure to the associated hazards.

5) Direct evidence on how the progressively developing welfare consequences change with increasing transport duration is limited.

6) Based on the presence of the continuous welfare consequences from the start of any transport, combined with the scarce data on the progressive welfare consequences, it can be concluded that any duration of transport will negatively affect animal welfare.

7) The severity of welfare consequences during the journey stage of transport will depend on the exact conditions pertaining to an individual journey (e.g. microclimatic conditions, space allowance and road conditions). These hazards potentially interact. The exposure to these hazards will continue at least as long as the journey continues.

8) Rabbits cannot be fed and provided with water in the containers. After 12 h from food withdrawal, rabbits will experience prolonged hunger and thirst. Therefore, transport duration (including on-farm feed and water withdrawal until being uncrated and slaughtered or provided feed) exceeding 12 h will compromise animal welfare.

9) This does not imply that no welfare consequences occur before these periods, as there are many factors other than transport duration that affect the risk of welfare consequences during a journey.

10) Planning, coordination and communication among the different actors involved with transport (e.g. farmer, drivers, slaughterhouse or destination farm staff) is crucial to reduce transport duration by minimising loading times and waiting time upon arrival of rabbits.

7.3.12.   Conclusions related to iceberg indicators

1) Death on arrival (DOA) is a relevant iceberg indicator for the assessment of welfare during transport and is shown to have a higher relationship with transport than with farming situations.

2) From the animal welfare point of view, the target DOA should be 0%. However, rabbits might die during transport due to factors that are not attributable directly to the transport conditions.

3) The AHAW Panel concluded with 66–100% certainty (likely to almost certain) that DOA higher than 0.1% might be due to welfare consequences experiencing during transport.

4) The main welfare consequences leading to DOA are heat stress, cold stress as well as the conditions of the rabbits (unfitness for transport).

5) The longer the journey duration the longer the time animals are experiencing these welfare consequences and higher the risk of DOA.

8.   Recommendations

8.1.   Recommendations for domestic birds

8.1.1.   Recommendations related to fitness for transport

1) Prior to catching and/or at the latest during catching and loading, the fitness for transport of each animal should be assessed. Birds that are not fit to be transported should receive appropriate treatment or be immediately humanely killed.

2) Farm staff (producers/stock carers and catchers) should receive training to inspect all animals and recognise birds unfit for transport as well as training to provide appropriate treatment or to cull unfit animals humanely.

3) To minimise the risk of birds becoming unfit for transport between the last inspection and the time of catching, the final inspection should be as close to the time of catching as possible (a maximum of 12 h prior to catching).

4) Birds must also be inspected during catching and crating. Birds unfit for travel that have not been identified during the preparation for transport stage should not be crated but receive appropriate treatment or be immediately humanely killed.

5) At all times before and during the loading stage a competent person should be responsible for animal welfare and a person trained in humane killing techniques must be available to dispatch injured and unfit animals and suitable equipment such as captive-bolt stunners should be available.

6) In automated catching and crating processes one or more people should be present at the point of loading of the containers to remove any injured animals.

7) Research is needed to determine fitness for transport. In particular, knowledge about the risk associated with transport of animals with a number of conditions potentially leading to pain (such as lameness, dislocations, fractures, open wounds) as well as the refinement of ABMs useful to identify these conditions, are needed.

8.1.2.   Recommendations related to handling stress and injuries

1) To mitigate handling stress, training of staff should acquire the knowledge and skills required to perform their allocated tasks efficiently, but in a way that minimise handling stress and risk of injuries to the birds, is identified as the most important mitigative measure.

2) Domestic birds should be carried upright by holding the wings against the body, and not inverted or by their neck or wings. Birds should not be swung, thrown or dropped during the process of catching and crating.

3) If birds are handled in inverted position, in order to reduce the risk of dislocated joints or fractures, they should be caught, lifted and carried by two legs, using breast slides in cages, maximum 3 birds/hand. Any wing flapping should be prevented by placing the bird against the operator's leg. The duration of inversion should be minimised, e.g. by placing containers close to the birds. To prevent injuries, birds should be placed carefully into the containers avoiding impacting the birds against any hard objects. Birds should be moved gently towards the back of the drawers and be loaded from the top, ensuring that heads, wings and legs are fully inside the container and that every bird is sitting upright before closing. A designated person should be dedicated to closing the containers after ensuring that the correct number of birds has been inserted and the other above-mentioned requirements are fulfilled.

4) Injuries due to hitting or pushing birds against the edges of the crate or container entrance can be prevented by using crates or containers with large doors or openings.

5) Injuries due to jamming and crushing of birds' body parts can be prevented by careful supervision when placing the birds into the containers and before and after loading and unloading the containers.

6) Once loaded into containers the crates or modules should be moved smoothly, lifted and placed carefully.

7) The ABMs for handling stress such as 'escape attempts', 'piling up' and 'distress calls' should be assessed during catching and crating of the loading stage, during unloading at the arrival stage and during the uncrating stage.

8) The ABMs for injuries such as hanging, non-functional limbs, severe lameness, protruding bones, bruises and wounds should be assessed during catching and crating at the loading stage and during unloading at arrival stage of the birds.

9) Severely injured birds should humanely killed immediately.

8.1.3. Recommendations related to Restriction of movement

1) To mitigate restriction of movement during transport, birds should be given sufficient space allowance to sit all at the same time without overlapping and to be able to change/adjust position. The generic allometric equation 'space allowance (cm^2/bird) = 290 × live weight (kg$^{2/3}$)' should be used to calculate the minimum required floor space (space allowance) to enable all birds to simultaneously adopt a sitting position and to be able to shuffle around.

2) For pullets and laying hens up to 2 kg, especially when well feathered, the space allowance derived from the planimetric measurements should be used instead of the allometric equation. Examples of space allowances for the different animal categories are reported in Table 11 (Section 3.7.3.4) of this text.

