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Impact of pre-slaughter factors on welfare of broilers

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ABSTRACT

Pre-slaughter factors adversely affecting bird welfare were studied at the slaughterhouse. The incidence of dead on arrival (DoA), bruises and dehydration was investigated in 64 different mixed-sex batches of broilers coming from 64 different farms rearing fast-growing genotypes (Ross or Cobb). The effects of catching team, method of catching, time of day for catching and transport, density per cage, transport duration, transport distance, lairage duration and water withdrawal were considered. The average DoA was 0.29%, ranging from 0.02% to 1.89% per batch. DoA rate has a higher probability of increase with the increase in transport distance ($t = 2.142$; $P = 0.037$; estimate = 0.009) and with catching the birds after midnight ($t = -2.931$; $P = 0.005$; estimate = 0.022). Longer transport durations for birds caught after midnight as well as longer lairage durations for birds caught after midnight are associated with the increase of DoA rate.

Bruises were observed in 3.37% of birds, ranging from 0.43% to 8.29% per batch. Bruises occurred mostly on wings (3.06%), followed by legs (0.19%) and breast (0.12%). A higher percentage of bruises occurred in batches with more birds per transport crate ($t = 2.185$; $P = 0.029$; estimate = 0.001). Dehydrated carcasses were observed in 22 out of 64 batches, accounting for 2.68% of condemnations. Signs of dehydration on carcasses were more frequently observed in batches subjected to longer withdrawal durations. Short transport distances, catching the birds before midnight and doing the transport by night are crucial in decreasing the DoA rate. Catching and crating processes seem to be responsible for the increase of percentage of bruises. Pre-slaughter operations should be adequate planned namely, transport and lairage durations, catching period and crating procedure in view to reduce negative effects on animal welfare.

1. Introduction

Current global production of broiler chickens is approaching 60 billion birds per annum (Food and Agriculture Organization of the United Nations, 2017). These birds are transported for slaughtering from their geographically dispersed farms. Prior to transportation, birds are subjected to fasting periods of varying duration. They are then manual or mechanically caught and placed into transport crates, which are subsequently loaded on to vehicles and transported to slaughterhouses (Nijdam et al., 2004). Upon arrival, the crates are unloaded from the vehicles and held in lairage for periods of differing durations (Tinker et al., 2005; Petracci et al., 2006).

The pre-slaughter procedures and practices impose varying degrees of stress upon the birds which will compromise their welfare status (Mitchell & Kettlewell, 2009; Jabocs et al., 2017). Catching and loading may be the most important moments because if birds are injured during this process it will have a profound effect on their response to the rest of their journey to the slaughterhouse (Whiting et al., 2007). Several reports indicate that appropriate handling procedure is essential in the reduction of mortality and trauma, such as haemorrhages, bruises and fractures (Nijdam et al., 2004; Caffrey et al., 2017). Broilers can be caught by hand wherein multiple birds are grasped by the legs, inverted, and carried by the catcher in both hands (Nijdam et al., 2005). Although birds appear to be more restless during one leg catching, the cautious handling of broilers to reduce stress seems to be more important than holding them by both legs (Langkabel et al., 2015).

Transport represents a brief period in the total lifespan of birds, however, there are indications that it is a time when both mental and physical suffering can be high (Knowles & Broom, 1990; Knezacek et al., 2010; Siegel & Honaker, 2014). During transportation the combination of stressors, rather than a single cause, is responsible for the decrease in welfare (Mitchell & Kettlewell, 1998; 2009).

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Although, it is generally accepted that animal transport of long duration is more likely to compromise animal welfare than short journeys, it is important to recognize that it is not journey duration *per se,* but the conditions of transport and the associated stress imposed, that are the source of welfare issues (Vecerek et al., 2006; Nielsen et al., 2011; Weeks et al., 2012). The microclimate within the trailer can be the most important factor affecting broiler welfare, as heat and cold stresses are two major contributors to both death and overall transportation stress in broilers (Mitchell & Kettlewell, 2004; Dadgar et al., 2010; Cockram & Dulal, 2018). Factors such as lack of feed, water and rest are all exacerbated by the length of exposure to challenging conditions, and thus, journey duration (Nielsen et al., 2011). The time of day for catching and transport, as well as density per crate are also important factors to be considered (Nijdam et al., 2004; Caffrey et al., 2017). Moreover, catching and transport to the slaughterhouse is considered by several authors as one of the most critical periods with regards to the risk of dehydration (Vanderhasselt et al., 2013).

