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Establishment of cumulative assessment groups of pesticides for their effects on the nervous system

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Abstract

Cumulative assessment groups of pesticides have been established for five effects on the nervous system: brain and/or erythrocyte acetylcholinesterase inhibition, functional alterations of the motor, sensory and autonomic divisions, and histological neuropathological changes in neural tissue. Sources of uncertainties resulting from the methodological approach and from the limitations in available data and scientific knowledge have been identified and considered. This report supports the publication of a scientific report on cumulative risk assessment to pesticides affecting the nervous system, in which all uncertainties identified for either the exposure assessment or the establishment of the cumulative assessment groups are incorporated into a consolidated risk characterisation.

Keywords: cumulative risk assessment, cumulative assessment groups, pesticides mixtures, nervous system, motor division, acetylcholinesterase inhibition

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Summary

From all possible effects of pesticides on the nervous system, five were found to be meeting the criteria established by the EFSA Panel on Plant Protection Products and their Residues (PPR Panel) and specific for consideration in cumulative risk assessment (CRA) (EFSA PPR Panel, 2013a). These specific effects were brain and/or erythrocyte acetylcholinesterase (AChE) inhibition, functional alterations of three divisions of the nervous system (motor, sensory and autonomic functions) and histological neuropathological changes in neural tissues. There was insufficient information to address the combined effects of pesticides with respect to developmental neurotoxicity and cognitive effects.

A cumulative assessment group (CAG) was established for each of the five specific effects and more than 400 active substances (AS) were screened for potential inclusion in these CAGs. Any AS possessing a chemical structure associated to a mode of action (MoA) of direct relevance for the effect or exhibiting selected indicators (toxicological endpoints) reflecting the specific effect in regulatory toxicological studies was included in the respective CAG.

In total, 47 ASs were included in the CAG for brain and/or erythrocyte AChE inhibition. They were 119 to be included in the CAG for functional alterations of the motor division, while the CAGs for functional alterations of the sensory and autonomic divisions both contained 101 ASs. The CAG for histological neuropathological changes in neural tissues contained 19 ASs only. All ASs included in the CAGs were characterised by no observed adverse effect levels (NOAELs) for short- and long-term cumulative exposure/risk assessment, derived from the most sensitive indicator, using all available information across studies, species and sexes. Index compounds (ICs) have been proposed to enable cumulative exposure and risk assessments with methods using relative potency factors (RPFs).

Based on the number and NOAELs of ASs in each CAG, it would be sufficient to perform CRAs with the CAGs for brain and/or erythrocyte AChE inhibition and for functional alterations of the motor division to cover the combined effects of pesticides associated with all five CAGs.

The number and the identity of the ASs included in the CAGs, as well as the allocated NOAELs, are subject to uncertainties. Sources of uncertainty resulting from the methods used to collect and assess toxicological data and from the limitations in the available data and scientific knowledge were therefore identified for appropriate consideration during the CRA conducted with these CAGs. The identified sources of uncertainty were related to the composition of the CAGs, the toxicological characterisation of the ASs, the slope and shape of the dose–response relationship, the contribution of metabolites and degradation products, the adequacy of the dose-addition model and the inter- and intraspecies differences in toxicological sensitivity.

With respect to the composition of the CAGs, the uncertainty about the total number of ASs in the CAG for functional alterations of the motor division that actually cause the effect was thoroughly addressed using weight of evidence and expert knowledge elicitation (EKE) techniques. In this process, ASs were allocated in subgroups of varying levels of evidence and a median estimate of 104 was derived for the number of ASs actually causing functional alteration of the motor division. A similar exercise was not conducted with the CAG for brain and/or erythrocyte AChE inhibition because the association between the chemical structure and the MoA is obvious for organophosphorus and *N*-methyl carbamate insecticides.

A mechanism for periodic update of the CAGs established in the present report should be put in place by EFSA in order to make use of relevant new information. It is also recommended to deploy a testing and assessment strategy to provide enough information supporting the establishment of CAGs covering developmental toxicity and, in the future, to characterise the ASs included in the CAGs using, as reference points, lower confidence limits of a benchmark dose (BMDL) suitable for regulatory purpose to remediate to the uncertainty resulting from the use of NOAELs.

This report should be read in conjunction with the EFSA scientific report on cumulative dietary exposure assessment to pesticides that have acute effects on the nervous system using SAS® software (EFSA, 2019a), the RIVM scientific report on cumulative dietary exposure assessment of pesticides that have acute effects on the nervous system using MCRA software (van Klaveren et al., 2019) and the EFSA scientific report on the cumulative dietary risk characterisation of pesticides that have acute effects on the nervous system (EFSA, 2019b).

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1. Introduction

Regulation (EC) No. 396/2005 on maximum residue levels (MRLs) of pesticides in or on food and feed provides that cumulative and synergistic effects of pesticides should be taken into account for dietary risk assessment when appropriate methodologies are available. Regulation (EC) No. 1107/2009 concerning the placing of plant protection products on the market also provides that the residues of the plant protection products shall not have any harmful effects on human health, taking into account known cumulative and synergistic effects where the scientific methods accepted by the Authority to assess such effects are available.

In view of this legal context, the European Food Safety Authority (EFSA) and the Panel on Plant Protection Products and their Residues (PPR Panel) started in 2007 the development of the necessary methodologies to carry out cumulative risk assessment of pesticide residues. This methodological development included a procedure to establish cumulative assessment groups (CAGs) of pesticides on the basis of their toxicological profile (EFSA PPR Panel, 2013a).

1.1. Background and Terms of Reference

In 2014, EFSA started a programme of activities aimed at implementing the cumulative risk assessment (CRA) of pesticides, using the methodologies developed by the PPR Panel. As part of this program, the Pesticides Unit (nowadays Pesticides Residues and Pesticides Peer Review units) has been requested by EFSA to prepare a scientific report on CAGs of pesticides for their effects on the nervous system.

1.2. Purpose of this scientific report

The EFSA implementation plan for CRA also requested the Pesticides Unit to carry out retrospective CRAs for the effects of pesticides on the nervous system, using the results of official controls conducted by Member States under the annual monitoring programmes foreseen by Regulation (EC) No 396/2005.

These assessments will use the CAGs established in the present report and will be presented in a separate EFSA scientific report which will deal with the following assessment questions:

- What is the acute cumulative risk of brain and/or erythrocyte acetylcholinesterase (AChE) inhibition resulting from combined dietary exposure to pesticide residues?
- What is the acute cumulative risk of functional alteration of the motor division of the nervous system (e.g. locomotor activity, muscle strength, coordination and equilibrium) resulting from combined dietary exposure to pesticide residues?

These CRAs will be conducted under the assumption of dose addition (EFSA, 2008). In 2015, European Commission informed EFSA that the Standing Committee on Plants, Animals, Food and Feed (PAFF Committee) agreed on the use of the combined margin of exposure (MOET, also known as Total Margin of Exposure) concept as the mode of expression of cumulative risks (see Section 2.2.3 for details on method). The CAGs established in the present report are compatible with this concept.

1.3. Precautionary principle and uncertainties

Regulation (EC) No 1107/2009 concerning the placing of plant protection products on the market does not prescribe precisely how CRA of pesticides should be performed. However, it provides that Member States 'shall not be prevented from applying the precautionary principle where there is scientific uncertainty as to the risks with regard to human and animal health' and 'shall take into consideration possible element of uncertainty in the information in order to ensure that the chances of failing to detect adverse effects or of underestimating their importance are reduced to a minimum'. These provisions are valid for the assessment of cumulative effects of pesticides and have been prevailing in the elaboration of principles to establish CAGs of pesticides by the PPR Panel (EFSA PPR Panel, 2013a,b), in view of the large areas of uncertainty related to the combined toxicity of chemicals on human health. This might contribute to explain differences with approaches developed under other jurisdictions with respect to the grouping strategy.

In this context, as the forthcoming CRAs will be performed using exclusively the active substances (ASs) included in the CAGs and following the dose-addition model, an uncertainty analysis will be conducted in order to appreciate how using the CAGs as established in this report may under- or overestimate the actual risk to consumers, as formulated in the above assessment questions. To prepare for this, this report will consider the following question:

- How sure is it that the CAG contains all the ASs causing the specific effect and only ASs causing this effect?
- How sure is it that these ASs combine their individual toxicities according to the dose-addition model at their actual level in food?

2. Data and methodologies

2.1. Data

Three data collections were carried out to retrieve information supporting the establishment of CAGs of ASs of plant protection products for their effects on the nervous system. Only chemical ASs were considered in these data collections.

The first of these data collections (RIVM, ICPS, ANSES, 2013) was outsourced to a consortium of the Dutch National Institute for Public Health and the Environment (RIVM), the International Centre for Pesticides and Health Risk Prevention in Italy (ICPS) and the French Agency for Food, Environmental and Occupational Health and Safety (ANSES). It covered the ASs approved until 31 May 2009 and identified as having effects on the nervous system by the Danish Technical University (DTU), under an earlier grant awarded by EFSA (Nielsen et al., 2012), and all ASs approved between 1 June 2009 and 31 December 2011. This data collection was used by the PPR Panel to established a first proposals of CAGs in 2013 (EFSA PPR Panel, 2013a) The sources of this data collection were official documents produced during the approval of ASs under Directive 91/414/EEC and Regulation (EC) No 1107/2009: Draft Assessment Reports (DARs), Renewal Assessment Reports (RARs) as well as the respective Addenda, peer review experts' meeting reports, EFSA conclusions and European Commission review reports. Original study reports submitted by applicants during the peer review process were also occasionally consulted when the sources of information were insufficiently detailed. Additionally, Joint Meeting on Pesticide Residues (JMPR) evaluation reports or open literature (e.g. PubMed) were also searched for additional information on modes of action (MoAs). All available acute and repeated dose *in vivo* toxicological studies performed by oral administration in mammals were considered. *In vitro* studies were used when they provided information on known or presumed neurotoxic MoAs. For each AS covered by this data collection, the main principles followed by the contractor were as follows:

- Both acute and repeated dose effects were reported.
- For each endpoint related to neurotoxicity, only the no observed adverse effect level (NOAEL) and lowest observed adverse effect level (LOAEL) observed in the most sensitive species and most sensitive sex were collected. In case the NOAELs in two different species were almost identical, the NOAELs and LOAELs for both species were recorded. Higher NOAELs and LOAELs observed in additional studies were not recorded. Comparable studies with respect to study design and strain of animals were combined to derive overall NOAELs and LOAELs.
- Observations indicative of neurotoxicity observed at (near) lethal doses were not collected when the reviewer attributed these effects to general toxicity.
- In case in a single study several critical endpoints were observed at the same NOAEL/LOAEL, they were recorded separately.
- Human data were always collected, even if the NOAELs/LOAELs for a certain endpoint were higher than those obtained in animal studies.
- Effects on AChE were reported when the inhibition was statistically significant ($p < 0.05$) and reached at least 20% decrease of the control level.
- Collected NOAELs which were lower than the NOAELs that formed the basis for the acceptable daily intake (ADI) or acute reference dose (ARfD) were flagged.

The second data collection was outsourced to the same consortium (RIVM, ICPS, ANSES, 2016). It covered all ASs approved after 1 January 2012 and until 31 May 2013, a number of new ASs not yet approved but pending approval at that time, and an additional list of non-approved ASs present in the European Union (EU) consumer's diet as evidenced in the 2011 Annual report on the Rapid Alert System for Food and Feed (European Commission, 2011) and in the 2010 Annual Report on Pesticide Residues in Food (EFSA, 2013). The sources of this data collection were official documents produced during the approval of ASs under Regulation (EC) No 1107/2009: DARs, RARs as well as the respective addenda, peer review expert meeting reports, EFSA conclusions and European Commission review reports. If necessary, original study reports were consulted for more details. When a European evaluation was not available or was outdated, assessment reports from recognised international bodies (e.g. JMPR, United

States Environmental Protection Agency (US-EPA)) were scrutinised. All repeated dose (short-term and long-term) toxicological studies based on oral administration (diet, gavage, capsule) were considered. *In vitro* studies were also used for information on MoAs. In contrast with the first data collection, the second data collection was organised in accordance with the specific effects identified for the nervous system by the PPR Panel (EFSA PPR Panel, 2013a) and their respective indicators. For each AS covered by this data collection, the main principles followed by the contractor were as follows:

- All studies rated as 'acceptable' or 'supportive' from all animal species reported in the regulatory documents with observations of specific effects were considered (mainly rat, mouse and dog).
- When more than one specific effect was observed for an AS in one study, each of them was collected under a separate entry.
- NOAELs/LOAELs for a same indicator of specific effect that were overlapping in two or more studies of the same duration in the same species were not combined and were reported in separate entries.
- The lowest NOAEL/LOAEL for a specific effect observed in the most sensitive sex in the study has been reported.
- When several indicators of a specific effect have been observed in one study, the most sensitive indicator(s) has been indicated in the column 'Endpoint of a specific effect', and the others have been reported in the column 'Remarks about the effect'.
- NOAELs/LOAELs for a specific effect have been collected regardless of the respective reference values (ADI/ARfD).
- Cases where age-related changes were not clearly separated from treatment-related effects were flagged.
- Information on statistical significance tests regarding observations in the studies was collected.
- Any limitation which could have had an impact on the acceptability of the study and the evaluation/occurrence of the specific effect was flagged.

In addition, EFSA conducted an internal complementary data collection to consolidate the information regarding 24 ASs, following comparable principles.

All the details of the data collections can be found in the respective external scientific report (RIVM, ICPS, ANSES, 2013, 2016) and the resulting data collection spreadsheets. The collected information slightly evolved over time based on the growing experience about the exact information needed to establish CAGs. It is acknowledged that the most recent data collection was performed with higher quality standards and that some relevant information might have been omitted in the previous data collections.