3) The height of the container should be such that the comb or head does not touch the ceiling when birds sit with their head and neck in a natural posture or when they change position. Examples of minimum heights of the containers are provided in Table 12 (Section 3.7.3.4) of this text for the different animal categories.

4) The ABMs 'sitting posture' and 'head posture' of the birds should be assessed at crating, before loading the containers on the vehicle. Birds should be visually assessed to ensure that all can sit simultaneously and that there is sufficient height for them to sit with their head and neck in a natural posture without the head/comb touching the ceiling of the container.

5) More research is needed about the height requirements and the impact on restriction of movement for various domestic birds.

8.1.4. Recommendations related to Sensory overstimulation

1) Escape attempts and distress calls should be assessed after crating during loading and during the stops, as once the vehicle is moving these ABMs cannot be assessed.

2) Vehicles for carriage of poultry should be enclosed (e.g. curtain sides) wherever possible, with due consideration for vehicle ventilation, to minimise exposure to sudden and rapidly changing light and sound levels.

3) Overstimulation of the chemical senses should be avoided by appropriate enclosure of the load, adequate ventilation and appropriate location of the air inlets and outlets regardless of the ventilation system (e.g. active or passive ventilation).

4) To prevent and/or mitigate sensory overstimulation, staff should be educated and trained to identify and prevent the hazards that cause sensory overstimulation.

8.1.5. Recommendations related to Motion Stress

1) Driving style will impact heavily upon the risks of motion stress through excessive acceleration, deceleration, braking and cornering. These should be avoided and driving style appropriate for the carriage of livestock should be ensured by the necessary training and education.

2) Good vehicle maintenance will reduce the risks associated with motion and vibration. Particular attention should be paid to tyre pressures and maintenance of the vehicle suspension.

3) In the longer term the introduction of improved suspension systems for all poultry vehicles may reduce the influence of some aversive vibration frequencies. This might be achieved by the use of air suspension systems although research is required to confirm what suspension characteristics will be most beneficial in terms of suppression of detrimental vibrations.

4) When planning animal transport, the best route should be chosen to avoid inappropriate roads and the traffic situation should be evaluated before departure and during the journey to be able to avoid obstructions on route.

8.1.6. Recommendations related to Heat Stress

1) To prevent heat stress, it is recommended that domestic birds should travel in the safe zone in which they will require minimal or no thermoregulatory effort to maintain constant deep body temperature during the journey. The safe zone may be described in terms of boundary conditions, e.g. a temperature of 18°C and an RH of 65%, a temperature of 21°C and an RH of 45% and a temperature of 32°C and an RH of 15%. The combinations of relative humidity and dry-bulb temperature giving rise to AET values for the safe, alert and danger zones can be found in Section 3.7.7.2 in Figure 20.
2) Domestic birds should never travel in the danger critical zone in order to avoid heat stress.
3) The AHAW Panel suggested domestic birds could travel in the alert zone (AET) or warning zone (ECI), if the duration of the journey is kept to a minimum (up to 4 h of journey time). This is because in the alert zone animals need to increase thermoregulatory efforts with increasing thermal load imposed and the longer the duration of the journey under these conditions the higher risk of heat stress. The upper boundary of the Alert Zone again may be defined in terms of combinations of temperature and RH e.g. 22°C and an RH of 95%, 26°C and an RH70% and a temperature of 30°C and an RH of 50%
4) Temperature and relative humidity should be continuously monitored and recorded inside the transport containers close to the birds in different locations in the vehicle, including the upper front and central areas.
5) Panting should be used as an ABM to assess heat stress during journey breaks and arrival. Upper and central containers should be prioritised for inspection.
6) If heat stress is suspected, upon arrival at the destination cloacal temperatures should be taken from sample birds in the locations of the load recognised to be at most risk of heat stress or in which birds are observed to be panting excessively. In this case, the crates should be unloaded following the recommended order of loading during high ambient temperatures to uncrate the birds at the locations of highest temperatures first.
7) The most efficient measure for preventing heat stress is to transport animals in vehicles using effective mechanical ventilation or air conditioning.
8) Other preventive measures include to plan journeys to take place during the night or during the coolest hours of the day, if the weather forecast predicts hot, sunny and/or humid conditions during other parts of the day.
9) To mitigate heat stress, during loading, journey breaks and unloading in hot weather, shade should be provided and birds should be exposed to breeze or portable or fixed fans.
10) In addition to reducing the number of birds per container, individual containers or entire modules should be left empty. This approach should be applied in the locations within the load known to be most at risk for heat stress.
11) An emergency plan should be ready in case of conditions above the suggested threshold for the alert zone.
12) In order to reduce the risk of thermal stress all slaughterhouses should be equipped with holding facilities with banks of fans to ventilate the stationary vehicle. The provision of roofs and some wall structure will provide shade, will reduce direct solar radiation on the vehicles and will provide a better control of the direction of the forced convective flow.
13) In order to reduce the risk of thermal stress the arrival, unloading and uncrating should be planned, and coordinated to minimise waiting time upon arrival of domestic birds and keep lairage duration to the minimum. This includes communication with farms, live bird catching teams and hauliers.
14) Further research should be carried to develop automated technology to detect animal responses inside the vehicle for assessing heat stress.

8.1.7. Recommendations related to Cold Stress

1) Domestic birds should travel in their comfort zone in order to avoid cold stress during transport. Temperature inside all transport containers should not be below 10°C during transport.

2) End-of-lay hens should travel in their comfort zone in order to avoid cold stress during transport. Temperature inside all transport containers should not be below 18°C during transport.

3) Huddling, fluffing up of feathers, shivering and cloacal temperature are the ABMs to be used to assess cold stress during loading, transport, upon arrival at the slaughterhouse or during unloading.