After transport, a suitable lairage period in proper holding areas, with environmental control, is necessary to reduce thermal stress of live birds (Vosmerova et al., 2010). However, short lairage times are recommended for poultry due to low energy availability in metabolically active birds, who may have suffered physiological changes and body weight loss due to fasting before transport handling (Nijdam et al., 2004; Delezie et al., 2007; Mitchell & Kettlewell, 2008). Pre-slaughter factors can also affect the process of converting muscle to meat and meat quality can be compromised having negative impact on consumer acceptability (Schwartzkopf-Genswein et al., 2012).

Slaughterhouses have been recognized as a relevant source of data for monitoring welfare conditions of birds (Grandin, 2017; Saraiva et al., 2016). Under the Directive 2007/43/EC which provided minimum standards to ensure the protection of broilers during intensive production the official veterinarian controls include monitoring and follow-up welfare parameters at the slaughterhouse (Council Directive 2007/43/EC). Following these requirements, and taking into account the results of studies carried out in this field, European level thresholds for some welfare parameters have been defined and can be easily assessed at the slaughterhouse (Saraiva et al., 2016; EFSA, 2004). For example, indicators of poor welfare in transit may include dead on arrival (DoA) greater than 0.5%.

The aim of this study was to analyse the effect of pre-slaughter factors on DoA rate, presence of bruises on wings, legs and breast and of dehydrated carcasses in commercial flocks of broilers.

2. Material and Methods

2.1. Sample characterization

The welfare indicators were collected in one of the largest broilers slaughterhouses in Portugal and the study took place during springtime. The incidence of DoA, bruises and dehydration was investigated in 64 different mixed-sex batches of broilers coming from 64 different farms with intensive system production rearing fast-growing genotypes (Ross or Cobb). The study did not include any long distance export or intra-community trade journeys. Birds had a mean age of 36 days (range 30 to 45d) with body weight of 1.85 ± 0.26kg (range 1.43 to 2.41kg). The average size of transport batches was 5110 ± 745 birds (range 2360 to 6804 birds).

2.2. Pre-slaughter procedure

Each vehicle, per slaughter day, undertook two journeys from two distinct farms. The first journey from farm to the slaughterhouse occurred before midnight (00h) and the second after midnight (some of them already in daylight) on the slaughter day. The method of catching was manual in all flocks and each vehicle transported broilers in 486 crates made from LCS plastic. Once at the slaughterhouse, the vehicle was unloaded and the crates with the broilers were placed in a holding area equipped with fans and sprinkler systems. According to the established slaughter schedule, the slaughter started at 5 a.m. when the crates were tipped over automatically and the broilers were dropped onto a conveyor and transferred to a carousel table from where they were hung on a shackle line.

2.3. Data collection

Dead broilers were removed at the carousel table and accounted by batch or consignment (load) immediately after slaughter and checked by the official veterinarian. The same official veterinarian recorded from 700 carcasses per batch (total 44.800) the number of bruises on wings, legs and breast and from 1.400 carcasses per batch (total 89.600) the number of carcasses condemned by dehydration. Bruises were classified according to the approximate age using visual and objective color assessment standards (Northcutt et al., 2000) so as to consider only those which had occurred during the pre-slaughter period. Dehydrated carcasses were recognised during *post mortem* inspection by being dry, tacky and badly bled (Butterworth & Niebuhr, 2009).

Information regarding age at slaughter (d); mean body weight (kg); batch size (number of broilers per vehicle); time of catching (before/after midnight); team for catching and vehicle; time of arrival at the slaughterhouse; duration of transport (min); distance of transport (km); crate floor area (cm^2/kg); density per crate (kg/m^2); number of birds/crate; lairage duration (min) and water withdrawal duration (min) was collected for the 64 batches.