The complete list of ASs (422 in total) covered by these data collections is given in Appendix A.

2.2. Methodologies

The establishment of CAGs followed a sequence of tasks comprising the identification of the specific effects on the system or organ considered, the definition of the hazard characterisation principles of these specific effects, the establishment of CAGs, the selection of an Index Compound (IC) and an analysis of uncertainties about the adequacy of the CAG with respect to the specific effect.

2.2.1. Identification of the specific effects

From all the effects of pesticides observed on the system or organ considered, this step consisted in identifying those which should be considered in CRA. Such effects, which can result from a combined action of pesticides, were generically designated as 'specific effects' in this report. This identification was based on information analysis and expert judgement aimed at:

- Excluding local effects: Local effects, not being produced by the potentially absorbed dose, were excluded. Furthermore, they do not form the basis of reference values in regulatory dietary risk assessment.
- Excluding non-adverse effects: Non-adverse effects are not used as basis for setting a toxicological reference value and were therefore also not considered as relevant for CRA. In discriminating between an adverse and a non-adverse effect, consideration was given to its adaptive nature, its transient or persistent nature, its magnitude, its association with other alterations, whether it was a precursor to a more relevant effect, and its impact on the overall function of the organism (Lewis et al., 2001; EFSA PPR Panel, 2013a).

- Excluding effects not relevant to humans: Effects not considered as relevant for human were not relevant for cumulative risk assessment.
- Evaluating the unambiguous nature of the effect: A specific effect needed to be unambiguous and well-defined in terms of site and nature.

These criteria were developed by the PPR Panel in 2013 (EFSA PPR Panel, 2013a) and resulted in CAGs of pesticides causing either a common phenomenological effect, or, in some cases where underlying MoAs are known, a common biochemical effect.

2.2.2. Characterisation of the specific effects

This step established the hazard characterisation principles applicable to the identified specific effects. In practice, this meant defining the descriptors/indicators of specific effects (endpoints) observed in toxicological studies building evidence that an AS causes the specific effect and deciding how NOAELs are derived to characterise the AS for this specific effect. This was done on the basis of the information available in application of the regulatory data requirements, following the respective study guidelines and in a way to ensure equal treatment of all ASs. If this was not the case, this had to be clearly highlighted.

2.2.3. Establishment of CAGs and selection of ICs

For each specific effect identified in the first step of the process, a CAG was established.

The population of each CAG by the appropriate ASs was based on a critical analysis of the information collected as described in Section 2.1. For each specific effect, the criteria used to perform this critical analysis were described with enough details to enable an independent assessor to repeat it.

Once CAGs were populated, one of the ASs was selected as the IC. The approach used to select the IC was defined on an ad-hoc basis for each specific effect, as explained in Sections 3.3.1 and 3.3.3.

However, it needs to be highlighted that any of the ASs of a CAG can be used as an IC without any impact on the MOET, and that a MOET can also be calculated without any IC.

Indeed, two options are possible to calculate MOET:

Directly, by calculating the reciprocal of the sum of the reciprocals of individual margins of exposure (MOEs) to each chemical contributing to the risk (EFSA, 2008):

$\frac{1}{MOET} = \frac{1}{MOE_1} + \frac{1}{MOE_2} + \frac{1}{MOE_3} \ldots + \frac{1}{MOE_n}$, where MOE_i is the margin of exposure for the i th chemical,

$MOE_i = \frac{RfP_i}{E_i}$ and RfP_i is the toxicological reference point (e.g. NOAEL, lower confidence limit of a benchmark dose (BMDL)) for chemical i and E_i its exposure.

Indirectly, by determining the sum of potency-normalised individual exposures as total IC equivalents and translating the IC equivalents into the MOET to the reference point of the IC. This approach, however, requires additional work to select an IC and calculate a relative potency factor (RPF_i) for each chemical.

$RPF_i = \frac{RfP_{IC}}{RfP_i}$ where RfP_{IC} and RFP_i are the reference points for the IC and chemical i,

$MOET = \frac{RfP_{IC}}{\sum_i E_i \times RPF_i}$ where the denominator sums over all chemicals including the IC.

The present report was elaborated in such a way to make both options possible. In particular, it includes the selection of ICs for each established CAG. It should be noted that direct or indirect calculations lead exactly to the same results. This is demonstrated as follows:

$\frac{1}{MOET} = \frac{\sum_i E_i \times \frac{RfP_{IC}}{RfP_i}}{RfP_{IC}}$ inverting the previous equation and substituting for RfP_i

$\frac{1}{MOET} = \frac{\sum_i E_i \times \frac{\cancel{RfP_{IC}}}{RfP_i}}{\cancel{RfP_{IC}}}$ cancelling out RfP_{IC} in numerator and denominator

So: $\frac{1}{MOET} = \sum_i \frac{E_i}{RfP_i} = \frac{1}{MOE_1} + \frac{1}{MOE_2} + \frac{1}{MOE_3} \cdots + \frac{1}{MOE_n}$ as in the direct calculation above.

An important consequence of this is that the choice of the IC has no influence at all on the result of the assessment, nor on the uncertainties affecting the MOET. This is because any change in RfP_{IC}, e.g. through choosing a different IC or errors in the RfP of the IC, affects both the numerator and denominator of the equation and cancels out, as shown above.

In order to perform the CRAs mentioned in Section 1.2 of the present report, cumulative exposure assessments were performed, either using ICs (Van Klaveren et al., 2019) or not (EFSA, 2019a).

2.2.4. Analysis of uncertainties

The CAGs established in this report were used to carry out cumulative exposure and risk assessments following the methodology developed by the PPR Panel. This methodology assumes that all ASs included in a CAG combine their effects by dose addition. To inform on whether the results tend to either over- or underestimate the actual risks, uncertainties relating to two questions have been considered.

Question 1

How sure is it that the CAG contains all the ASs causing the specific effect and only ASs causing this effect?
If the CAG does not contain all ASs causing the specific effect, the results of the assessment will tend to underestimate the risk. If, in contrast, it includes ASs not causing the effect, the results of the assessment will tend to overestimate the risk.

Question 2

How sure is it that these ASs combine their individual toxicities according to the dose-addition model at the actual dietary exposure level? Where possible, clusters of ASs for which dose addition is virtually certain should be defined.
The rationale of using dose addition to perform CRA of pesticide residues was given in the Scientific Opinions of the PPR Panel on the identification of pesticides to be included in CAGs on the basis of their toxicological profile (EFSA PPR Panel, 2013a) and on the relevance of dissimilar MoA and its appropriate application for CRA of pesticides residues in food (EFSA PPR Panel, 2013b).
Although dose addition is expected in principle when chemicals in a mixture act by the same MoA, and differ only in their potencies, its use is recommended by the PPR Panel to assess the cumulative effects of pesticides eliciting the same adverse effect by different MoAs. Similarly, the ESFA Scientific Committee recommends adoption of the mixture assessment concept of dose addition as a pragmatic and precautious default assumption, unless there are indications that the alternative concept of response addition is more appropriate (EFSA Scientific Committee, 2019).

For the CAGs subject to a CRA as indicated in Section 1.2 (effects on motor division and brain and/or erythrocyte AChE inhibition), Question 1 was addressed in the present report using a combination of weight of evidence and expert knowledge elicitation (EKE) techniques, described in the following section. With respect to Question 2, this report reviewed the available information regarding the MoAs leading to these effects, but a full assessment, relying on expert judgement, was only possible during the respective CRAs (EFSA, 2019b) after identification of the ASs driving the risks.

For the other CAGs (functional alteration of the sensory and autonomic functions, histological neuropathological changes in neural tissue), a similar exercise was not done, because these effects are less critical than functional effects on the motor division in terms of cumulative risks (see Section 3.3.3).

2.2.5. Weight of evidence and expert knowledge elicitation technique

The amount, reliability, relevance and consistency of evidence for causing effects on the nervous system vary between ASs. This makes it uncertain which substances should be included in a given CAG, with some substances being more likely to belong to a certain CAG than others. This can be quantified by assessing the probability that each substance actually causes the specific effect. This could be done separately for each substance but, due to the large number of substances involved, it was more practical to form subgroups of substances for which the weight of evidence is similar, and then assess what proportion of chemicals in each subgroup causes the effect. This was done by developing a structured procedure which combines techniques for weight of evidence assessment (EFSA Scientific Committee, 2017) and EKE (EFSA, 2014). This procedure comprised the following sequence of tasks:

1) Defining in precise terms the specific effect that is to be assessed.
2) Identifying lines of evidence that were important for assessing whether the AS causes the effect: lines of evidence typically included the indicators as defined in Section 2.2.2 but were not necessarily restricted to these indicators. Depending on the specific effect, additional factors contributing to the evidence could be defined.
3) Rating the weight of each line of evidence: the lines of evidence were assessed with respect to their reliability and relevance to the assessment question. This assessment was conducted by expert discussion and resulted in the allocation of a coefficient or weight to each line of evidence, varying from 1 to 10 and which was a relative measure of the contribution that positive findings for each line of evidence results in an increase of the probability of a chemical causing the effect.
4) Reviewing the evidence for each AS included in the CAG in order to identify which lines of evidence are positive.
5) Integration of the lines of evidence by multiplying all coefficients corresponding to the lines of evidence for each AS. This gave a score to each AS which is proportionate to the number and strength of the positive lines of evidence and reflects the overall weight of evidence on whether the AS is causing the effect. The individual and aggregated scores for every substance were recorded and colour coded in a large table (see Annex A), to facilitate their use by the experts in the following steps.
6) Clustering the ASs in different groups of similar weight of evidence on the basis of their score. This was done by ordering the ASs in decreasing order of the calculated scores, identifying points in the ranked list where there are large changes in score, and using this to inform decisions about how to divide the list into subgroups. These decisions were made by expert discussion, balancing the need for a practical number of subgroups against the homogeneity of scores and lines of evidence within each subgroup.
7) Assessing how many of the ASs in each subgroup actually cause the specific effect. This was done by a structured EKE procedure, using a modified version of the 'Sheffield' EKE protocol described by EFSA (EFSA, 2014) to elicit a discrete probability distribution quantifying the experts' uncertainty about the number of substances in each subgroup that actually cause the effect. For each subgroup, experts first worked individually, reviewing the evidence and making their own judgements. This was not based simply upon the weight of evidence scores, but on evaluation of all relevant considerations (e.g. information on the MoA) using expert judgement. This was followed by a facilitated discussion of the individual distributions and reasoning, leading to agreement on a consensus distribution and reasoning for each subgroup. Both the individual and consensus distributions were elicited using the 'roulette' method (EFSA, 2014, pp. 169–170), as this is well suited to eliciting a discrete distribution and the experts found it easy to use when making their judgements. Finally, results for all the subgroups were displayed together for the experts to review and, where necessary, adjust.
8) When developing the consensus distribution for some of the more diverse subgroups, the experts found it helpful first to divide the subgroup into subsets of substances for which the probability of causing functional alterations of the motor division was thought to be similar, express those probabilities on an approximate scale, and then use this to inform their collective judgement on the consensus distribution for the subgroup as a whole.
9) The elicited distributions for the subgroups were combined by 1D Monte Carlo simulation (EFSA Scientific Committee, 2018) to calculate a probability distribution for the total number of substances that actually cause the specific effect. This was done twice, first assuming independence between subgroups and then assuming perfect positive dependence, to explore the potential impact of dependency on the results.

The results of this procedure comprised (a) a probability distribution for the number of substances in each subgroup that cause the specific effect, each with accompanying rationale, and (b) two probability distributions for the total number of substances causing the effect, one assuming independence between subgroups, and the other assuming positive dependence.

Additional sources of uncertainties will be considered in a subsequent report when assessing overall uncertainty in the CRA (EFSA, 2019b).

3. Assessment

3.1. Identification of the specific effects

On the basis of the results of the project commissioned by EFSA to DTU (Nielsen et al., 2012) and of the first data collection performed by the consortium RIVM/ICPS/ANSES (RIVM, ICPS, ANSES, 2013), the PPR Panel identified five specific effects of pesticides on the nervous system (EFSA PPR Panel, 2013a), which were confirmed as follows:

- Functional alteration of the motor division of the nervous system (e.g. locomotor activity, muscle strength, coordination and equilibrium).
- Functional alteration of the sensory division of the nervous system (e.g. reflex action, sensory motor responses).
- Functional alteration of the autonomic division of the nervous system (i.e. modulation of the autonomic activity by cholinergic neurons).
- Brain and/or erythrocyte AChE inhibition (neurochemical effect).
- Histological neuropathological changes in neural tissue (e.g. axonal degeneration and demyelination).

The rationale behind the identification of these effects by the PPR Panel was given in details in the Scientific Opinion. It considered the high complexity of the nervous system with respect to its anatomic organisation and variety of physiological functions.

With respect to the criteria listed in Section 2.2.1, all specific effects result from systemic exposure, are adverse, relevant for humans, specific and can be observed as primary effects.

The functional alterations of the motor, sensory and autonomic divisions of the nervous system are phenomenological effects considered as relevant for CRA, because, although of variable nature at biochemical level, they concern very specific and specialised functions of the organism.

AChE inhibition is quantified by objective instrumental measurements, directly related to the MoA. Therefore, the unambiguousness of the nature of this effect is strongly demonstrated.

AChE inhibition and functional alterations of the motor, sensory and autonomic divisions of the nervous system, by their nature, can be triggered by acute and chronic exposures.

Neuropathological effects are unambiguously associated to histopathological observations, which can also be of variable nature, although consisting in most cases of axonal and myelin degeneration. As they commonly result in the alteration of the specialised function of nervous cells, they are also considered as specific effects justifying the establishment of a CAG. Owing to its nature, this specific effect is triggered by chronic exposures only.