4) Bird containers closest to the known air inlets (and at highest risk of water ingress and higher air speeds) should be prioritised for inspection and checking and birds in all containers at the edges of load must be considered to be at the highest risk of cold stress as compared to birds in the centre of the load.

5) Temperature should be continuously monitored and recorded inside the transport containers close to the birds in different locations in the vehicle, including close to the position of the air inlets and the edge of the load.

6) Preventive measures for cold stress include: vehicles and/or modules should have curtains, which can be used according to weather conditions, or, even better, should have solid sides with defined inlet and outlet apertures which may function in a similar manner to modified curtained vehicles but give more thermal protection during cold weather.

7) Avoid loading modules and crates with birds near the air inlet (leave empty).

8) Avoid the coldest hours of the day for transportation of animals to avoid extreme climatic conditions.

9) Avoid any wetting of birds or crates and drawers if ambient temperatures suggest a risk of cold stress (during the journey, loading, standing).

10) Have an emergency plan ready in case of conditions above the suggested threshold.

11) Cold stress can be mitigated by adjusting ventilation or closing curtains.

12) Upon arrival at the slaughterhouse lairage waiting times should be kept to a minimum and loads with more vulnerable birds should be unloaded and slaughtered immediately.

8.1.8. Recommendations related to Prolonged Hunger

1) To prevent prolonged hunger during transport, the total time of feed deprivation should not exceed 6 h.

2) To mitigate against prolonged hunger during transport, the total time of feed deprivation should not exceed 12 h.

3) For end-of-lay hens, to mitigate against prolonged hunger during transport, the total time of feed deprivation should not exceed 10 h. However, it should be kept to a minimum as the time of onset of prolonged hunger might be shorter than 10 h. Further research is needed to ascertain the time end-of-lay hens start to experience prolonged hunger.

4) From a welfare perspective, feed withdrawal on farm should be avoided as there is no scientific evidence of a welfare benefit of fasting domestic birds before transport.

5) Upon arrival, birds should be unloaded without any delay and provided with feed and water (on destination farm) or be slaughtered immediately after arrival.

6) If the expected transport duration exceeds 12 h, provision of feed during transport might prevent, correct or mitigate prolonged hunger. However, if birds are to be fed in transport containers, the devices for feeding must be designed in a way enabling all birds' access to sufficient amounts of feed. They must also have free access to drinking water to be able to digest any feed provided.

7) For end-of-lay hens, if the expected transport duration exceeds 10 h, provision of feed during transport might prevent, correct or mitigate prolonged hunger. However, if end-of-lay hens are to be fed in transport containers, the devices for feeding must be designed in a way enabling all hens' access to sufficient amounts of feed. They must also have free access to drinking water to be able to digest any feed provided.

8) Unless all birds can access feed from their original position in the container, the space allowance, including height, must enable them to move freely within the container to access feed and water.

9) Further research is recommended to identify feasible ABMs for assessing prolonged hunger during transport in birds transported in containers.

8.1.9.   Recommendations related to Prolonged Thirst

1) To prevent prolonged thirst during transport, the total time of water deprivation should not exceed 6 h.
2) To mitigate against prolonged thirst during transport, the total time of water deprivation should not exceed 12 h.
3) On farm, water should be available until the time of catching and crating.
4) Upon arrival, birds should be unloaded without any delay and provided with water (on destination farm) or be slaughtered immediately after arrival.
5) If the expected transport duration exceeds 12 h, provision of water during transport might prevent, correct or mitigate prolonged thirst. However, if water is provided in transport containers, the devices must be designed in a way enabling all birds' access to sufficient amounts of water.
6) Unless all birds can access water from their original position in the container, the space allowance, including height, must enable them to move freely within the container to access water.
7) Further research is recommended to identify feasible ABMs for assessing prolonged thirst during transport in birds transported in containers.

8.1.10.   Recommendations related to maximum transport duration

1) For animals transported in containers, the transport duration should be considered as the whole time the animals are kept in the containers. It is recommended to take into account the following time periods for the definition of maximum duration time transport: the time of feed withdrawal that has been applied on farm to prepare the transport, the time needed to crate all animals for the transport, the time the animals are in the containers/crates (before, during and after the journey itself) and the time needed to uncrate the animals (from the first one to the last one).
2) Based on an overall assessment of the welfare consequences that are continuously present or developing over time, journeys up to a maximum of 12 h, including on farm feed withdrawal are recommended for domestic birds.
3) Based on an overall assessment of the welfare consequences that are continuously present or developing over time, journeys up to a maximum of 10 h, including on farm feed withdrawal are recommended for end-of-lay hens.
4) Related to thermal stress, if birds travel in the alert zone (AET) or warning zone (ECI), journey duration is to be kept to a maximum of 4 h.
5) Planning, coordination and communication among the different actors involved with transport (e.g. live bird catching teams, haulers, drivers, slaughterhouse or destination farm staff) is crucial to reduce transport duration and mitigate the associated welfare consequences.
6) Further research to investigate relationships between journey time, journey conditions and ABMs considered to reflect affective states of domestic birds for all animal categories, including knowledge about the progressively developing welfare consequences and their changes over time.

8.1.11.   Recommendations related to iceberg indicators

1) The percentage of DOA should be collected and monitored for each individual transport.
2) The AHAW Panel suggested that DOA should be investigated when it exceeds 0.1% in all domestic birds.
3) DOA should be minimised by reducing the hazards that can lead to high levels of DOA. Preventive measures are detailed in the corresponding chapters (e.g. vehicle design and ventilation).

8.2.   Recommendations for day-old chicks

1) The only way to avoid de welfare consequences during transport is not to transport chicks and have the fertilised eggs transported and hatched on farm.

8.2.1.   Recommendations related to fitness for transport

1) Prior to catching and/or at the latest during catching and loading, the fitness for transport of each animal should be assessed. Day-old chicks that are not fit to be transported should receive appropriate treatment or be immediately humanely killed.