2.4. Statistical analysis

Generalized linear models (GzLM) were conducted to study the effects of pre-slaughter factors (explanatory variables) on DoA rate (model I), percentage of bruises ($\leq$ 4% or $>$ 4%; model II) and percentage of dehydrated carcasses (model III), using stepwise procedures to select significant predictors on dependent variables (Mccullaghn & Nelder, 1989).

Gaussian errors and log link function was applied for DoA rate (model I) using the initial explanatory variables: body weight, transport duration, transport distance, lairage duration, withdrawal duration, stocking density and catching period. The interaction effects were evaluated for catching period and lairage duration, as well for catching period and transport duration. All of explanatory variables mentioned above were numerical except the catching period which was categorical in a binary scale (before/after 00h).

Binomial errors and logit link function was applied for bruises (model II) using the initial explanatory variables: batch size, team for catching, transport duration, transport distance, lairage duration, withdrawal duration and catching period in a binary scale (before/after 00h). In the stepwise GzLM analysis, variables and their first-level interaction were integrated into the final model if they significantly ($P < 0.05$) changed the deviance. The final models were recalculated with the heterogeneity factor and Akaike Information Criterion (AIC) was measured for goodness of fit. The effect of each factor in the final model was expressed as an odds ratio (OR) and this value is the equivalent to the relative risk, assessing each specific factor relative to its reference class.

Gaussian errors and log link function was applied for dehydration (model III) using the initial explanatory variables: body weight, transport duration, lairage duration, withdrawal duration and catching period (before/after 00h). GzLM model was not found for dehydration and data was analysed thought univariate chi-square for categorical variables: catching period in a binary scale (before/after 00h); lairage duration and withdrawal duration in a 3-point scale ($<$ 8h; $>$ 8h and $<$ 12h; $>$ 12h) and Mann-Whitney for numeric variables: body weight and transport duration. Chi-square and Mann-Whitney tests were

Table 1
Descriptive statistic of pre-slaughter period (n = 64).

Pre-slaughter factors	Mean	SD	Minimum	Median	Maximum
Transport duration (min)	72	5	22	59	184
Velocity of transport (km/h)	55.4	1.8	32.0	52.5	93.0
Transport distance (km)	70.9	6.6	15.0	45.0	196.0
Lairage duration (min)	412	32	17	385	879
Water withdrawal duration (min)	578	30	160	525	1050
Crate floor area (cm^2/kg)	199.5	2.0	172.5	200.8	232.5
Birds per crate (number)	11.1	0.2	8	11	15

performed using SPSS 25.0. GzLM was performed using R.3.52 version Packages MASS.

3. Results

The mean percentage of birds DoA was 0.29 ± 0.21%, ranging from 0.02% to 1.89% per batch. Eleven batches presented DoA higher than 0.5% and two batches presented DoA higher than 1.0%. The batches with DoA rate lower than 0.5%, between 0.5% and 1.0%, and higher than 1.0% were subjected to an average transport distance of 63km, 89km and 171km, respectively. The average mortality in case of short distances (<50km) was 0.23 ± 0.07% and in case of longer distances (>150km) was 0.60 ± 0.10%.

The mean prevalence of bruises on wings, legs and breast was 3.37 ± 0.02%, ranging from 0.43% to 8.29% per batch. Twenty five batches presented bruises in more than 4.0% of birds. Bruises were much frequent on wings (3.06%) compared with the legs (0.19%) and breast (0.12%). Dehydrated carcasses were observed in 22 out of 64 batches, representing 2.68% of condemnations. The characterisation of pre-slaughter period is described in Table 1.

The transport duration (72min ± 5min) and transport distance (70.9 ± 6.6km) were, on average, short. The average withdrawal duration was 578min ± 30min with a maximum of 1020min.

Batches with dehydrated birds presented higher average lairage durations (480min vs. 386min) and higher withdrawal durations (645min vs. 562min), in comparison with those without dehydrated birds.

The GzLM model I analysing the effects of pre-slaughter factors (explanatory variables) on DoA rate are presented in Table 2. The significant effects are represented in Figure 1.