Interdependencies

There are interdependencies between these specific effects because they are associated to common MoAs. For instance, some pesticide targets (e.g. neurotransmitter receptors, ion channels) are expressed in some but not all neurons. These targets can be found in the central nervous system or in the peripheral nervous system, or in both. However, given the tremendous complexity of the nervous system and the huge number of different tasks it performs, neurons play different roles, and thus they can be divided into three classes: sensory neurons, motor neurons, and interneurons (which connect one neuron to another). Interneurons are thus involved in processing information, both in simple reflex circuits (consisting of only a few neurons) and in more complex circuits in the brain (which involve more complex neuronal networks, i.e. integrated nervous pathways). For these reasons, the major pesticide classes with known MoA can impair the motor, sensory and autonomic function despite interacting with different neuronal targets. Hence, pesticides targeting gamma-aminobutyric acid (GABA) receptors in interneurons can induce either motor or sensory signs of toxicity because of the physiological role of these neurons in the nervous system. This is reflected in the data which show that many ASs with different MoAs affect all 3 functional divisions of the nervous system: motor, sensory and autonomic (see Section 3.3.1).

AChE inhibition is obviously an important mechanism of neurotoxicity leading to functional alterations observed in the motor, sensory and autonomic divisions of the nervous system. This interdependency should however not suggest that some assessments might be seen as refinements of other assessments in the usual sense of this word when used in regulatory pesticide risk assessment. Indeed, the risk assessments which will be conducted with respect to these effects will each address a specific assessment question, e.g. 'What is the risk of functional alteration of the motor division of the

nervous system resulting from the exposure to pesticide residues?' or 'What is the risk of cholinesterase inhibition resulting from the exposure to pesticide residues?'. These questions are different and need to be addressed through individual assessments.

Effects not leading to CAGs

Based on the information collected by DTU (Nielsen et al., 2012), a number of reported effects of pesticides on the nervous system were not considered as relevant for CRA, because they were either not adverse, observed at doses above the maximum tolerated dose (MTD) (e.g. vacuoles in brain) or not statistically significant and within the historical control range (i.e. induction of neoplasm, particularly observed astrocytoma).

Neurochemical effects of pesticides on the nervous system, other than AChE inhibition, were not proposed as specific effects because they are not directly measured in regulatory studies.

Owing to the lack of specific requirements for developmental neurotoxicity (DNT) testing of pesticides in EU at the time of the Scientific Opinion of the Panel (EFSA PPR Panel, 2013a), the number of studies of this type available to the panel was not enough to propose specific effects and CAGs with respect to this type of toxicity. Therefore, DNT was not addressed in this report.

Similarly, behavioural tests assessing the effects of pesticides on the cognitive function (e.g., learning and memory) are often used as a higher tier of neurotoxicity evaluation. Because of the scarcity of information related to this type of test in the data collection, the effects of pesticides on this function were not addressed in this report.

3.2. Characterisation of the specific effects

All indicators of effects of pesticides on the nervous system were reviewed in view of characterising the five specific effects. In this process, several observations (such as prostration, opisthotonus, laboured breathing, tachypnoea, dyspnoea, exophthalmos, lethargy, coma, hypothermia, emesis and alopecia) were found as often occurring secondary to general systemic toxicity after high doses. Accordingly, they were not deemed appropriate observations to characterise any of the five specific effects.

3.2.1. Functional alteration of the motor division of the nervous system

The specific indicators of toxicity observable in toxicological studies contributing to the evidence that an AS causes a functional alteration of the motor division of the nervous system were classified in four categories:

- Reduced motor activity: hypoactivity, recumbency (if not observed in isolation), etc.
- Increased motor activity: tremor, choreoathetosis, hyperactivity, convulsions, etc.
- Alteration of muscle strength: reduced grip strength, increased or decreased muscle tone, muscle fasciculation, weakness, ptosis, inability to stand, paresis, paralysis, etc.
- Coordination: ataxia, abnormal gait, landing foot splay, etc.

In this list of indicators, 'etc.' is to be understood as covering synonyms of the indicators listed, but not as an indication that additional indicators were envisaged. For instance, 'hunched position/posture', 'lateral posture' and 'curved body position' are considered as synonyms of 'recumbency'.

Recumbency was considered as an indicator of functional alteration of the motor division when other indicators were also present, or, when this was not the case, if the AS had a chemical structure known to be capable to induce this specific effect.

Reduced and increased motor activity, although looking at first sight as opposite effects, were considered as two equally valid manifestations of the specific effect. These two types of effects are indeed regularly observed with the same AS.

3.2.2. Functional alteration of the sensory division of the nervous system

The specific indicators of toxicity observable in toxicological studies contributing to the evidence that an AS causes a functional alteration of the sensory division of the nervous system were classified in three categories:

- Decreased reactivity: hyporeactivity, righting reflex (air drop), touch response (handling reactivity), approach response, pupil response, tail pinch response, analgesic reflex (nociception response), patellar reflex, etc.
- Increased reactivity: hyperreactivity, exaggerated auditory response (startle reflex), etc.

- Proprioception and sensory deficits: proprioception deficit, paraesthesia (except for pyrethrins and pyrethroids, where it can be considered as a local effect in studies with administration through the diet), hyperaesthesia, etc.

In this list of indicators, 'etc.' is to be understood as covering synonyms of the indicators given, but not as an indication that additional indicators are envisaged.

3.2.3. Functional alteration of the autonomic division of the nervous system

The specific indicators of toxicity observable in toxicological studies contributing to the evidence that an AS causes a functional alteration of the autonomic division of the nervous system were:

- miosis
- mydriasis
- increased salivation
- lacrimation
- piloerection
- urination

or any synonym of these indicators.

The combination of two or more autonomic signs of toxicity provides a stronger support for a specific effect on the autonomic division. Salivation is considered an indicator of alteration of autonomic division, as a result of systemic exposure, and not a local effect, when other indicators are also present, or, when this is not the case, if the AS has a chemical structure known to be capable to induce this type of neurotoxicity.

Similarly, piloerection is considered as an indicator of functional alteration of the autonomic division when other indicators are also present, or, when this is not the case, if the AS has a chemical structure known to be capable to induce this specific effect.

3.2.4. Brain and/or erythrocyte AChE inhibition

This neurochemical effect is directly defined by its indicator. It was however considered relevant only when the inhibition leads to a statistically significant ($p < 0.05$) decrease of the AChE activity of 20% or more compared to concurrent control groups (JMPR, 1999; US Environmental Protection Agency, 2000).

3.2.5. Neuropathological effects

The specific indicators of toxicity observable in toxicological studies contributing to the evidence that an AS causes histologic neuropathological effects were:

- axonal degeneration (such as sciatic nerve axonopathy)
- myelin degeneration
- neuronal degeneration/necrosis.

It is known that the presence of certain artefacts in microscopic sections of tissues can result in misinterpretations leading to diagnostic pitfalls. For this reason, special care needs to be dedicated to the interpretation of histopathological findings by paying due consideration to observations in control animals and dose–response relationship.

Sciatic nerve axonopathy, without concurrent changes in motor neurons or spinal tracts, may be consistent with an increase of age-related effects due to systemic toxicity and diminished repair capacity of the nerve. Therefore, this indicator is rather considered to be of confirmatory nature when other evidence is available.

3.3. Establishment of CAGs, setting of NOAELs and selection of ICs

3.3.1. General provisions

Establishment of CAGs:

Based on the three data collections (RIVM, ICPS, ANSES, 2013, 2016; EFSA internal data collection) referred to in Section 2.1 and indicators listed in Section 3.2, CAGs were elaborated for the five specific effects of pesticides on the nervous system.

An AS was included in a CAG if it has a known MOA capable to induce directly the specific effect or if at least one of the respective indicators was observed at a statistically significant and/or biologically relevant level in at least one toxicological study with this AS and the study was assessed as 'acceptable' in the DAR, RAR or equivalent document, unless:

- This observation was age related or occurred at or above the MTD, or,
- Consideration of the dose-response relationship showed that the observation was not treatment related.[1]

Studies assessed as 'supportive' or 'unacceptable' in the final DAR or RAR were not considered in any stage of the elaboration and characterisation of the CAGs. Data from reproductive toxicity studies were not considered for the establishment of CAGs since toxicological endpoints were not always clearly reported as pertaining to dams or pups. Teratogenicity studies were also disregarded as they do not adequately examine toxic effects on the nervous system.

When a metabolite or degradation product present in food had been investigated by regulatory studies and found to meet the above conditions, it was also included in the CAG.

With respect to the functional alterations of the motor, sensory and autonomic divisions, it was considered appropriate to include ASs in CAGs on the sole basis of a known relevant MoA, even in the absence of factual observation of indicators of a specific effect in the available toxicological studies. This is because, if a pesticide affects a target site which is critical to nervous system function, then it can be expected that effects may occur in all three functional divisions due to the interdependencies discussed in Section 3.1. This is supported by the available evidence for most of the known neurotoxic MoA listed in Section 3.3.2. Appendix C shows the observed indicators of functional alterations of the three divisions of the nervous system for *N*-methyl carbamate insecticides, macrocyclic lactone insecticides, neonicotinoid insecticides, organophosphorus pesticides, organochlorine insecticides, phenylpyrazole insecticides, pyrethrin and pyrethroids ester insecticides, chlormequat, mepiquat, sulfoxaflor, indoxacarb and amitraz. Except for amitraz for which effects on the autonomic function are not observed, effects are always seen at varying intensities in the motor, sensory and autonomic divisions of the nervous system for all these chemical classes. Therefore, while assuming functional effect on the sole basis of a demonstrated MoA is a reasonable default approach, it may overestimate the risk in a few cases.

Setting of NOAELs:

The data collection spreadsheets mentioned in Section 2.1 were used to characterise each AS included in a CAG for the respective specific effect. NOAELs for short- and long-term cumulative exposure/risk assessments were derived for each AS from the most sensitive indicator, using all available information across studies, species and sexes.

All indicators listed in Section 3.2 were equally valid for the setting of NOAELs for specific effects.

In case two or more studies of similar design within the same species investigated the same indicators of a specific effect, they were combined where relevant to derive the respective NOAEL based on the whole information.

In case only a LOAEL was available for a certain indicator, a default NOAEL was determined from this LOAEL by applying an additional uncertainty factor (UF), as recommended by the guidance of EFSA on default values to be used in the absence of measured data (EFSA Scientific Committee, 2012). In the present report, the value of this additional UF was however not defined on a case-by-case basis, but, instead, was set at 10 in all cases.

Human studies reported in the spreadsheets were never used for the establishment of CAGs, as the provisions of Commission Regulation (EU) No 283/2013 authorising their use (scientific validity, ethical generation and leading to lower regulatory limit values compared to animal studies) were never met.

For ASs included in the CAGs related to AChE inhibition and functional alterations of the motor, sensory and autonomic divisions, both acute and chronic NOAELs were established, as these effects can be triggered by both acute and chronic exposures.

In order to establish NOAELs for ASs with a known MoA regarding functional alterations of the motor, sensory and autonomic divisions, preference was given to neurotoxicity studies, unless dog or mouse was more sensitive than rats for these effects and studies in these species resulted in lower

[1] In other words, dose–response relationship was established when the effect was observed in at least two dose levels (not only at the top dose) and the comparison of the responses at the different dose levels did not exclude their relationship with the treatment.

NOAELs. When indicators of functional alteration of the nervous system were not observed at statistically significant level in neurotoxicity studies, the highest tested dose in these studies was defined as the NOAEL for the respective division. In the absence of acute neurotoxicity studies, available 28-day or 90-day neurotoxicity studies were used to set NOAELs for acute exposure/risk assessments. In the absence of 28-day or 90-day neurotoxicity studies, several options were considered to set NOAELs for chronic exposure/risk assessment, including the use of acute neurotoxicity studies (*N*-methyl carbamates), the use of other regulatory studies, the use of the NOAELs for AChE inhibition or the use of the NOAEL leading to the ADI. Similar options were considered to set NOAELs for acute exposure/risk assessment when neurotoxicity studies were totally missing. The impact of missing neurotoxicity studies will be addressed in the context of the overall uncertainty analysis (Second bullet point of Section 4.5 (overall uncertainty analysis)).

EFSA conclusions on the pesticide risk assessment in the context of Regulation (EC) No 1107/2009 finalised until end 2018 and dealing with ASs included in the CAGs were considered to retrieve any element of expert judgement regarding their effects on the nervous system and to ensure consistency of the NOAELs proposed for the specific effect with the ADI and ARfD of the AS at the time of the preparation of the present report. For ASs not reviewed by EFSA, the scientific evaluations conducted by the body constituting the main source of the data collection were also considered (e.g. JMPR evaluations).

Selection of Index Compounds:

To enable risk assessors to perform cumulative exposure/risk assessments using an IC and RPFs, an IC was proposed from the ASs included in the CAG. The IC was preferably selected between ASs of high potency and with highly convincing evidence that it causes the specific effect using the following criteria:

- Quality of the study (study meeting the requirements of regulation (EC) No 1107/2009, considered acceptable, statistical robustness of the findings)
- Strength of the specific effect (NOAEL, number of indicators of the specific effect observed)
- Evidence of dose–response relationship
- Consistency in the occurrence of the specific effect across genders, species and studies

Two ICs were selected when a CAG is to be used for both acute and chronic exposure/risk assessments. This is the case for the CAGs related to the brain and/or erythrocyte AChE inhibition and to the functional alterations of the motor, sensory and autonomic divisions of the nervous system.

In subsequent CRAs using ICs, RPFs will need to be calculated to normalise the toxicity of all ASs in each CAG to the IC, by dividing the NOAEL of the IC by the NOAEL of the AS.