2) Farm staff should receive training to inspect all animals and recognise day-old chicks unfit for transport as well as training to provide appropriate treatment or to cull unfit day-old chicks humanely.

3) To minimise the risk of day-old chicks becoming unfit for transport between the last inspection and the time of catching, the final inspection should be as close to the time of catching as possible.

4) Day-old chicks must also be inspected during catching and crating. Day-old chicks unfit for travel that have not been identified during the preparation for transport stage should not be crated but receive appropriate treatment or be immediately humanely killed.

5) At all times before and during the loading stage a competent person should be responsible for animal welfare and a person trained in humane killing techniques must be available to dispatch injured and unfit animals and suitable equipment should be available.

6) Research is needed to determine fitness for transport. In particular, knowledge about the risk associated with transport of animals with a number of conditions potentially leading to pain as well as the refinement of ABMs useful to identify these conditions, are needed.

8.2.2.   Recommendations related to handling stress

1) The ABMs for handling stress 'posture and orientation on conveyor belt', 'chicks falling on the floor', 'escape attempts' and 'distress calls' should be assessed while chicks are put in the boxes.

2) If they have to be transported, it is recommended to handle the chicks manually with care or adjust all automatic equipment so that: i. velocity does not exceed 0.4 m/s, ii. drop height is not above 280 mm and iii. Speed of belts is below 27 m/min.

3) Poor design of system components where chickens become caught, trapped, smothered, crushed must be avoided.

4) Training of the staff should be performed to decrease rough handling.

5) If chicks fall on the floor, they should be lifted by both hands supporting the body.

8.2.3.   Recommendations related to Sensory overstimulation and Motion stress

1) It is recommended not to submit the chicks to high or rapidly changing light, to loud or sudden noise, to shaking and vibration in the crate.

8.2.4.   Recommendations related to Heat Stress

1) The ABMs of heat stress such as 'respiration frequency', 'mean surface body temperature' and 'cloacal temperature' should be assessed.

2) It is recommended that the cloacal temperature of chicks does not exceed 41°C.

3) It is recommended that the body surface temperature does not exceed 38°C.

4) It is recommended to keep day-old chicks in an environment where dry-bulb temperature does not exceed 35°C in their vicinity.

8.2.5.   Recommendations related to Cold Stress

1) The ABMs of cold stress 'huddling', 'distress calls', 'mean surface temperature', 'cloacal temperature', 'respiration frequency' and 'lethargy' should be assessed.

2) It is recommended to keep day-old chicks in an environment where temperature is not below 30°C.

3) It is recommended that the cloacal temperature of chicks does not go below 40°C.

4) It is recommended that the mean body surface temperature does not go below 34°C.

8.2.6.   Recommendations related to Prolonged Hunger and Thirst

1) To prevent day-old chicks experiencing prolonged hunger and thirst, the maximum time before first access to feed and water (including time spent in the hatchery, holding time,

loading, transport and unloading time) must not exceed 48 h. This time should be measured from the first chicks to hatch until the last chick has access to feed and water.

2) Hatching the birds on-farm should be considered in order to eliminate the transport and the welfare consequences of prolonged hunger and thirst associated with it.

3) If the expected transport duration is longer than 48 h, feed and water should be provided at the hatchery before the transport (in hatching tray or at hatchery).

4) Alternatively, feed and water might be also provided during transport (in the boxes).

5) More research is needed to assess the maximum time before access to feed and water that is not detrimental for day-old chicks.

6) Communication with live bird catching teams and hauliers and with the destination farm for planning and coordinating the arrival of day-old chicks, will reduce transport duration by minimising loading times and waiting time upon arrival of live birds and decrease prolonged hunger and thirst.

8.2.7. Recommendations related to maximum time duration

1) Planning, coordination and communication among the different actors involved with transport (e.g. live bird catching teams, haulers, drivers and destination farm staff) is crucial to reduce transport duration and mitigate the associated welfare consequences.

8.2.8. Recommendations related to iceberg indicators

1) The percentage of DOA should be collected and monitored for each individual transport.

2) DOA in day-old chick is an iceberg indicator of poor welfare during transport and should be investigated if it is higher than 0.1%.

3) DOA should be minimised by reducing the hazards that can lead to high levels of DOA. Preventive measures are detailed in the corresponding chapters (e.g. vehicle design and ventilation).

8.3. Recommendations for rabbits

8.3.1. Recommendations related to fitness for transport

1) Prior to catching and/or at the latest during catching and loading, the fitness for transport of each animal should be assessed. Rabbits that are not fit to be transported should receive appropriate treatment or be immediately humanely killed.

2) Farm staff (producers/stock carers and catchers) should receive training to inspect all animals and recognise rabbits unfit for transport as well as training to provide appropriate treatment or to cull unfit animals humanely.

3) To minimise the risk of rabbits becoming unfit for transport between the last inspection and the time of catching, the final inspection should be as close to the time of catching as possible (a maximum of 12 h prior to catching).

4) Rabbits must also be inspected during catching and crating. Rabbits unfit for travel that have not been identified during the preparation for transport stage should not be crated but receive appropriate treatment or be immediately humanely killed.

5) At all times before and during the loading stage a competent person should be responsible for animal welfare and a person trained in humane killing techniques must be available to dispatch injured and unfit animals and suitable equipment such as captive-bolt stunners should be available.

6) Research is needed to determine fitness for transport. In particular, knowledge about the risk associated with transport of animals with a number of conditions potentially leading to as well as the refinement of ABMs useful to identify these conditions, are needed.

8.3.2. Recommendations related to handling stress and injuries

1) The ABMs for handling stress such as 'escape attempts', 'piling up' and 'vocalisations' should be assessed during catching and crating of the loading stage, during unloading at the arrival stage and during the uncrating stage.