The percentage of birds found DoA increased with transport distance (t = 2.142; P = 0.037; estimate = 0.009). The time of catching also had a significant effect on DoA. For birds caught after midnight, the increase of transport duration increased the birds found DoA. For birds caught after midnight, the increase of lairage duration increased the DoA rate and for birds caught before midnight, the increase of lairage duration did not increase the DoA.

The model II analysing the effects of pre-slaughter factors (explanatory variables) on percentage of bruises (≤ 4% or > 4%) are presented in Table 3. The significant effects are represented in Figure 2.

A higher percentage of bruises (> 4%) occurred in larger batch sizes. Batches with lower stocking density and therefore with a higher floor area of crate per kilogram of body weight (cm^2/kg) also presented a higher percentage of bruises (> 4%). Batches with lower lairage duration and lower transport distance showed higher probability of having bruises' prevalence above 4%. The catch of birds before midnight increased the probability of birds having higher percentage of bruises (> 4%).

The water withdrawal durations had a significant effect on prevalence of dehydrated carcasses (χ^2 = 7.273, df = 2, P = 0.026). A significant effect was observed for catching period and dehydration (χ^2 = 4.403, df = 1, P = 0.036). Birds subjected to longer withdrawal durations (> 12h) and caught before midnight were more likely to become dehydrated.

4. Discussion

The transport of domestic fowl (*Gallus gallus*) is the largest translocation of a single class of livestock in the world. Any problems with transport tend therefore to be important in terms of the number of individuals whose welfare may be affected (EFSA, 2004). In the present study, several pre-slaughter factors were investigated in 64 batches of broilers transported from farms to slaughter to determine potential causes of poor welfare based on indicators collected at the slaughterhouse namely, DoA rate, presence of bruises and of dehydrated carcasses.

The percentage of birds found DoA was 0.29 ± 0.21%, from which 11 batches presented DoA higher than 0.5% and two batches presented DoA higher than 1.0%. The lowest DoA rates were observed for the shortest transport distances and in case of distances over 150km the average mortality was 0.60%. Vecerek et al. (2016) compared mortality rates during transport to slaughter and observed for broilers a DoA rate of 0.15% for a transport distance up to 50km and for transport distances over 200km a DoA rate of 0.54%. A significant effect of transport distance (t = 2.142; P = 0.037; estimate = 0.009) was found for model I on DoA rate, indicating that is expected an increase in DoA of 0.9% with the increase of transport distance in 100km, a result in line with findings obtained by Vecerek et al. (2016), showing average mortality to be very dependent on the distance of travel (Kittelsen et al., 2015). Other large surveys in several countries have found for several species a significant risk of increased mortality rates with journey length (Vecerek et al., 2006; Voslářová et al., 2007; Weeks et al., 2012).

The percentage of birds found DoA was higher when the catching occurred after midnight (t = -2.931; P = 0.005; estimate = -3.813).

Table 2
Effect of explanatory variables on model I for dead on arrival (DoA) rate.

Model for DoA (AIC = 8.573) Explanatory variables	Estimate	Std. Error.	t	P>|t|	exp	Conf. interval (95%)
(Intercept)	-1.649	0.853	-1.932	0.058	0.192	(0.030-0.823)
Catch. per. (+00h)	-3.813	1.301	-2.931	0.005**	0.022	(0.001-0.282)
Transp. dur. (min)	-0.014	0.008	-1.043	0.087	0.986	(0.970-1.001)
Transp. dist. (km)	0.009	0.004	2.142	0.037*	1.009	(1.000-1.019)
Lair. dur. (min)	0.001	0.001	0.724	0.472	1.001	(0.999-1.003)
Catch. per. (± 00h)*Transp. dur. (min)	0.031	0.007	4.109	0.000***	1.031	(1.018-1.049)
Catch. per. (± 00h)*Lair.dur. (min)	0.007	0.002	2.998	0.004**	1.007	(1.003-1.013)

AIC: Akaike Information Criterion; Catch. per.: Catching period; ,+00h: After midnight; Transp. dur.: Transport duration; Transp. dist.: Transport distance; Lair. dur.: Lairage duration; ± 00h: Before/After midnight.
Significant differences:
* $P < 0.05$,
** $P < 0.01$,
*** $P < 0.001$.