3.3.2. Chemical classes with known or presumed neurotoxic MoA

Based on knowledge of toxicological properties of pesticides and scientific peer-reviewed open literature, the following chemical classes and ASs were considered as having a known neurotoxic MoA in mammals:

a) *N*-methyl carbamate insecticides (reversible AChE inhibitors):

- Benzofuranyl methylcarbamate: benfuracarb, carbofuran, carbosulfan;
- Carbamate: carbaryl;
- Dimethyl carbamate: pirimicarb;
- Formamidine: formetanate (also agonist of the octopamine receptor in insects which is equivalent to the alpha-2 adrenergic receptor in mammals);
- Oxime carbamate: aldicarb, methomyl, oxamyl, thiodicarb;
- Phenyl methylcarbamate: methiocarb.

b) Macrocyclic lactone insecticides (GABA-gated chloride channel agonist):

- Avermectins: abamectin, emamectin benzoate;
- Milbemycin: milbemectin.

c) Neonicotinoid insecticides (agonist of nicotinic acetylcholine receptor (nAChR)): acetamiprid, clothianidin, dinotefuran, imidacloprid, thiacloprid, thiamethoxam.

d) Organophosphorus pesticides (irreversible AChE inhibitors):

- Organophosphates: chlorfenvinphos, dichlorvos, monocrotophos;

- Organothiophosphates: azinphos-ethyl, azinphos-methyl, cadusafos, chlorpyrifos, chlorpyrifos-methyl, diazinon, dimethoate, ethion, ethoprophos, fenitrothion, fenthion, fosthiazate, malathion, methidathion, omethoate, oxydemeton-methyl, parathion, parathion-methyl, phenthoate, phosalone, phosmet, phoxim, pirimiphos-methyl, profenofos, pyrazophos, triazophos;
- Phosphonates: trichlorfon;
- Phosphonothioates: fonofos;
- Phosphoramidates: fenamiphos;
- Phosphoramidothioates: acephate, methamidophos;
- Phosphonic acids: ethephon (growth regulator);
- Although considered as a weak AChE inhibitor, the fungicide tolclofos-methyl has also a typical organophosphorus structure and is metabolically activated to the oxon moiety.

e) Organochlorine insecticides (GABA-gated chloride channel blockers):

- Cycloalkanes: lindane;
- Cyclodienes: dieldrin, endrin, endosulfan (allocated to subgroup 5 in the EKE process), heptachlor.

f) Phenylpyrazole insecticides (GABA-gated chloride channel blockers): fipronil.

g) Pyrethrins and pyrethroid ester insecticides (bind to the voltage-gated sodium channel (VGSC) preventing its transition from an activated (ion-conducting) to an inactivated (non-conducting) state): acrinathrin, alpha-cypermethrin, beta-cyfluthrin, beta-cypermethrin, bifenthrin, cyfluthrin, cypermethrin, deltamethrin, esfenvalerate, fenpropathrin, fenvalerate, lambda-cyhalothrin, permethrin, tau-fluvalinate, tefluthrin, tetramethrin, zeta-cypermethrin.

h) Chlormequat and mepiquat are alkyl quaternary ammonium plant growth regulators. They are partial agonists of the nAChR. Chlormequat chloride showed *in vitro* a weak agonistic activity on muscarinic receptors, with potency 10^5 lower than for atropine, and also showed to be a partial agonist of the nAChR, with a potency of about 1% of that of acetylcholine (EFSA, 2009).

i) Sulfoxaflor is a sulfoximine insecticide. Toxicity and mechanistic studies in rats, rabbits, dogs and mice indicate that sulfoxaflor is an activator of the mammalian nAChR, but to a much lesser degree than in insects and in a species-specific manner.[2]

j) Indoxacarb belongs to a relatively new class of sodium channel blocker insecticides (SCBIs) with a MoA distinct from all other sodium channel-targeting insecticides, including pyrethroids, but similar to that of cationic local anaesthetics (Zhang et al., 2016).

k) Amitraz: formamidine acaricide, octopamine receptor agonist in insects which is equivalent to alpha-2 adrenergic receptor in mammals. Several effects of amitraz in mammals are mediated by its interaction with alpha-2 adrenoceptors (Costa et al., 1989).

All ASs falling under items a) to k) above have MoAs directly relevant for the functional alterations of the motor, sensory and autonomic divisions of the nervous system and are included in the respective CAGs (see Section 3.1). With respect to brain and/or erythrocyte AChE inhibition, only groups a) and d) are relevant.

Less convincing (e.g. being scarce or lack of consistency) mechanistic evidence is available for the following ASs, which were considered as having a presumed mode of neurotoxic action in mammals:

l) Metaldehyde is a molluscicide. It leads to an increase in monoamine oxidase (MAO) activity and to a decrease in GABA, norepinephrine (NE) and 5-hydroxytryptamine (5-HT) concentrations which leads to neuronal excitation (GABA) and decreased seizure threshold (NE, 5-HT), both of which can result in convulsions (Yas-Natan et al., 2007).

m) Triadimefon and triadimenol are triazole fungicides. Triadimefon and triadimenol are inhibitors of dopamine transporter, leading to increased synaptic concentrations of dopamine (Walker and Mailman, 1996).

n) Dithiocarbamates (mancozeb, maneb, metiram, propineb, thiram and ziram): although the molecular mechanisms underlying the neurotoxicity of dithiocarbamate fungicides in general are not well understood, the generation of CS_2 *in vivo* has been shown to cause Lys-Lys thiourea cross-linking of neurofilament proteins resulting in the accumulation of neurofilaments in the axon, which causes the characteristic lesion of axonal swelling (Valentine et al., 1997). This is

[2] https://toxnet.nlm.nih.gov/cgi-bin/sis/search/a?dbs+hsdb:@term+@DOCNO+8245

presumed to cause peripheral neuropathy and subsequent motor and some sensory symptoms under chronic exposure.

o) Molinate is a thiocarbamate herbicide. The neurotoxic effects observed may be a consequence of its metabolite molinate sulfone, which inhibits aldehyde dehydrogenase (ALDH) by covalently binding to the active-site Cys residue. Inhibition of ALDH, particularly the ALDH2 isoenzyme, in the brain results in accumulation of endogenous neurotoxins, such as 3,4-dihydroxyphenylacetaldehyde (DOPAL), a dopamine metabolite, or 4-hydroxy-2-nonenal (4HNE), a product of lipid peroxidation, which may account for the observed neurotoxicity (Allen et al., 2010). As ALDH2 is widely expressed in the frontal and temporal cortex, hippocampus, mid-brain, basal ganglia and cerebellum, it plays a crucial function in protecting neurons in the brain and the spinal cord (Chen et al., 2016).

p) Pymetrozine, a pyridine azomethine insecticide, acts by overstimulating and eventually silencing vanilloid-type transient receptor potential (TRPV) channels in insect chordotonal stretch receptor neurons. However, there is no indication of cross-reactivity of TRPV channels in mammals. In this regard, afidopyropen, an insecticide with a similar MoA than pymetrozine that stimulated TRPV channels from two different insect species, did not affect the function of the mammalian TRPV channel TRPV4 (Kandasamy et al., 2017).

q) Dicofol is believed to act by at least four mechanisms, possibly all functioning simultaneously. It may reduce potassium transport across the membrane. Dicofol also alters the porous channels through which sodium ions pass. These channels activate (open) normally but are inactivated (closed) slowly, thus interfering with the active transport of sodium out of the nerve axon during repolarisation. Dicofol inhibits neuronal adenosine triphosphatases (ATPases), particularly Na^+ K^+-ATPase, and Ca^{2+}-ATPase which play vital roles in neuronal repolarisation. Dicofol also inhibits the ability of calmodulin, a calcium mediator in nerves, to transport calcium ions that are essential for the release of neurotransmitters.[3]

These ASs (items l to q) are included in CAGs only in case of observation of relevant indicators in toxicological studies.

This is also the case for the following ASs for which no information is available on the mode of neurotoxic action:

2,4-D (phenoxyacetic herbicide), bromide ion (comes from methyl bromide, fumigant), carbetamide (carbanilate herbicide), chlorpropham (carbanilate herbicide, growth regulator), desmedipham (carbanilate herbicide), dicamba (benzoic acid herbicide), dicofol (bridge diphenyl acaricide), ethephon (growth regulator), fenpropidin (unclassified fungicide), fenpropimorph (morpholine fungicide), flufenacet (anilide herbicide), fluquinconazole, tebuconazole and tetraconazole (triazole fungicides), glufosinate (organophosphorus herbicide), isoxaflutole (oxazole herbicides), lufenuron (benzoylphenylurea), metribuzin (triazinone herbicide), triallate (thiocarbamate herbicide), oxasulfuron (sulfonylurea), penflufen (anilide herbicide), pyridate (pyridazine herbicide), spirotetramat (tetramic acid insecticide), tembotrione (benzoylcyclohexanedione herbicides), thiophanate-methyl (benzimidazole, carbamate fungicide).

In this section, only the most well-established neurotoxic MoAs were described. However, other MoAs could occur simultaneously for example in the case of organochlorine insecticides (e.g. endosulfan and lindane) acting mainly as GABA-A receptor antagonists, but also inhibiting Ca- and Mg-ATPase (Jayaraj et al., 2016). Hence, there is some uncertainty regarding allocating specific pesticides to CAGs according to pesticide classes.

3.3.3. Cumulative assessment groups

This section presents the CAGs proposed to be used for future CRAs. They differ to some extent from those initially elaborated by the PPR Panel and published in the Scientific Opinion of 2013 (EFSA PPR Panel, 2013a), because only one (RIVM, ICPS, ANSES, 2013) of the three data collections used in this report was available to the Panel when it adopted its opinion and comments submitted during the public consultation on a draft of the present report were considered (EFSA, 2019c).

Functional alteration of the motor, sensory and autonomic divisions:

In total, 85 ASs have a known MoA of direct relevance for functional alterations of the motor, sensory and autonomic divisions and are included in the three CAGs: abamectin, acephate, acetamiprid, acrinathrin, aldicarb, alpha-cypermethrin, amitraz, azinphos-ethyl, azinphos-methyl, benfuracarb, beta-

[3] http://www.t3db.ca/toxins/T3D0144

cyfluthrin, beta-cypermethrin, bifenthrin, cadusaphos, carbaryl, carbofuran, carbosulfan, chlorfenvinphos, chlormequat, chlorpyriphos, chlorpyriphos-methyl, clothianidin, cyfluthrin, cypermethrin, deltamehrin, diazinon, dichlorvos, dieldrin, dimethoate, dinotefuran, emamectin, endosulfan, endrin, esfenvalerate, ethephon, ethion, ethoprophos, fenamiphos, fenitrothion, fenpropathrin, fenthion, fenvalerate, fipronil, fonofos, formetanate, fosthiazate, heptachlor, imidacloprid, indoxacarb, lambda-cyhalothrin, lindane, malathion, mepiquat, metamidophos, methidathion, methiocarb, methomyl, milbemectin, monocrothophos, omethoate, oxamyl, oxydemeton-methyl, parathion, parathion-methyl, permethrin, phenthoate, phosalone, phosmet, phoxim, pirimicarb, pirimiphos-methyl, profenofos, pyrazophos, pyrethrins, sulfoxaflor, tau-fluvalinate, tefluthrin, tetramethrin, thiacloprid, thiamethoxam, thiodicarb, tolclofos-methyl, triazophos, trichlorfon, zeta-cypermethrin.

The chemical classes of these ASs and their MoAs are described in Section 3.3.2, points a) to k).

Based on factual observations of relevant indicators, additional ASs were included in these CAGs as follows:

- CAG on the functional alteration of the motor division (Tables B.1 and B.2): 2,4-D, bromide ion, carbetamide, chlorpropham, desmedipham, dicamba, dicofol, fenpropidin, fenpropimorph, flufenacet, fluquinconazole, glufosinate, isoxaflutole, lufenuron, mancozeb, maneb, metaldehyde, metiram, molinate, oxasulfuron, penflufen, propineb, pymetrozine, pyridate, spirotetramat, tebuconazole, tembotrione, tetraconazole, thiophanate-methyl, thiram, tri-allate, triadimefon, triadimenol, ziram. These additional 34 ASs lead to a total of 119 ASs included in this CAG.
- CAG on the functional alteration of the sensory division (Tables B.2 and B.6): cymoxanil, dicamba, dicofol, fenpropimorph, flufenacet, glufosinate, halosulfuron-methyl, metaldehyde, molinate, oxasulfuron, propineb, sulcotrione, tebuconazole, tembotrione, thiram, tri-allate. These additional 16 ASs lead to a total of 101 ASs included in this CAG.
- CAG on the functional alteration of the autonomic division (Tables B.3 and B.7): 2,4-D, carbetamide, chlorpropham, dicamba, dicofol, flufenacet, fluquinconazole, glufosinate, metaldehyde, metamitron, molinate, pyridate, tebuconazole, thiram, tri-allate, triadimenol. These additional 16 ASs lead to a total of 101 ASs included in this CAG.

In the CAG on the functional alteration of the motor division, the ICs for the acute and chronic risk assessments are proposed as oxamyl and emamectin benzoate, respectively. Oxamyl was selected based on the robustness of data and the number of endpoints affected in the acute neurotoxicity study. Emamectin benzoate was selected based on the robustness of data and consistency of effects across studies and animal species (rat, dog and mouse).

In the CAG on the functional alteration of the sensory division, the ICs for the acute and chronic risk assessments are proposed as oxamyl and endrin, respectively. Oxamyl was selected on the basis of the robustness of findings (dose relationship in mid and high dose males and statistical significance). Endrin was selected based on the consistency of findings observed in two strains of rats.

In the CAG on the functional alteration of the autonomic division, the ICs for the acute and chronic risk assessments are proposed as oxamyl and methamidophos, respectively. Both were selected based on the robustness of data.