2) The ABMs for injuries such as 'wounds' and 'bruises', should be assessed during catching and crating, loading and unloading of rabbits.

3) Training of staff to acquire the knowledge and skills required to perform their allocated tasks efficiently, but in a way that minimise handling stress and risk of injuries to the rabbits, is identified as the most important mitigative measure.

4) Rabbits should be removed from the husbandry cages individually by holding and lifting by the neck by one hand, with or without support of the body with the other hand. Once outside the cages, their body should always be supported with the other hand.

5) Injuries due to hitting or pushing rabbits against the edges of the crate or container entrance can be prevented by using crates or containers with large doors or openings.

8.3.3. Recommendations related to Restriction of movement

1) To mitigate restriction of movement during transport, should be able to sit when transported.

2) To mitigate restriction of movement during transport, rabbits should be given a minimum space allowance calculated with the generic allometric equation 'space allowance (cm²/rabbit) = 270 × live weight (kg$^{2/3}$)' (See examples of space allowances for the different rabbit weight classes reported in Table 31 in Section 5.7.3.4).

3) However, more space is needed when rabbits are transported at high effective temperature or during long journeys and they should be allowed to rest in ventral recumbency, i.e. adopt a recumbent position with the front legs extended and hind legs bent to the body. In this case, position 2 of the planimetric measures should be used to provide for the minimum required floor space for rabbits (see Figure 34 and Table 30 in Section 5.7.3.3).

4) The height of the container should be at least 35 cm to ensure slaughter rabbits (up to 3 kg) can sit with their ears extended in a comfortable position.

5) The height of the container should be at least 40 cm to ensure rabbit breeders (between 4.5 kg and 6 kg) can sit with their ears extended in a comfortable position.

6) The posture of animals can be visually assessed during loading

7) Adjusting the number of rabbits to the size of the containers is recommended as a preventive measure.

8) The ABMs 'sitting posture', 'lying posture' and 'ear position when sitting' of the rabbits should be assessed at crating, before loading the containers on the vehicle.

8.3.4. Recommendations related to Sensory overstimulation

1) Vehicles for carriage of rabbits should be enclosed (e.g. curtain sides) wherever possible, with due consideration for vehicle ventilation, to minimise exposure to sudden and rapidly changing light and sound levels.

2) Overstimulation of the chemical senses should be avoided by appropriate enclosure of the load, adequate ventilation and appropriate location of the air inlets and outlets regardless of the ventilation system (e.g. active or passive ventilation).

3) Staff education and training to identify and prevent the hazards that cause sensory overstimulation is an effective preventive and mitigative measure.

8.3.5. Recommendations related to Motion Stress

1) Excessive acceleration, braking, stopping, cornering, gear changing, vibrations and uneven road surfaces during transport result in motion stress in rabbits and thus should be avoided.

2) The vehicle should be designed, constructed and maintained so as to reduce the risks associated with motion and vibration. Particular attention should be paid to tyre pressures and maintenance of the vehicle suspension.

3) Drivers should receive training and education in driving style appropriate for the carriage of livestock, namely they should avoid excessive acceleration, deceleration, braking and cornering.

4) When planning animal transport, the best route should be chosen to avoid inappropriate roads and the traffic situation should be evaluated before departure and during the journey to be able to avoid obstructions on route.

8.3.6.   Recommendations related to Heat Stress

1) To prevent heat stress, rabbits should travel in their safe zone, so they require no or minimal thermoregulatory efforts during the journey. If THI remains below 27.8, rabbits will not experience heat stress during transport (safe zone).
2) Rabbits should never travel in the danger zone (THI is above 28.9) in order to avoid heat stress.
3) Rabbits could travel in the alert zone (THI values of 27.8 and 28.9). In the alert zone, animals need to increase thermoregulatory efforts and the longer the duration of the journey under these conditions the higher risk of heat stress. However, due to lack of evidence, the AHAW Panel cannot recommend any maximum journey durations in the alert zone.
4) Temperature and relative humidity should be continuously monitored and recorded inside the transport containers close to the rabbits in different locations in the vehicle, including the upper front and central areas.
5) Panting should be used to assess heat stress during loading, transport, upon arrival at the slaughterhouse or during unloading. Upper and central containers should be prioritised for inspection.
6) The most efficient measure for preventing heat stress is to equip the vehicle with mechanical ventilation or reliable well-functioning air conditioning. Other measures are indicated in the next recommendations.
7) Journeys should be undertaken during the night or coolest hours of the day, if the weather forecast predicts hot, sunny and/or humid conditions during other parts of the day.
8) In hot weather, shade should be provided and rabbits should be exposed to breeze or portable fans during loading.
9) In addition to reducing the number of rabbits per container to reduce the impact of high ambient temperatures, individual containers or entire modules should be left empty of animals in the locations within the load known to be most at risk for heat stress. The configuration of the filled containers should be adjusted to increase air flow around and through them.
10) Water should be available until the loading starts.
11) Crate structure (e.g. increased size or number of perforations) should be modified to increase efficacy of passive ventilation regimes.
12) An emergency plan should be ready in case ambient temperatures increase above the suggested threshold for the alert zone.
13) In order to reduce the risk of thermal stress all slaughterhouses should be equipped with holding facilities with banks of fans to ventilate the stationary vehicle. The provision of roofs and some wall structure will provide shade, will reduce direct solar radiation on the vehicles and will provide a better control of the direction of the forced convective flow.
14) Further research should be carried to develop automated technology to detect animal responses inside the vehicle for assessing heat stress.