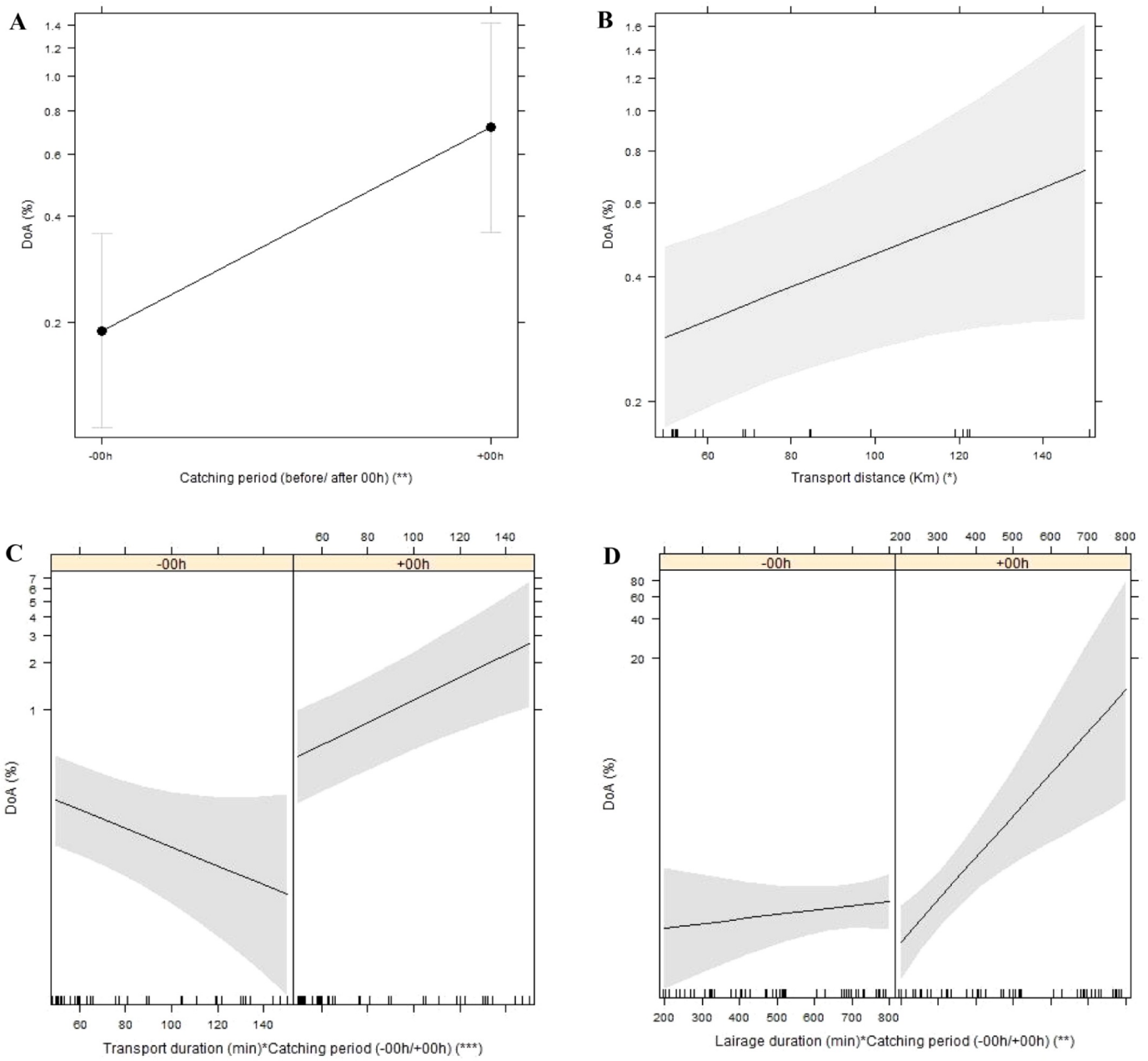

Fig. 1. Effects of catching period (before/after 00h) (A), transport distance (km) (B), transport duration (min) and catching period (before/after 00h) interaction (C) and lairage duration (min) and catching period (before/after 00h) interaction (D) on DoA rate.
DoA: Dead on Arrival; -00h: before midnight; +00h: after midnight.
Values are significantly different at *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$.