Furthermore, all proposed ICs have a known MoA.

In the NOAEL-setting process, it was noted that for 19 ASs belonging to chemical classes with a known MoA (amitraz, azinphos-ethyl, chlorfenvinphos, chlormequat, dichlorvos, dieldrin, endrin, ethion, fosthiazate, methiocarb, monocrotophos, oxydemeton-methyl, parathion, parathion-methyl, phenthoate, phoxim, pyrazophos, tetramethrin and triazophos) neurotoxicity studies are not available. For an additional set of 14 ASs (carbetamide, chlorpropham, desmedipham, fenpropidin, fluquinconazole, isoxaflutole, mancozeb, maneb, metiram, oxasulfuron, propineb, pyridate, tetraconazole, thiophanate-methyl), functional alterations of the motor division were observed in single and/or repeated dose studies, but specific neurotoxicity studies were not available. This lack of neurotoxicity studies is a source of uncertainty affecting the setting of NOAELs for functional alterations of the motor, sensory and autonomic divisions. This will need to be considered in the interpretation of the cumulative risk assessments when they will be performed.

For 50 ASs belonging to chemical classes with a known MoA, the acute NOAELs characterising their effects on the three functional divisions of the nervous system were derived from the same neurotoxicity study. The comparison of the NOAELs for the three divisions indicates that the motor division is the most sensitive to the effects of chemicals with neurotoxic MoA. Indeed:

- The comparison between the NOAELs of individual ASs in the motor and sensory divisions showed that the NOAELs are lower in the motor division in 28 cases and lower in the sensory division in two cases. In 20 cases, the NOAELs were the same.
- The comparison between the NOAELs of individual ASs in the motor and autonomic divisions showed that the NOAELs are lower in the motor division in 23 cases and lower in the autonomic division in three cases. In 24 cases, the NOAELs were the same.

In view of this comparison, it is concluded that the cumulative risk assessments for the effects of pesticides on the motor division might be enough to cover the cumulative risks on all functional divisions.

Brain and/or erythrocyte AChE inhibition:

This effect is defined at biochemical level and is always associated with organophosphorous and *N*-methyl carbamate insecticides, and only ASs of these chemical classes of pesticides are included in the CAG. Since erythrocyte AChE is a membrane bound enzyme, oxidative stress induced by other chemical classes may also lead indirectly to a decrease in erythrocyte AChE activity (Banerjee et al., 1999; El-Demerdash, 2011). ASs acting via this indirect pathway are however not included in the CAG as they do not meet the precise definition of the specific effect.

In total, 47 ASs are included in the CAG for the brain and/or erythrocyte AChE inhibition (Tables B.4 and B.8). These ASs are: acephate, aldicarb, azinphos-ethyl, azinphos-methyl, benfuracarb, cadusaphos, carbaryl, carbofuran, carbosulfan, chlorfenvinphos, chlorpyriphos, chlorpyriphos-methyl, diazinon, dichlorvos, dimethoate, ethephon, ethion, ethoprophos, fenamiphos, fenitrothion, fenthion, fonofos, formetanate, fosthiazate, malathion, methamidophos, methidathion, methiocarb, methomyl, monocrotophos, omethoate, oxamyl, oxydemeton-methyl, parathion, parathion-methyl, phenthoate, phosalone, phosmet, phoxim, pirimicarb, pirimiphos-methyl, profenofos, pyrazophos, thiodicarb, toloclophos-methyl, triazophos, trichlorfon.

The ICs for the acute and chronic risk assessments are proposed as oxamyl and omethoate, respectively. Oxamyl was selected based on the robustness of data. Omethoate was selected based on the consistency of findings across studies and animal species (rat, dog, mouse and rabbit).

Generally, the NOAELs related to AChE inhibition are notably lower than the NOAELs of the respective ASs for their effects on the motor, sensory and autonomic divisions of the nervous system as well as for their neuropathological effects.

Neuropathological effects:

With respect to this specific effect, no chemical class or MoA linking a biochemical mechanism and the observed histopathological changes has been identified. For a few ASs, only presumed MoAs have been proposed. Therefore, ASs are included in this CAG based on factual observations only.

In total, 19 ASs are included in the CAG for neuropathological effects (Table B.9). These ASs are: chlorfenapyr, cymoxanil, cypermethrin, emamectin benzoate, fenpropidin, flufenacet, indoxacarb, isoxaflutole, lindane, mancozeb, molinate, oxasulfuron, quinoclamine, tau-fluvalinate, tembotrione, thiram, tri-allate, trichorfon, ziram.

The IC is proposed as emamectine, which was selected based on the consistency of findings across studies and animal species (rat, dog and mouse).

A comparison between this CAG and the CAG for functional alterations of the motor division showed that:

- The CAG for neuropathological effects contains about 6 times less ASs than the CAG for the motor division. This is explained by the fact that functional and biochemical mechanisms, not necessarily associated to neuropathological effects, are major causal factors of alteration of the motor function.
- Three out of the 19 ASs included in the CAG for neuropathological effects are not in the CAG for the motor division. These ASs are chlorfenapyr, cymoxanil and quinoclamine.
- The number of ASs in the CAG for motor division with NOAELs for chronic effects exceeds by about 60 the number of ASs in the CAG for neuropathological effects. Four ASs (chlorfenapyr, cymoxanil, quinoclamine and ziram) have NOAELs for neuropathological effects, but not NOAEL for chronic effects in the motor division.
- From the ASs included in the CAG for neuropathological effects and having a NOAEL for chronic effects in the motor division, five have lower NOAELs for neuropathological effects (lindane, mancozeb, thiram, tri-allate and trichlorfon), three have lower NOAELs in the motor division and seven have the same NOAELs for both effects.

In view of this comparison, it is anticipated that the cumulative risks for the neuropathological effects will be covered by cumulative risk assessments for the effects on the motor division and therefore may not require separate assessment.

Nine tables (see Appendix B) were prepared to support all possible acute and chronic exposure/risk assessments that could be conducted using the five CAGs. For each AS included in the CAG, these tables indicate which indicator of the specific effect is used for hazard characterisation, the respective reference point (NOAEL/LOAEL) and the reference of the study from which this reference point was retrieved. They also mention the source of the information (e.g. DAR, 2011; JMPR, 1997), the EFSA conclusions considered (e.g. EFSA (2008)), some additional elements of the assessment and the available information on MoAs. In accordance with Article 63 of Regulation (EC) No 1107/2009, the names of persons involved in these studies are confidential and are not shown in the study reference details.

3.3.4. Use of the CAGs to assess consumer safety

As indicated in Section 3.3.3, it is expected that CRA conducted with the CAG for the effects on the motor division will show higher risks than those carried out with the CAGs for the effects on the sensory and autonomic divisions and for the neuropathological effects. Therefore, in order to assess the combined effects of pesticide residues present in consumer diet on the nervous system, it would be enough to perform CRA with the CAGs for AChE inhibition and for the functional alterations of the motor division, if similar protection goals would apply to all these effects.

In conducting these CRA, the potential contribution of metabolites and degradation products to the specific effects should be taken into account. It should be considered whether the residue definition for risk assessment established with respect to the critical effect(s) (e.g. effects on which the ADI and/or ARfD are based) can be used. If this is not appropriate, another residue definition should be considered on a case-by-case basis which is consistent with the specific effect. In doing so, it is recommended to use the guidance of the PPR Panel on the establishment of the residue definition for dietary risk assessment (EFSA PPR Panel, 2016).

4. Uncertainty analysis

4.1. General considerations

The actual and first-hand information supporting the establishment of CAGs lies in the original studies submitted by the applicants for approval of ASs. For reasons of resources, these studies have only occasionally been consulted for the purpose of the present exercise. Instead, regulatory documents, where information from the original studies is reported in a condensed form have been used as the primary source of information.

Information of relevance for the establishment of CAGs might not have been captured properly when these regulatory documents were drafted, as their main purpose is to establish the reference values of the ASs. This constitutes a general source of uncertainty which may result in some underestimation of the actual risk, because the most common issue with these regulatory documents is likely to be the omission to report effects at doses exceeding the overall NOAEL of the respective study.

In addition, for several ASs, especially for ASs which are not approved anymore in EU, the quality of the database does not conform to the current standards and causes an additional source of uncertainty. This also leads to some possible over- or underestimation of the contribution of the respective ASs to the actual cumulative risk.

A particular source of uncertainties with respect to the effects of pesticides on the nervous system stems from the fact that a neurotoxicity study is not always available, despite the fact that Commission Regulation (EU) No 283/2013 setting out the data requirements for ASs provides that such study should be performed for ASs with structures that are similar or close to those capable of inducing neurotoxicity. This absence of a neurotoxicity study may result in overestimated NOAELs for some ASs (and thus underestimating the actual risk) as information on some indicators is missing in this case.

Specific sources of uncertainties related to the CAGs for functional alterations of the motor division, brain and/or erythrocyte AChE inhibition and other specific effects are addressed in Sections 4.2–4.4. In Section 4.5, recommendations are given about the overall sources of uncertainties to be systematically reviewed when CRAs are conducted with the CAGs established in the present report.

4.2. CAG for the functional alterations of the motor division

4.2.1. Question 1: Does the CAG for the functional alteration of the motor division contain all ASs contributing to this effect and only ASs causing this effect?

Question 1 implies providing responses to two discrete questions.

The first one is *whether all ASs causing the effect are well included* in the CAG. To evaluate the chance of omitting ASs contributing to the effect, the reader should refer to the first bullet point of Section 4.5 (overall uncertainty analysis).

The second one is *whether ASs not causing the effect are included* in the CAG. For the CAG on motor division, the possibility of including ASs not contributing to the effect has been addressed by Weight of Evidence and EKE techniques described in Section 2.2.5. The process was conducted as follows:

a) A key step in EKE is specification of the question to be addressed in a well-defined manner and, if possible, in such a way that the answer to the question is potentially observable, at least in principle (EFSA, 2014). The question of interest for CRA is, for each AS, 'Does this chemical cause any functional alteration of the motor division of the nervous system (locomotor activity, muscular strength and coordination)?' In regulatory practice, causation of toxic effects is determined by established standard procedures for the conduct, reporting and interpretation of toxicity studies. The elicitation question was therefore defined as follows: 'If the required set of studies (including neurotoxicity studies if relevant) was performed and reported perfectly, and the results were analysed and interpreted according to the standard procedure, would this chemical be assessed as positive for effects on the motor division of the nervous system?' For the purpose of risk assessment, these two framings of the question are equivalent.

b) The lines of evidence and their respective weights are:

- Belonging of the AS to one of the following chemical classes: organophosphates, *N*-methyl carbamates, organochlorines, macrocyclic lactones, pyrethroids, neonicotinoids or phenylpyrazoles: 32,000. This exaggerated weight was applied in order to ensure that ASs with and without relevant chemical structures are separated completely in the ranking process, and placed in different subgroups, to increase the homogeneity of evidence in each subgroup and make it easier for experts to judge the probability of CAG membership.
- AS for which a MoA is presumed (rather than known) in the data collection tables: 3.
- Observation of decreased motor activity[4]: 4 if hypoactivity is reported; 2 if any indicator of reduced activity, but not hypoactivity, is reported. From the indicators of reduced motor activity, hypoactivity is the one which is the most closely related to the adverse effect of a chemical. The other indicators must be considered more carefully and are rather considered as confirmatory information supporting other observations.
- Observation of increased motor activity[3]: 3 if only one indicator is reported; 4 if more than one indicator is reported.
- Observation of effects on the muscular strength: 3 if only one indicator is reported or in the absence of any neurotoxicity study[5]; 4 if more than one indicator is reported.
- Observation of effects on the motor coordination: 3 if only one indicator is reported or in the absence of any neurotoxicity study[4]; 4 if more than one indicator is reported.
- Concomitant observation of indicators of functional alteration of the sensory function: 3 in the absence of any neurotoxicity study[4]; 4 if any indicator is reported.
- Observation of indicators in more than one species: 3.
- Observation of a dose-response relationship for the most sensitive indicator: 4.

c) Based on the lines of evidence, scores were calculated for all ASs of the CAG. These scores were used to distribute the ASs into six subgroups of decreasing scores. The first four subgroups contained ASs from chemical classes associated to neurotoxicity. The next two subgroups contained ASs of different chemical classes. The 7 ASs identified from the CAG on

[4] In case of observations of decreased and increased activity for a same AS, the combined weight is however limited to an upper limit of 8.