8.3.7.   Recommendations related to Cold Stress

1) Rabbits should travel in their comfort zone in order to avoid cold stress during transport. To achieve this, temperature inside all transport containers should not be below 10°C during transport.
2) Piloerection, huddling and shivering should be used as ABMs to assess cold stress during loading, transport, upon arrival at the slaughterhouse or during unloading.
3) Containers closest to the known air inlets (and at highest risk of water ingress and higher air speeds) should be prioritised for inspection and checking. Rabbits in all containers at the outside of the consignment are at greater risk of cold stress, compared to animals in the centre of the consignment.
4) Temperature should be continuously monitored and recorded inside the transport containers close to the rabbits in different locations in the vehicle, including the upper front and central areas.
5) Preventive measures for cold stress include: vehicles and/or modules should have curtains, which can be used according to weather conditions, or, even better, should have solid sides

with defined inlet and outlet apertures which may function in a similar manner to modified curtained vehicles but give more thermal protection during cold weather.

6) Loading modules and crates with rabbits near the air inlet should be avoided (leave empty).
7) In cold weather, the coldest hours of the day should be avoided for transportation of animals.
8) If ambient temperatures suggest a risk of cold stress (during the journey, loading, standing), any transport of wet rabbits or crates and drawers should be avoided.
9) An emergency plan should be in place in case of conditions below the suggested threshold.
10) Cold stress can be mitigated by adjusting ventilation or closing curtains.
11) Upon arrival at the slaughterhouse lairage waiting times should be kept to a minimum and loads with more vulnerable rabbits should be unloaded and slaughtered immediately.

8.3.8. Recommendations related to Prolonged Hunger

1) To prevent prolonged hunger during transport, the total time of feed deprivation should not exceed 6 h.
2) To mitigate against prolonged hunger during transport, the total time of feed deprivation should not exceed 12 h.
3) From a welfare perspective, feed withdrawal on farm should be avoided as there is no scientific evidence of a welfare benefit of fasting rabbits before transport.
4) Upon arrival, rabbits should be unloaded without any delay and provided with feed (on destination farm) or be slaughtered immediately after arrival.
5) If the expected transport duration exceeds 12 h, provision of feed during transport might prevent, correct or mitigate prolonged hunger. However, if rabbits are to be fed in transport containers, the devices for feeding must be designed in a way enabling all rabbits access to sufficient amounts of feed. They must also have free access to drinking water to be able to digest any feed provided. Unless all rabbits can access feed from their original position in the container, the space allowance, including height, must enable them to move freely within the container to access feed and water.
6) Further research is recommended to identify feasible ABMs for assessing prolonged hunger during transport in rabbits transported in containers.
7) Communication with live bird catching teams and hauliers and with the slaughterhouse or destination farm for planning and coordinating the arrival of rabbits, will reduce transport duration by minimising loading times and waiting time upon arrival of live birds and prevent prolonged hunger.

8.3.9. Recommendations related to Prolonged Thirst

1) To prevent prolonged thirst during transport, the total time of water deprivation should not exceed 12 h. This duration should be shorter if rabbits are transported above their thermal comfort zone and experience heat stress.
2) Water should be available on farm until the time of catching and crating.
3) Upon arrival, rabbits should be unloaded without any delay and provided with water (on destination farm) or be slaughtered immediately after arrival.
4) In case transport duration exceeds 12 h, rabbits are to be provided with water in transport containers in such way that all rabbits can access sufficient amounts of water.
5) Further research is recommended to identify feasible ABMs for assessing prolonged thirst during transport of rabbits.

8.3.10. Recommendations related to maximum transport duration

1) For animals transported in containers, the transport duration should be considered as the total time the animals are kept in the containers, and eventually the feed withdrawal period on farm. For animals transported in containers, the transport duration should be considered as the whole time the animals are kept in the containers. It is recommended to take into account the following time periods for the definition of maximum duration time transport: the time of feed withdrawal that has been applied on farm to prepare the transport, the time needed to crate all animals for the transport, the time the animals are in the

containers/crates (before, during and after the journey itself) and the time needed to uncrate the animals (from the first one to the last one).

2) Based on an overall assessment of the welfare consequences that are continuously present or developing over time, journey duration should not exceed 12 h, including on farm feed withdrawal (if applied).

3) Planning, coordination and communication among the different actors involved with transport (e.g. live bird catching teams, haulers, drivers, slaughterhouse or destination farm staff) are crucial to reduce transport duration and mitigate the associated welfare consequences.

4) Further research is recommended to investigate relationships between journey time, journey conditions and ABMs to reflect affective states of rabbits for all animal categories, including knowledge about the progressively developing welfare consequences and their changes over time.

8.3.11. Recommendations related to iceberg indicators

1) The percentage of DOA should be collected, reported and monitored for each individual transport.

2) The AHAW Panel suggested that DOA should be investigated when it exceeds this level might be 0.1% in in rabbits.

3) DOA should be minimised by reducing the hazards that can lead to high levels of DOA. Preventive measures are detailed in the corresponding chapters (e.g. vehicle design and ventilation).

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Abbreviations

ABM	animal-based measure
AET	Apparent Equivalent Temperature
ALERT	upper limit of the safe zone
ALT	alanine transaminase
CBT	core body temperature
CFD	Computational Fluid Dynamics
CFIA	Canadian Food Inspection Agency
DANGER	upper limit of the alert zone
DOA	Dead on arrival
ECI	Enthalpy Comfort Index
H/L	heterophil to lymphocyte (ratio)
LCT	lower critical temperature
MEMS	Microelectromechanical Systems
MS	Member State
NCP	National Contact Point
RH	Relative humidity
TCZ	thermal comfort zone
THI	Temperature-Humidity Index

TNZ	thermoneutral zone
UCT	upper critical temperature
VCR	vestibulo-collic reflex
VOR	vestibulo-ocular reflex
VSR	vestibulo-spinal reflex

Appendix A – Literature searches for the different animal categories

Broilers

General search

Date: 3 November 2020. Web of Science. Advanced search. Topic. The search was limited to records published after EFSA AHAW Panel, 2011. No language or document type restrictions were applied in the search string.