Table 3
Effect of explanatory variables on model II for bruises.

Model for bruises (AIC = 81.299) Explanatory variables	Estimate	Std. Error.	t	P > |t|	exp	Conf. interval (95%)
(Intercept)	-8.933	5.502	-1.624	0.104	0.000	(0.000-4.106)
Batch size (No. of birds)	0.001	0.000	2.185	0.029*	1.001	(1.000-1.002)
Catch. per. (+00h)	-2.616	1.101	-2.376	0.017*	0.067	(0.006-0.539)
Transp. dist. (km)	-0.025	0.009	-2.881	0.004**	0.982	(0.942-1.017)
Lair. dur. (min)	-0.006	0.002	-2.542	0.011*	0.994	(0.989-0.998)
Crate floor area (cm^2/kg)	0.044	0.022	1.962	0.049*	1.045	(1.002-1.096)

AIC: Akaike Information Criterion; Catch. per.: Catching period; +00h: After midnight; Transp. dist.: Transport distance; Lair. dur.: Lairage duration.
Significant differences:
* $P < 0.05$,
** $P < 0.01$,

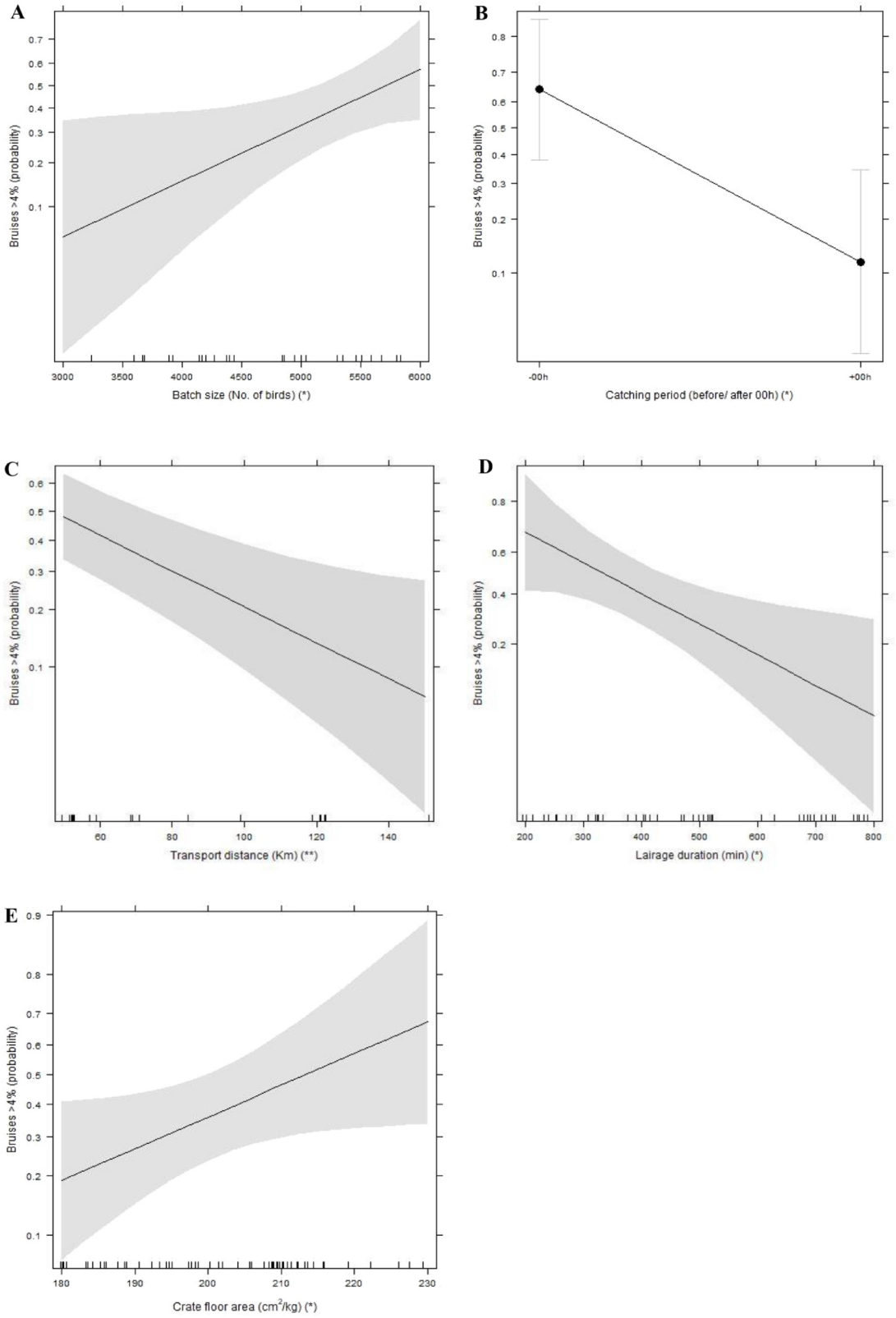

Fig. 2. Effects of batch size (number of birds) (A), catching period (before/after 00h) (B), transport distance (km) (C), lairage duration (min) (D) and crate floor area (cm^2/kg) (E) on percentage of bruises.
-00h: before midnight; +00h: after midnight.
Values are significantly different at *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$.

The interactions between catching period and transport duration (t = 4.109; $P < 0.001$) and between catching period and lairage duration (t = 2.998; $P = 0.004$) indicates that DoA increases with the increase of transport and lairage durations for birds caught after midnight. Nijdam et al. (2004) in a work that sought to determine factors influencing mortality showed that the risk of dying during transport or waiting increases considerably as time increases. According to Cockram and Dulal (2018) the duration of loading, transport, and lairage increases the mortality risk. However, the present study shows that there are other factors which can influence the increase of DoA rate, particularly the time of catching. Therefore, the increased risk of dying with longer time in transport or waiting could be problematic in case of a late catching period. According to the present study, a late catching must occur in farms near the slaughterhouse and have short lairage durations so as to reduce DoA. Jacobs et al. (2017) observed that after nocturnal transportation, birds show less change of body temperature compared to morning transportation, indicating that birds experienced less stress at night than in the morning. Comparing *post mortem* findings in dead-on-farm and DoA broilers, Kittelsen et al. (2015) found that lung congestion and trauma was present in a greater percentage of birds that died during transportation than in those that died on the farm. Furthermore, Lund et al. (2013) underlined the importance of increased focus on handling based on the chronicity of the lesions found on DoA broilers, which were primarily related to management and handling procedures. Moreover, climatic conditions have been found to be associated with DoA (Chauvin et al., 2011), being heat stress recognized as a major risk factor (Mitchell & Kettlewell, 2004; Petracci et al., 2006; Whiting et al., 2007). Seasonal variation was not relevant in this study since it was carried out in spring, however a higher DoA rate would be expected in summer and winter months, as confirmed by several reports (Vecerek et al., 2006; Vieira et al., 2011).