[5] Neurotoxicity studies are essential to observe indicators related to motor coordination, muscular strength and sensory function. In the absence of such studies for certain ASs, it was conservatively assumed that these ASs would show these indicators if these studies were conducted. These cases were highlighted in the tabulated evidence and the effect of this assumption was considered by the experts when making their judgements.

neurochemical effects were not scored and treated separately as a seventh subgroup of ASs. The compositions of these seven subgroups are as follows:

- Subgroup 1 (18 ASs): acetamiprid, aldicarb, carbofuran, clothianidin, cypermethrin, deltamehrin, emamectin, ethoprophos, fenpropathrin, fenvalerate, fipronil, formetanate, metamidophos, milbemectin, omethoate, oxydemeton-methyl, pyrethrins and thiamethoxam.
- Subgroup 2 (20 ASs): abamectin, acrinathrin, azinphos-methyl, beta-cyfluthrin, beta-cypermethrin, bifenthrin, cadusaphos, carbaryl, dichlorvos, esfenvalerate, fenitrothion, fosthiazate, imidacloprid, oxamyl, permethrin, tau-fluvalinate, tefluthrin, thiacloprid, trichlorfon and zeta-cypermethrin.
- Subgroup 3 (14 ASs): alpha-cypermethrin, benfuracarb, cyfluthrin, diazinon, dimethoate, fenamiphos, fenthion, lambda-cyhalothrin, lindane, methidathion, monocrothophos, pirimicarb, pyrazophos and thiodicarb.
- Subgroup 4 (20 ASs): acephate, carbosulfan, chlorpyrifos, chlorpyrifos-methyl, dieldrin, dinotefuran, endrin, fonofos, heptachlor, malathion, methiocarb, methomyl, parathion, parathion-methyl, phosalone, phosmet, phoxim, pirimiphos-methyl, tetramethrin and triazophos.
- Subgroup 5 (19 ASs): bromide ion, carbetamide, chlormequat, dicamba, dicofol, endosulfan, flufenacet, fluquinconazole, indoxacarb, mepiquat, metaldehyde, metribuzin, molinate, oxasulfuron, pyridate, sulfoxaflor, tebuconazole, thiram and triallate.
- Subgroup 6 (22 ASs): 2,4-D, amitraz, chlorpropham, desmedipham, fenpropidin, fenpropimorph, glufosinate, isoxaflutole, lufenuron, mancozeb, maneb, metiram, penflufen, propineb, pymetrozine, tembotrione, tetraconazole, thiophanate-methyl, triadimefon, triadimenol, spirotetramat and ziram.
- Subgroup 7 (7 ASs): azinphos-ethyl, chlorfenvinphos, ethephon, ethion, phenthoate, profenofos and tolclofos-methyl.

d) The probability estimations for subgroups 1 to 7 concluded that:

In subgroup 1 (18 ASs):

- All ASs in this group have relevant structure and mechanism and show clear evidence of dose-response relationship and effects on two or more species.
- All show both reduced and increased motor activity.
- All showed effect on coordination, muscle strength, and on the sensory division of the nervous system, except one AS for which no neurotoxicity study was available, but in this case muscle strength effects were seen in another study.
- Overall, it was judged almost certain (99–100%) that all substances cause effects on the motor division.

In subgroup 2 (20 ASs):

- All ASs have relevant structure or mechanism.
- All ASs show dose-response relationship.
- All ASs show several indicators of effects on the motor division.
- For each ASs, one of the lines of evidence was missing (missing indicators, missing observations in the sensory division or missing observation in a second species).
- Overall, it was judged almost certain (99–100%) that all 20 substances cause motor division effect.

In subgroup 3 (14 ASs):

- All ASs have relevant structure and mechanism and substantial evidence of effects (indicators).
- For four ASs, the available data do not show a dose response, but this is either mitigated by the observations available for another mixture of the same isomers (alpha-cypermethrin), a metabolite (dimethoate) and/or possibly due to the limitations of the studies (e.g. no neurotoxicity study available for monocrotophos). It is also unlikely that the effects on the top dose were observed on doses above the MTD as such cases would have been excluded by the data collection process.
- Overall, it was judged almost certain (99–100%) that all ASs cause effects on the motor division of the nervous system.

In subgroup 4 (20 ASs):

- All ASs have relevant structure or mechanism.
- All ASs have few positive indicators of effects on the motor division.
- However, data available for most substances is thought to be old and less complete/robust. For six ASs, a neurotoxicity study is not available.
- For some substances there is positive evidence from human/medical data of the effects on the motor function, which was not captured by the data collection.
- Overall, it was judged almost certain (99–100%) that all 20 substances cause motor division effect.

In subgroup 5 (19 ASs):

- Only two ASs have known structural alerts of neurotoxicity.
- Five ASs (chlormequat, endosulfan, indoxacarb, mepiquat and sulfoxaflor) have a known neurotoxic MoA. In addition, dicofol, being structurally related to DDT is believed to act by at least four mechanisms. Three ASs (oxasulfuron, thiram and triallate) cause neuropathology.
- All ASs have several types of motor division effects, all show a dose–response, and all but three have effects in two or more species.
- Sixteen ASs have effects on the sensory division.
- 5–6 ASs were judged extremely likely (95–99%) to cause motor division effects, 1–2 ASs were judged unlikely (10–33%), 1–2 as likely as not (33–66%) and most ASs likely (66–90%) or very likely (90–95%).
- Overall, it was judged that the number of ASs causing motor division effects is likely (66–90%) to be between 15 and 17, with a plausible range from 12 to 19.

In subgroup 6 (22 ASs):

- Six ASs have known or presumed MoA (amitraz, mancozeb, maneb, metiram, propineb and ziram).
- For all AS show at least one indicator, and many two or more.
- Neurotoxicity studies were available for 12 of the 22 ASs, detecting motor division effects in most but not all cases.
- A dose–response was reported for nine ASs only.
- In seven cases, effects were seen in two or more species.
- Five ASs were judged extremely likely (95–99%) to cause motor division effects; four were judged likely (66–90%); five as likely as not (33–66%); two unlikely (10–33%); three very unlikely (5–10%) and three extremely unlikely (1–5%).
- Overall, it was judged that the number of ASs that cause motor division effects is likely (60–90%) to be between 10 and 12, with plausible range between 8 and 15.

In subgroup 7 (7 ASs):

- Five out of the seven ASs have clearly relevant structure/mechanism, for the other two this is questionable.
- Four of the seven ASs inhibit brain AChE, one (ethephon) inhibits erythrocytes AChE but not brain, and for two ASs information on brain AChE inhibition is missing.
- For three ASs, a neurotoxicity study is available. None of these studies show effects on the motor division, but two of these studies are old and not likely to detect such effects.
- The group judged that it is almost certain (99–100%) that chlorfenvinfos, ethion, profenofos, azinphos-ethyl and penthoate cause effects on the motor division, but that it is extremely unlikely (1–5%) that either ethephon or tolclofos-methyl cause the effects.
- Overall it was judged extremely likely (95–99%) that five ASs cause motor division effects extremely unlikely (1–5%) that 6 cause the effects.

The probability terms used in the above considerations are recommended in the EFSA guidance on communication of uncertainty (EFSA, 2019d).

Appendix D provides details on the evidence collected for each AS, its score and the subgroup it belongs to. After the EKE session, the available technical information was further reviewed and this resulted in the identification of a few inaccuracies in the information reported in Appendix D, e.g. regarding the availability of neurotoxicity studies. However, these inaccuracies are impactless on the result of the EKE session.

e) Assessing the total number of ASs causing motor division effects:

- It is nearly certain that all the ASs of subgroups 1, 2, 3 and 4 cause alterations on the motor function. The same is true for five of the seven substances in subgroup 7.
- The number of ASs of subgroup 5 causing alterations on the motor function is most likely between 15 and 17, with plausible range from 12 to 19.
- The number of ASs of subgroup 6 causing alterations on the motor function is most likely between 10 and 11, with plausible range from 8 to 15.
- The elicited distributions for the seven subgroups are plotted together in Figure 1. The number of substances considered differs between subgroups so, to facilitate comparison, the elicited distributions were rescaled to percentage of substances.
- The elicited distributions for the seven subgroups were combined by 1D Monte Carlo simulation, assuming independence between subgroups. This produced a combined distribution for the total number of ASs in the CAG that actually cause alterations of motor function. The median estimate was 104 ASs, with a 90% confidence interval of 100 to 107 (see Figure 2).
- A second Monte Carlo simulation was conducted assuming perfect positive dependence between subgroups. This produced an alternative distribution for the total number of ASs in the CAG that actually cause alterations of motor function. The median estimate was again 104 ASs but with a 90% confidence interval of 97–109 (see Figure 3).
- The experts considered that there will be some, but less than perfect, positive dependence between their judgements for the different subgroups. The distributions in Figures 2 and 3 therefore provide a lower and upper bound for the impact of this dependence on the width of the combined distribution. This will be considered as part of overall uncertainty analysis when the cumulative risk assessment is performed.

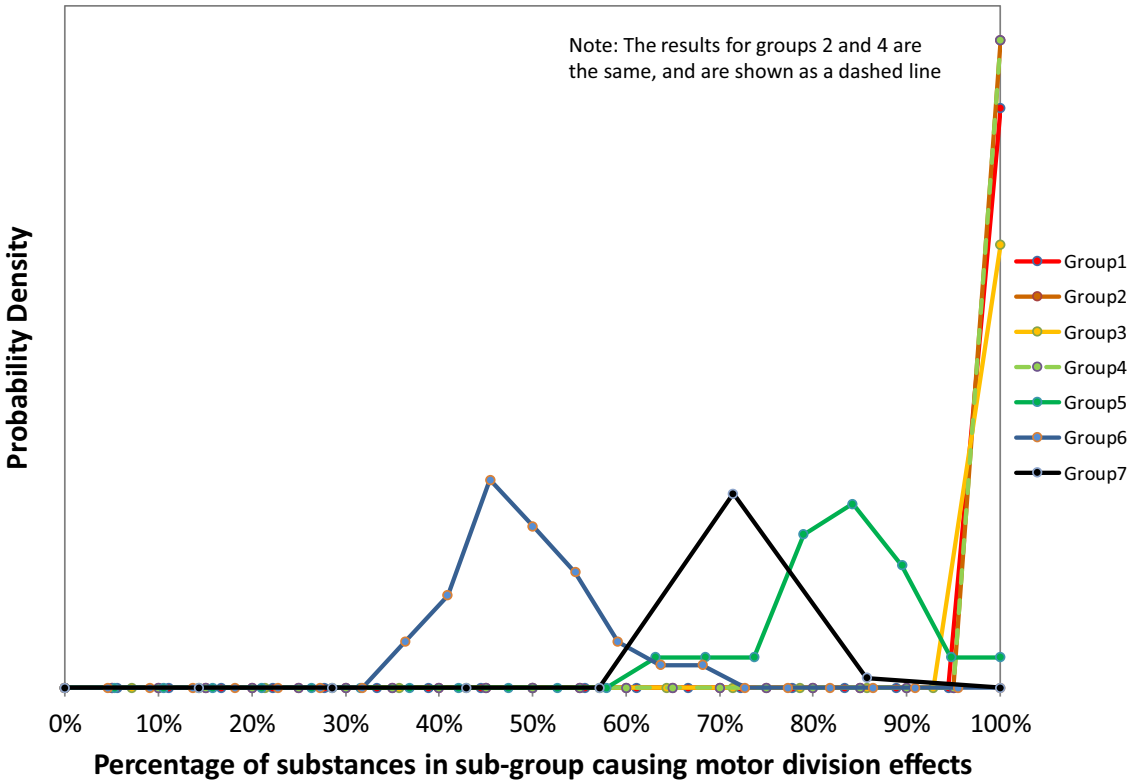

Figure 1: Distributions quantifying uncertainty about the percentage of substances in each subgroup that cause motor division effects. The vertical axis (probability density) quantifies the experts' judgement of the likelihood of different proportions of substances causing motor division effects within each subgroup

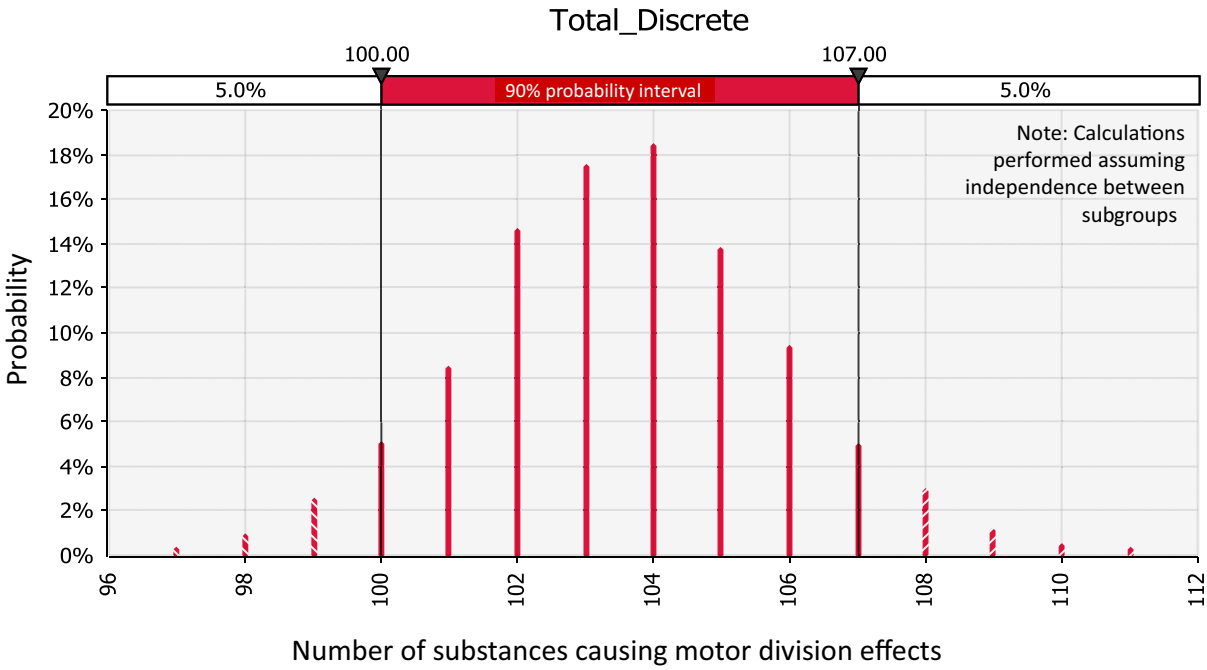

Figure 2: Distribution quantifying uncertainty about the total number of substances from subgroups 1–7 that cause motor division effects, obtained using Monte Carlo simulation assuming that the elicited distributions for the seven subgroups are independent