Search string TS = ((broiler* OR poultry OR chicken) AND (transport*) AND (welfare)).

Result = 169. Result after screening: 51.

Specific ToR search

Selected welfare consequences: Handling stress, Soft tissue lesions and integument damage, Bone lesions, Sensorial under/over- stimulation, Motion stress, Resting problems, Restriction of movement, Cold stress, Heat stress, Prolonged hunger, Prolonged thirst.

Date: 16 April 2021. Web of Science. 2011–2021. Advanced search. Topic.

The search was limited to records published after 2011. Language English only. No document type restrictions were applied in the search string.

Welfare consequences: Handling stress, Skin lesions and integument damage, Bone lesions

Catching and crating of poultry takes place in the housing system, and depopulation of crates happens at the slaughterhouse. Therefore, this search was not limited to transport.

TS = ((broiler* OR poultry OR chicken) AND (handling OR catch* OR harvest OR crat*) AND (welfare OR injur* OR lesion OR trauma OR bone OR bruis* OR joint))

Result: 238.

Welfare consequences: Sensorial over/understimulation, Resting problems, Restriction of movement, Motion stress

These welfare consequences are direct results of the transport situation (vehicle moving) in our context. This search was limited to the transport situation, assuming that only welfare consequences related to transport were relevant.

Search string: TS = ((broiler* OR poultry OR chicken) AND (resting OR sensorial OR sensory OR motion OR movement OR space OR stocking) AND (transport)).

Result: 144.

Welfare consequences: Heat stress, cold stress, prolonged hunger, prolonged thirst

For these, it is relevant to include farming conditions, as the thresholds for welfare will be the same regardless of whether the animal is being transported or not. Therefore, search was not limited to transport. However, welfare was used to limit the number of hits, and due to the assumption that relevant papers on these welfare consequences would be considering welfare.

Search string: TS = ((broiler* OR poultry OR chicken) AND (heat stress OR hyperthermia OR cold stress OR hypothermia OR thermal stress OR hunger OR starvation OR thirst OR dehydration) AND (welfare)).

Result = 264.

Laying hens

General search

Date: 26 October 2020. Web of Science. 2011–2020. Advanced search. Topic. The search was limited to records published after 2011. Language English only. No document type restrictions were applied in the search string.

Search string TS = ((lay* OR hen OR hens) AND (transport*) AND (welfare)).

Result = 76. Result after screening: 12.

Specific ToR search

Selected welfare consequences: Handling stress, Soft tissue lesions and integument damage, Bone lesions, Sensorial under/over- stimulation, Motion stress, Resting problems, Restriction of movement, Cold stress, Heat stress, Prolonged hunger, Prolonged thirst. Date: 16 April 2021. Web of Science.

2011–2021. Advanced search. Topic. The search was limited to records published after 2011. Language English only. No document type restrictions were applied in the search string.

<u>Welfare consequences: Handling stress, Skin lesions and integument damage, Bone lesions</u>

Catching and crating of poultry takes place in the housing system, and depopulation of crates happens at the slaughterhouse. Therefore, this search was not limited to transport. TS = ((laying hen OR hen OR pullet) AND (handling OR catch* OR harvest OR crate*) AND (welfare OR injur* OR lesion OR trauma OR bone OR bruis* OR joint)).
Result: 56.

<u>Welfare consequences: Sensorial over/understimulation, Resting problems, Restriction of movement, Motion stress</u>

As these are direct results of the transport situation (being in a crate + vehicle moving) in our context, this search was limited to transport.
Search string: TS = ((laying hen OR hen OR pullet) AND (resting OR sensorial OR sensory OR motion OR movement OR space OR stocking) AND (transport)).
Result: 22.

<u>Welfare consequences: Heat stress, cold stress, prolonged hunger, prolonged thirst</u>

For these, it is relevant to include farming conditions, as the thresholds for welfare will be the same regardless of whether the animal is being transported or not. Therefore, the search was not limited to transport. However, welfare was used to limit the number of hits, and due to the assumption that relevant papers on these welfare consequences would be considering welfare.
Search string: TS = ((laying hen OR hen OR pullet) AND (heat stress OR hyperthermia OR cold stress OR hypothermia OR thermal stress OR hunger OR starvation OR thirst OR dehydration) AND (welfare)).
Result = 102.

Ducks, geese, turkeys and quails

<u>General search</u>

Date: 27 October 2020. Web of Science. 2011–2020. Advanced search. Topic. The search was limited to records published after EFSA AHAW Panel, 2011. No language or document type restrictions were applied in the search string.
Search string: TS = ((duck* OR geese OR quail* OR turkey* OR game bird*) AND (transport*) AND (welfare)).
Result = 48. Result after screening: 22.

<u>Specific ToR search</u>

Selected welfare consequences: Handling stress, Soft tissue lesions and integument damage, Bone lesions, Sensorial under/over- stimulation, Motion stress, Resting problems, Restriction of movement, Cold stress, Heat stress, Prolonged hunger, Prolonged thirst.
Date: 20 April 2021. Web of Science. 2011–2021. Advanced search. Topic.
The search was limited to records published after 2011. Language English only. No document type restrictions were applied in the search string.

<u>Welfare consequences: Handling stress, Skin lesions and integument damage, Bone lesions</u>

Catching and crating of poultry take place in the housing system, and depopulation of crates happens at the slaughterhouse. Therefore, this search was not limited to transport.
TS = ((duck OR geese OR turkey OR quail) AND (handling OR catch* OR harvest OR crat*) AND (welfare OR injur* OR lesion OR trauma OR bone OR bruis* OR joint)).
Result: 137.