The average percentage of birds with bruises was 3.37 ± 0.02%, ranging from 0.43% to 8.29% per batch. Batches with more birds (t = 2.185; $P = 0.029$) presented a higher prevalence of bruises. Furthermore, batches with a lower stocking density and more space per crate (t = 1.962; $P = 0.049$) presented more bruises, which is in line with findings obtained by Knowles and Broom (1990), indicating that transport systems with less space per bird can be more suitable in preventing bruising. This may be explained by the fact that birds sustain each other's body, reducing falling or the need to spread the wings and legs to keep balance. However, the present study demonstrated that bruises did not increase with transport duration indicating that bruises were more likely to have occurred on farms during catching, crating and loading. According to Vosmerova et al. (2010), pre-transport handling procedures may be more stressful for broilers than the transport itself. The main hazard responsible for occurrence of bruises is inadequate handlers (Marahrens et al., 2011) and individual features of the catching teams' elements might explain different degrees of lesions (Langkabel et al., 2015). Stocking density can also be managed to lessen negative influences on animal welfare (Fisher et al., 2009). Birds may benefit from slightly higher densities in spring, or if weather conditions are anticipated to be cold (EFSA, 2004; Marahrens et al., 2011). The prevalence of bruises was much higher in the wings (3.06%) when compared to the legs (0.19%) and breast (0.12%). Catching the birds before midnight also contributed to the increase of bruises (> 4%) and batches with more birds per crate presented more bruises (Figure 2). These results can be related to the first catching (before midnight) usually occur in pavilions of broilers with less body weight where catchers hold more birds simultaneously while have less control of birds movement. This difficult the placement of bird inside the crate, increasing agitation and striking the crate entrance. Thus, lesions on the wings usually occur during crating because of an increase in wing flapping, and when a large number of birds are squeezed into the same crate. Birds should be handled cautiously, tranquillity of the flock should be maintained as to avoid wing flapping, and modular containers should be positioned as close to the animals as possible (Langkabel et al., 2015; Fisher et al., 2009). The setting up of standard operating procedures might help to attain a situation within which the welfare of the animals is maintained at all stages of catching and crating (Langkabel et al., 2015).