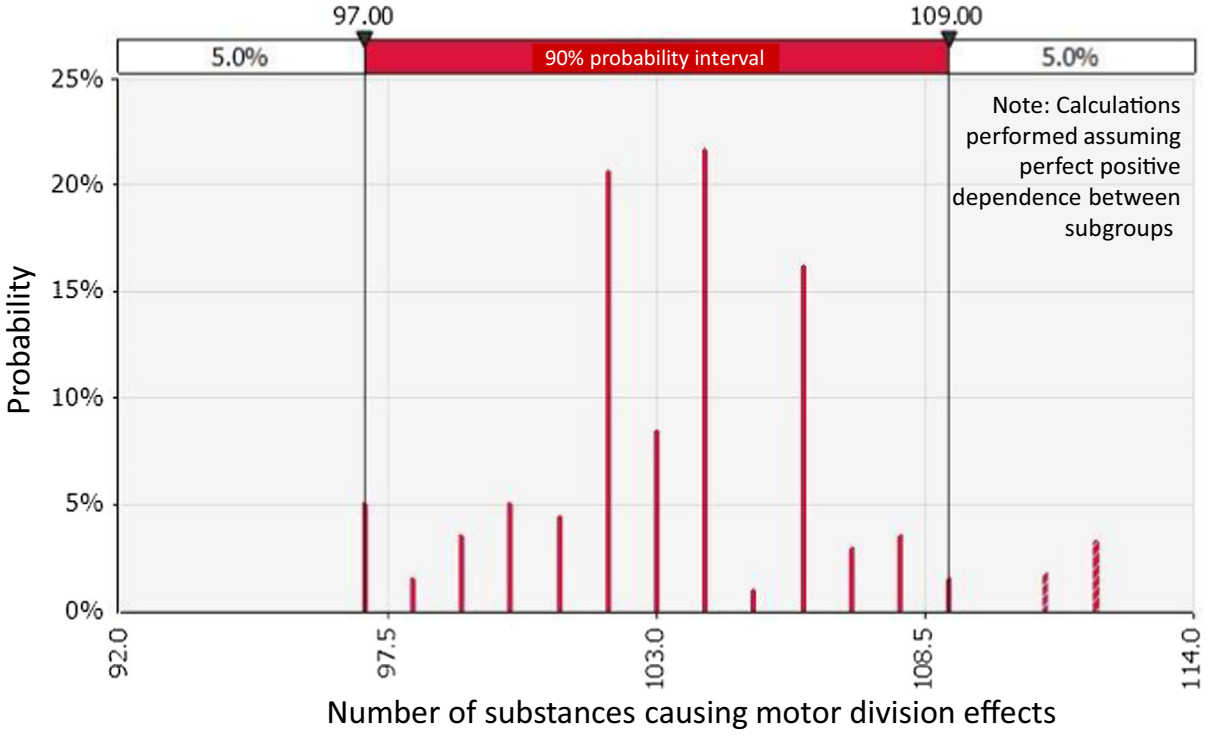

Figure 3: Distribution quantifying uncertainty about the total number of substances from subgroups 1–7 that cause motor division effects, obtained using Monte Carlo simulation assuming perfect positive dependence between the elicited distributions for the seven subgroups

4.2.2. Question 2: How sure is it that these ASs combine their individual toxicities according to the dose addition model at their actual level in food?

Question 2 was ultimately addressed as part of the CRA for the CAG on functional alterations of the motor division, on the basis of the observed risk-drivers (EFSA, 2019b). This was facilitated by the information given in Section 3.3.2 about 11 MoAs identified to be of direct relevance for functional alterations of the motor division, and, in turn, defining 11 clusters of ASs within which dose addition is virtually certain for the respective effect.

Between AS not belonging to the same cluster, the uncertainty about how closely combined effects conform to those predicted by dose addition was addressed in EFSA (2019b) considering the chemical structures and the empirical information on the respective combined toxicity if available. The extent to which this uncertainty impacted the risk assessment depended on various considerations, including the extent to which individual consumers have simultaneous exposures to multiple ASs with different MoAs.

4.3. CAG for the erythrocyte and/or brain AChE inhibition

4.3.1. Question 1: Does the CAG for the brain and/or erythrocyte AChE inhibition contain all ASs contributing to this effect and only ASs causing this effect?

As stated above, question 1 implies providing responses to two discrete questions.

As to *whether all ASs causing the effect are well included* in the CAG, the reader should refer to the first bullet point of Section 4.5 (overall uncertainty analysis).

As to *whether ASs not causing the effect are included* in the CAG, the chance of including ASs not contributing to the effect is here virtually non-existent because all ASs but 2 are organophosphorous or *N*-methyl carbamate insecticides acting biologically via AChE inhibition. The two exceptions are tolclofos-methyl and ethephon, which have other biological actions, but have shown weak, but however significant, inhibition of AChE in experimental studies.

4.3.2. Question 2: How sure is it that these ASs combine their individual toxicities according to the dose addition model at their actual level in food?

Question 2 was addressed when the CRA for brain and/or erythrocyte AChE inhibition was performed in the light of the observed risk drivers (EFSA, 2019b) and their MoAs (All ASs in the CAGs for AChE inhibition belong to the chemical classes a) or d) listed in Section 3.3.2).

4.4. CAGs for the functional alterations of the sensory and autonomic divisions of the nervous system, and for the neuropathological effects

As indicated in Section 3.3.4, it is suggested not to perform CRAs for these CAGs, as those carried out with the CAG for the alterations of the motor division are expected to provide more critical results. Therefore, uncertainty is not considered further for these CAGs.

It is, however, noted that MoAs leading to neuropathological effects are not the same as those leading to the other specific effects. For this reason, the assessment of the modes of neurotoxic action conducted in Section 4.2.2 cannot be used to support uncertainty analyses related to CRA performed for neuropathological effects. For these effects, information on relevant MoAs is scarce, but where available, more details can be found in Table B.9 of Appendix B.

4.5. Overall uncertainty analysis

In subsequent CRAs performed with the CAGs established in the present report, an evaluation of all uncertainties affecting these assessments will be conducted. To address the uncertainties resulting from the composition of the CAG and from the assumption that ASs in the CAG combine their effects by dose addition, it is recommended to consider systematically all relevant sources of uncertainties, including the following:

- Uncertainty related to the composition of the CAG.
  - How certain is it that the CAG includes all the substances contributing to the specific effect of interest? If the CAG does not contain ASs contributing to the risk, the outcome of the

risk assessment might be underestimated. The assessors should consider the probability that ASs causing the specific effect might have not been identified during the data collection procedure (possibility that information of relevance in original toxicological studies is omitted or misreported in summary documents used as source of information) or omitted by the application of the criteria used to populate CAGs, and evaluate their potential contribution to the risk.

  - How certain is it that the CAG includes only ASs contributing to the specific effect of interest? If the CAG contains ASs not contributing to the risk, the outcome of the risk assessment might be overestimated. This needs to be considered in the light of probabilities of CAG membership assessed in Section 4.2.1 (CAG for the functional alteration of the motor division) and Section 4.3.1. (CAG for brain and/or erythrocyte AChE inhibition) and of the individual contribution of each AS to the risk. For the CAGs related to the functional alterations of the sensory and autonomic division and to histological neuropathological changes in neural tissues, this source of uncertainty should be addressed based on appropriate lines of evidence.

- Uncertainty related to the characterisation of ASs included in the CAG.
  - Are all ASs in the CAG characterised with acute and/or chronic NOAELs?
  - Can these acute and/or chronic NOAELs be either under- or overestimated?

    These questions need to be evaluated in the light of the data collection procedure and of the principles used to establish NOAELs. In this respect, the assessors will at least consider the adequacy of the data collection procedure to the principles adopted for the hazard characterisation (including the adopted indicator), the quality of the toxicological dossiers of the ASs included in the CAG (availability of the ad hoc studies for the hazard characterisation, e.g. neurotoxicity studies) and the hazard characterisation principles defined for each CAG (including the eventual measures taken to establishing surrogate NOAELs, e.g. from LOAELs or from chronic studies when acute studies are not available, etc.).
- Uncertainty regarding relative contribution of ASs to the cumulative risk resulting from the use of NOAELs rather than BMDLs.
- Uncertainty regarding the slope and the shape of the dose-response relationship and consequently regarding the effect size at the actual levels of exposure.
- Uncertainty about the contribution of metabolites and degradation products to the cumulative risk. Not only ASs, but also their metabolites and degradation products may contribute to the specific effect. If this contribution is not considered, this needs to be treated as a source of uncertainty when a CRA is performed for any of the CAG related to the effects of pesticides on the nervous system.
- Uncertainty about the adequacy of the dose addition model: How closely will the actual risks for the specific effect of interest conform to those predicted by dose addition? It is recommended to focus on the observed combinations of ASs at the percentiles of the exposure distribution of interest for the risk managers. The evaluation will consider whether risk drivers have similar or dissimilar MoAs. Empirical information on their combined effects in peer-reviewed scientific literature should be considered if available. The distribution of ASs according to their MoA in Section 3.3.2 is useful in the evaluation of this source of uncertainty. It must, however, be kept in mind that this distribution is based on the best known mechanisms and that the use of this distribution implies a simplification of the inherent complexity of the functioning of the nervous system.
- Uncertainties about the inter- and intraspecies variability in toxicological sensitivity. This source of uncertainty concerns the adequacy for human risk assessment of the toxicological characterisation of ASs based on animal data.
- Uncertainties resulting from the use of acute and chronic exposure calculation models not necessarily reflecting the actual time course of toxicokinetic and toxicodynamic processes. For instance, being exposed to a *N*-methylcarbamate insecticide at breakfast and to an organophosphate at dinner may not lead to the same risk as being exposed to the same compounds in the reverse order. This is because although both organophosphorus and *N*-methylcarbamate insecticides bind to AChE, inhibition by the latter is spontaneously reversible whereas that by organophosphorus insecticides is considered irreversible. Functional recovery

following exposure to organophosphorus insecticides requires the synthesis of new enzyme before AChE activity returns to normal values. The cumulative exposure calculation model, which sums up all doses ingested within a 24-hour period, does not take these toxicokinetic and toxicodynamic properties into account and does not produce different results for different sequences of exposure to chemicals.

- If the type of administration (diet or gavage) of the test substance in toxicological studies might be of importance, then this should be flagged in the data tables and considered in the uncertainty analysis.

5. Conclusions

Cumulative assessment groups for the effects of pesticides on the nervous system were previously established by the PPR Panel in 2013. The five specific effects of pesticides on this system that are of relevance for CRA have been confirmed: functional alterations of the motor, sensory and autonomic divisions, histological neuropathological changes in neural tissue, and brain and/or erythrocyte AChE inhibition. The CAGs have been updated on the basis of additional information collected from more recent data collections.

NOAELs have been defined to characterise the ASs included in the CAGs for the respective specific effects. ICs have been proposed to enable cumulative exposure and risk assessments with methods using RPFs.

For an efficient use of resources, the assessment of the cumulative risks of pesticides residues for the nervous system should be focussed on their specific effects on the motor division and on brain and/or erythrocyte AChE inhibition because the highest risks are expected to be observed for these effects.

Sources of uncertainties resulting from the methodological approach and from the limitations in available data and scientific knowledge have been identified and considered in accordance with the anticipated assessment question which will govern CRA conducted with these CAGs.

6. Recommendations

Due to the current scarcity of data with respect of DNT of pesticides, it is currently premature to evaluate if specific effects of pesticides in this area deserve the establishment of CAGs and the performance of CRAs. Therefore, a testing and assessment methodology should be developed and applied on a consistent basis to provide enough information supporting the establishment of CAGs covering DNT if appropriate.

If the outcome of CRAs conducted with the CAGs established in this report exceeds regulatory thresholds of acceptance, empirical research is needed on how ASs driving the risk combine their effects at the anticipated dietary exposure levels, especially if they act according to dissimilar MoAs, and on the extent to which this combination of effects deviates from dose addition.

If the outcome of CRAs conducted with these CAGs, as currently characterised by NOAELs, exceeds the regulatory thresholds of acceptance, an alternative cumulative exposure/risk assessment should be considered with BMDLs used as reference points, after agreement on benchmark dose levels suitable for regulatory purposes. This is not likely to change significantly the outcome of the assessment but would make it independent from the dose selection in toxicological studies and better reflect the actual relative potencies of ASs in the CAG.

The approaches developed in the present report to evaluate uncertainties should be integrated into the CRA which follows. This could be done by incorporating the probabilities of CAG membership into a probabilistic calculation of cumulative risk and taking account of other uncertainties (including those identified in this report and any others arising in the risk assessment) when assessing the overall uncertainty by expert judgement. A simpler alternative would be to do a sensitivity analysis, starting with all subgroups of substances included and removing them one at a time in order of increasing probability of CAG membership, and use the results of this to inform expert judgement of the contribution of CAG membership uncertainty to overall uncertainty. Though less rigorous, this would avoid the need for probabilistic calculations.

The CAGs established in this report should be regularly updated in the light of the toxicological information provided to EFSA in the context of its regulatory activities. This would be the opportunity to include additional non-approved ASs found in food commodities with relevant MoA in the respective CAGs. This would solve a source of underestimation of cumulative risks (EFSA, 2019b).