<u>Welfare consequences: Sensorial over/understimulation, Resting problems, Restriction of movement, Motion stress</u>

As these are direct results of the transport situation (being in a crate + vehicle moving) in our context, this search was limited to transport. Search string: TS = ((duck OR geese OR turkey OR quail)

AND (resting OR sensorial OR sensory OR motion OR movement OR space OR stocking) AND (transport)).
Result: 141.

Welfare consequences: Heat stress, cold stress, prolonged hunger, prolonged thirst

For these, it is relevant to include farming conditions, assuming the thresholds for welfare will be the same or comparable whether the animal is being transported or not. Therefore, the search was not limited to transport. However, welfare was used to limit the number of hits (otherwise 725), and due to the assumption that relevant papers on these welfare consequences would be considering welfare.
Search string: TS = ((duck OR geese OR turkey OR quail) AND (heat stress OR hyperthermia OR cold stress OR hypothermia OR thermal stress OR hunger OR starvation OR thirst OR dehydration) AND (welfare)).
Result = 27.

Game birds

Selected Welfare consequences: Handling stress, Sensorial under/overstimulation, Motion stress, Resting problems, Restriction of movement, Cold stress, Heat stress, Prolonged hunger, Prolonged thirst

Date: 20 April 2021. Web of Science. 2011–2020. Advanced search. Topic. The search was limited to records published after EFSA AHAW Panel, 2011. No language or document type restrictions were applied in the search string.
Search string: TS = ((game bird OR pheasant OR partridge OR guinea fowl OR pigeon) AND (handling OR catch* OR heat stress OR hyperthermia OR cold stress OR hypothermia OR thermal stress OR hunger OR starvation OR thirst OR dehydration OR transport OR welfare)).
Result: 336.

Day-old chicks

General search

Date: 15 December 2020. Web of Science. 2011–2020. Advanced search. Topic. The search was limited to records published after EFSA AHAW Panel, 2011. No language or document type restrictions were applied in the search string.
Search string TS = ((chick* OR hatch*) AND (transport*) AND (welfare)).
Result = 136. Result after screening: 9.

Specific welfare consequences search

Date: 20 April 2021. Web of Science. 2011–2021. Advanced search. Topic.
The search was limited to records published after 2011. Language English only. No document type restrictions were applied in the search string.
Selected welfare consequences: Handling stress, Sensorial under/overstimulation, Motion stress, Cold stress, Heat stress, Prolonged hunger, Prolonged thirst.

Welfare consequences: Cold stress, Heat stress, Prolonged hunger, Prolonged thirst

Search string: TS = ((chick* OR hatch*) AND (heat stress OR hyperthermia OR cold stress OR hypothermia OR thermal stress OR hunger OR starvation OR thirst OR dehydration) AND (welfare)).
Result: 219.

Welfare consequences: Sensorial under/overstimulation, Motion stress

TS = ((chick* OR hatch*) AND (resting OR sensorial OR sensory OR motion OR movement) AND (transport)).
Result: 146.

Welfare consequences: Handling stress

TS = ((chick* OR hatch*) AND (handling OR catch*) AND (welfare)).
Result: 87.

Rabbits

General search

Date: 27 October 2020. Web of Science. 2011–2020. Advanced search. Topic. The search was limited to records published after 2011. Language English only. No document type restrictions were applied in the search string.

Search string TS = ((rabbit*) AND (transport*) AND (welfare)).

Result = 22. Result after screening: 12.

Specific welfare consequences search

Selected welfare consequences: Handling stress, Soft tissue lesions and integument damage, Bone lesions, Sensorial under/overstimulation, Motion stress, Resting problems, Restriction of movement, Cold stress, Heat stress, Prolonged hunger, Prolonged thirst.

Date: 20 April 2021. Web of Science. 2011–2021. Advanced search. Topic.

The search was limited to records published after 2011. Language English only. No document type restrictions were applied in the search string.

Welfare consequences: Handling stress, Skin lesions and integument damage, Bone lesions

Catching and crating of poultry take place in the housing system, and depopulation of crates happens at the slaughterhouse. Therefore, this search was not limited to transport.

TS = ((rabbit) AND (handling OR catch* OR harvest OR crate*) AND (welfare OR injur* OR lesion OR trauma OR bone OR bruis* OR joint)).

Result: 760.

The reason for the large number of references was that papers using rabbits in experiments, especially as a model for studying and treating injuries in bone, appeared in the search. Limiting the search by including 'Welfare' in the search string resulted in less than 10 papers. Therefore, the search string was kept as it is above despite the large numbers of irrelevant results.

Welfare consequences: Sensorial over/understimulation, Resting problems, Restriction of movement, Motion stress

As these are direct results of the transport situation (being in a crate + vehicle moving) in our context, this search was limited to transport.

Search string: TS = ((rabbit) AND (resting OR sensorial OR sensory OR motion OR movement OR space OR stocking) AND (transport)).

Result: 118.

Welfare consequences: Heat stress, cold stress, prolonged hunger, prolonged thirst

For these, it is relevant to include farming conditions, as the thresholds for welfare will be the same regardless of whether the animal is being transported or not. Therefore, the search was not limited to transport. However, welfare was used to limit the number of hits, and due to the assumption that relevant papers on these welfare consequences would be considering welfare.

Search string: TS = ((rabbit) AND (heat stress OR hyperthermia OR cold stress OR hypothermia OR thermal stress OR hunger OR starvation OR thirst OR dehydration) AND (welfare)).

Result = 23.

Annex A – Protocol for the development of the opinion

Annex A is the protocol undertaken for the scientific development of this opinion and can be found in the online version of this output ('Supporting information' section) at: https://doi.org/10.2903/j.efsa.2022.7441

Annex B – Sources of uncertainty

Annex B is a table listing and describing all sources of uncertainty identified during the development of the scientific opinion, and can be found in the online version of this output ('Supporting information' section) at: https://doi.org/10.2903/j.efsa.2022.7441