The maximum transport duration of 184min and distance of 196km were not very high. In contrast, the lairage duration was high with mean value of 412min and maximum of 879min, as well as the water withdrawal duration with mean value of 578min and maximum of 1050min. Twenty one batches (32.81%) were subjected to more than 12h (720min) of fasting and even with good weather, dehydrated carcasses were observed in 22 out of 64 batches. In commercial practice, water is usually withdrawn just before the first bird of a flock is caught and crated for transport to the slaughterhouse. This depopulation process often takes several hours and water deprivation continues as the birds are transported to the slaughter plant (Delezie, 2006). According to the Council Regulation 1/2005, suitable water shall be available in adequate quantities in the case of a journey lasting more than 12h, although this is hardly feasible for birds (Council Regulation No 1/2005). This is particularly important in hot weather, due to the increased risk of birds restricted from access to water easily became dehydrated, and depending on their age and physiological state, birds vary in the ability to cope with periods of feed and water withdrawal (Fisher et al., 2009). Moreover, the duration of the pre-slaughter stages, the thermal environment, fasting, ill-health, and injury can reduce the physiological capacity of the birds to maintain homoeostasis which can result in exhaustion and death (Caffrey et al., 2017; Cockram & Dulal, 2018). The welfare of broilers during the pre-slaughter can be greatly improved through changes in the human approach, by the implementation of standard operating procedures and by ensuring adequate planning. Training is essential for those involved in handling animals and driving vehicles (Northcutt et al., 2000). Planning the catching, loading and transport from the farm in coordination with planned slaughter times is essential and it will ensure that birds that have been the longest without feed and water are the first to be killed. All those involved should consider carefully the weather conditions, the transport distances and the transport time (night/day), as well as, the body weight and the health state of birds. It is emphasized that when a broiler transport vehicle is stationary, or when a modular load is stacked in lairage, the ventilation of the transport containers is entirely passive and dependent upon buoyancy forces. The reduced ventilation of the load in these circumstances may not be adequate to fully dissipate the heat and moisture loads produced by the birds. This in turn will increase the risk of thermal (heat) stress and possibly, an increase in mortality. It should be stressed, perhaps that in many circumstances the term DoA is inappropriate as birds may die as a result of the factors described above rather than on the journey, and often mortality in transport is not estimated until the birds are hung on the shackle line. Thus, birds should be unloaded from vehicles as soon as possible after arrival, avoiding unnecessary delay. Broilers should be placed immediately in environmentally controlled lairage areas, as this will reduce stress and reduced welfare during standing and lairage and may decrease apparent DoA in addition to avoiding negative impacts on carcass and meat quality (e.g. live shrink).

5. Conclusion

Important risk factors affecting broiler welfare during transport from farms to slaughterhouses can be identified by assessing welfare indicators such as DoA rate, presence of bruises and dehydration. In the present study it was shown that pre-slaughter operations should be adequate planned and carried out for short transport distances and lairage duration, catching the birds by night or before midnight and ensure adequate catching and crating procedures. Close attention to, and control of, all of these factors is essential to ensure high standards of animal welfare in the transportation of broiler chickens.

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