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Abbreviations

4HNE	4-hydroxy-2-nonenal
5-HT	5-hydroxytryptamine
AChE	acetylcholinesterase
ADI	acceptable daily intake
ALDH	aldehyde dehydrogenase
ANSES	French Agency for Food, Environmental and Occupational Health and Safety
ARfD	acute reference dose
AS	active substance
ATPase	adenosine triphosphatase
BMDL	lower confidence limit of the benchmark dose
CAG	cumulative assessment group
CRA	cumulative Risk Assessment

DAR	Draft Assessment Report
DNT	developmental neurotoxicity
DOPAL	3,4-dihydroxyphenylacetaldehyde
DTU	Danish Technical University
EKE	expert knowledge elicitation
EHC	Environmental Health Criteria Monograph
GABA	gamma-aminobutyric acid
IC	Index Compound
ICPS	International Centre for Pesticides and Health Risk Prevention
JMPR	Joint Meeting on Pesticides Residues
LOAEL	lowest observed adverse effect level
LOEL	lowest observed effect level
MAO	monoamine oxidase
MoA	mode of action
MOE	margin of exposure
MOET	total margin of exposure
MRL	maximum residue level
MTD	maximum tolerated dose
nAChR	nicotinic acetylcholine receptor
NE	norepinephrine
NOAEL	no observed adverse effect level
NOEL	no observed effect level
NTE	neuropathy target esterase
PAFF	Committee Standing Committee on Plants, Animals, Food and Feed
PPR	EFSA Panel on Plant Protection Products and their Residues
RAR	Renewal Assessment Report
RfP	reference point
RIVM	National Institute for Public Health and the Environment
RPF	relative potency factor
SCBI	sodium channel blocker insecticide
TRPV	vanilloid-type transient receptor potential
UF	uncertainty factor
US-EPA	United States Environmental Protection Agency
VGSC	voltage-gated sodium channel

Appendix A – List of active substances considered in view of establishing CAGs for effects of pesticides on the nervous system

Active substances covered by the first outsourced data collection (RIVM, ICPS, ANSES, 2013):

1-Methylcyclopropene	Cyazofamid	Fluazifop-P	Mesosulfuron	Pyridate
1-Naphthylacetamide (1-NAD)	Cyclanilide	Fluazinam	Mesotrione	Pyrimethanil
1-Naphthylacetic acid (1-NAA)	Cycloxydim	Fludioxonil	Metalaxyl-M	Pyriproxyfen
2,4-D	Cyflufenamid	Flufenacet (formerly fluthiamide)	Metaldehyde	Quinmerac
2,4-DB (metabolised to 2,4-D)	Cyfluthrin	Flumioxazin	Metamitron	Quinoclamine
2-Phenylphenol (incl. sodium salt orthophenyl phenol)	Cyhalofop-butyl	Fluometuron	Metazachlor	Quinoxyfen
6-Benzyladenine	Cymoxanil	Fluopicolide	Metconazole	Quizalofop-P-tefuryl
Abamectin (aka avermectin)	Cypermethrin	Fluoxastrobin	Methiocarb (aka mercaptodimethur)	Rimsulfuron (aka renriduron)
Acetamiprid	Cyproconazole	Flupyrsulfuron-methyl (DPX KE 459)	Methomyl	Silthiofam
Acibenzolar-S-methyl (benzothiadiazole)	Cyprodinil	Fluquinconazole	Methoxyfenozide	Sintofen (aka Cintofen)
Aclonifen	Cyromazine	Flurochloridone	Metiram	S-Metolachlor
Alpha-Cypermethrin (aka alphamethrin)	Daminozide	Fluroxypyr	Metosulam	Sodium 5-nitroguaiacolate
Aluminium ammonium sulfate	Dazomet	Flurtamone	Metrafenone	Sodium hypochlorite
Aluminium phosphide	Deltamethrin	Flusilazole	Metribuzin	Sodium o-nitrophenolate
Amidosulfuron	Desmedipham	Flutolanil	Metsulfuron-methyl	Sodium p-nitrophenolate
Amitrole (aminotriazole)	Dicamba	Flutriafol	Milbemectin	Spinosad
Azimsulfuron	Dichlorprop-P	Folpet	Molinate	Spirodiclofen
Azoxystrobin	Diclofop	Foramsulfuron	Myclobutanil	Spiroxamine
Beflubutamid	Diethofencarb	Forchlorfenuron	Napropamide	Sulcotrione
Benalaxyl	Difenoconazole	Formetanate	Nicosulfuron	Sulfosulfuron
Benfluralin	Diflubenzuron	Fosetyl	Omethoate	Sulfuryl fluoride
Bensulfuron	Diflufenican	Fosthiazate	Oryzalin	tau-Fluvalinate
Bentazone	Dimethachlor	Fuberidazole	Oxadiargyl	Tebuconazole
Benthiavalicarb	Dimethenamid-P	Gibberellin	Oxadiazon	Tebufenozide
Benzoic acid	Dimethoate	Glufosinate	Oxamyl	Tebufenpyrad
Beta-Cyfluthrin	Dimethomorph	Glyphosate (incl. trimesium aka sulfosate)	Oxasulfuron	Teflubenzuron
Bifenazate	Dimoxystrobin	Haloxyfop-P/R	Oxyfluorfen	Tefluthrin
Bifenox	Dinocap	Hexythiazox	Paclobutrazol	Tepraloxydim
Bispyribac	Diquat (dibromide)	Hymexazol	Penconazole	Terbuthylazine
Boscalid	Dithianon	Imazalil (aka enilconazole)	Pencycuron	Tetraconazole
Bromadiolone	Diuron	Imazamox	Pendimethalin	Thiabendazole

Bromoxynil	Dodemorph	Imazaquin	Penoxsulam	Thiacloprid
Bromuconazole	Dodine	Imazosulfuron	Pethoxamid	Thiamethoxam
Bupirimate	Epoxiconazole	Imidacloprid	Phenmedipham	Thifensulfuron-methyl
Buprofezin	Esfenvalerate	Indoxacarb	Phosmet	Thiophanate-methyl
Calcium phosphide	Ethephon	Iodosulfuron	Picloram	Thiram
Captan	Ethofumesate	Ioxynil	Picolinafen	Tolclofos-methyl
Carbendazim	Ethoprophos	Iprodione	Picoxystrobin	Tolylfluanid
Carbetamide	Ethoxysulfuron	Iprovalicarb	Pirimicarb	Tralkoxydim
Carboxin	Etofenprox	Isoproturon	Pirimiphos-methyl	Triadimenol
Carfentrazone-ethyl	Etoxazole	Isoxaben	Prochloraz	Tri-allate
Carvone	Etridiazole	Isoxaflutole	Profoxydim (aka Clefoxydim)	Triasulfuron
Chloridazon (aka pyrazone)	Famoxadone	Kresoxim-methyl	Prohexadione (incl. Prohexadione-calcium)	Triazoxide
Chlormequat (chloride)	Fenamidone	Lambda-Cyhalothrin	Propamocarb	Tribenuron (aka metometuron)
Chlorothalonil	Fenamiphos (aka phenamiphos)	Lenacil	Propaquizafop	Triclopyr
Chlorotoluron	Fenazaquin	Linuron	Propiconazole	Trifloxystrobin
Chlorpropham	Fenbuconazole	Lufenuron	Propineb	Triflumizole
Chlorpyrifos	Fenbutatin oxide	Magnesium phosphide	Propoxycarbazone	Triflumuron
Chlorpyrifos-methyl	Fenhexamid	Malathion	Propyzamide	Triflusulfuron
Chlorsulfuron	Fenoxaprop-P	Maleic hydrazide	Proquinazid	Trinexapac (aka cimetacarb ethyl)
Cinidon ethyl	Fenoxycarb	Mancozeb	Prosulfocarb	Triticonazole
Clethodim	Fenpropidin	Maneb	Prosulfuron	Tritosulfuron
Clodinafop	Fenpropimorph	MCPA	Prothioconazole	zeta-Cypermethrin
Clofentezine	Fenpyroximate	MCPB	Pymetrozine	Zinc phosphide
Clomazone	Fipronil	Mecoprop	Pyraclostrobin	Ziram (incl. impurity TMTU)
Clopyralid	Flazasulfuron	Mecoprop-P	Pyraflufen-ethyl	Zoxamide
Clothianidin	Flonicamid (IKI-220)	Mepanipyrim	Pyrethrins	
Copper compounds	Florasulam	Mepiquat	Pyridaben	

Active substances covered by the second outsourced data collection (RIVM, ICPS, ANSES, 2016):

2-chloroethanol	Carbaryl	Ethametsulfuron	Mandipropamid	Prothiofos
8-Hydroxyquinoline incl. oxyquinoleine	Carbofuran	Ethion (aka diethion)	Meptyldinocap	Pyrazophos
Acephate	Carbosulfan	Ethylene oxide	Metaflumizone	Pyridalyl
Acequinocyl	Chlorantraniliprole	Fenarimol	Metalaxyl	Pyriofenone
Acrinathrin	Chlordane	Fenitrothion	Metam (incl. -potassium and -sodium)	Pyroxsulam
Aldicarb	Chlorfenapyr	Fenpropathrin	Methamidophos	Quintozene
Aluminium sulfate	Chlorfenvinphos	Fenpyrazamine	Methidathion	Resmethrin
Ametoctradin	Chlorobenzilate	Fenthion	Methoxychlor	Sedaxane
Aminopyralid	Chromafenozide	Fenvalerate	Metobromuron	Spinetoram

Amisulbrom	Cyantraniliprole	Ferric phosphate	Monocrotophos	Spiromesifen
Amitraz	Cyflumetofen	Fluazifop	Nicotine	Spirotetramat
Anthraquinone	DDT	Flubendiamide	Orthosulfamuron	Sulfoxaflor
Azadirachtin	Diazinon	Flufenoxuron	Oxadixyl	Tecnazene
Azinphos-ethyl	Dichlofluanid	Fluopyram	Oxydemeton-methyl	Tembotrione
Azinphos-methyl	Dichlorvos	Fluxapyroxad	Parathion	Tetradifon
Benalaxyl-M	Dicloran	Fonofos	Parathion-methyl	Tetramethrin
Benfuracarb	Dicofol	Halosulfuron methyl	Penflufen	Thiencarbazone
Benomyl	Dicrotophos	HCH	Penthiopyrad	Thiodicarb
Benzalkonium chloride	Didecyldimethylammonium chloride	Heptachlor	Permethrin	Tolfenpyrad
Beta-cypermethrin	Dieldrin	Hexachlorobenzene	Phenthoate	Topramezone
Bifenthrin	Dinotefuran	Hexaconazole	Phosalone	Triadimefon
Bitertanol	Diphenylamine	Indolylbutyric acid	Phosphane	Triazophos
Bixafen	Dithiocarbamates	Ipconazole	Phoxim	Trichlorfon
Bromide ion	Emamectin benzoate	Iron sulfate	Pinoxaden	Trifluralin
Bromopropylate	Endosulfan	Isoprocarb	Procymidone	Valifenalate
Cadusafos (aka ebufos)	Endrin	Isopyrazam	Profenofos	Vinclozolin
Camphechlor	EPN	Lindane	Propargite	

Active substances covered by the EFSA data collection:

Aluminium phosphide	Etoxazole	Pymetrozine
Benthiavalicarb	Fenpyroximate	Pyriproxyfen
Bifenazate	Iprovalicarb	Tebuconazole
Copper compounds	Lufenuron	Tebufenpyrad
Cyromazine	Magnesium phosphide	Tetraconazole
Difenoconazole	Metamitron	Thiamethoxam
Diflubenzuron	Metribuzin	Thiophanate-methyl
Etofenprox	Milbemectin	Tolylfluanid

Appendix B – Tables supporting Cumulative Risk Assessments using the CAGs for effects of pesticides on the nervous system

Note 1: In following tables, the names of persons involved in testing on vertebrate animals are confidential and not shown in the study reference details.

Table B.1: CAG on functional effects on motor division: toxicological characterisation of ASs to be considered in acute exposure/risk assessments

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
2,4-D	Abnormal gait, uncoordinated movements and reduced motor activity	75	250	Acute neurotoxicity rat [redacted] Administration via gavage	Source: RAR 2013 EFSA conclusions on 2,4-D (2014) considered	Unknown
Abamectin	Ataxia	1.5	6	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2005, addendum 2007 EFSA conclusions on abamectin (2008) considered	GABA-gated chloride channel agonist
Acephate		74.2	–	49-day neurotoxicity study [redacted] Administration via diet Highest tested dose	Source: JMPR 2002	AChE inhibitor
Acetamiprid	Reduced motor activity, tremor	10	30	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2001	Agonist of nAChR
Acrinathrin	Abnormal gait	1	3	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2007 EFSA conclusions on acrinathrin (2013) considered	Binding to VGSC
Aldicarb	Decreased motor activity, tremor, ataxic gait, decreased hindlimb strength	0.1	0.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996	AChE inhibitor
Alpha-Cypermethrin	Ataxia	2.3	6.8	90-day dog [redacted] Administration via diet	Source: DAR 2000	Binding to VGSC
Amitraz	Hypoactivity	1	4	90-day dog [redacted] Administration via gavage	Source: DAR Neurotoxicity studies not available	Partial agonist of presynaptic α2-adrenergic receptor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Azinphos-ethyl	AChE inhibition (erythrocyte)	0.0125	0.025	90-day dog [redacted] Administration via diet	Source: JMPR 1973 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Azinphos-methyl	Reduced motor activity, reduced grip strength, abnormal gait	2	6	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996, Addendum 6, 2000 (neurotoxicity)	AChE inhibitor
Benfuracarb	Increased motor activity, tremor, hyperactivity	2	20	28-day rat [redacted] Administration via gavage	Source: DAR 2004 Acute neurotoxicity study not available	AChE inhibitor
Beta-Cyfluthrin	Reduced motor activity, tremor, uncoordinated gait	2	10	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2006, DAR 1996	Binding to VGSC
Beta-cypermethrin	Abnormal gait, tremor	1	10	90-day dog [redacted] Administration via capsule	Source: DAR 2013	BINDING to VGSC
Bifenthrin	Increased motor activity, tremor, convulsion, abnormal gait	35	75	Acute neurotoxicity rat [redacted] Administration via diet	Source: DAR	Binding to VGSC
Cadusafos	Decreased hindlimb strength	25	40	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR, addendum 2005	AChE inhibitor
Carbaryl	Tremors, decreased motor activity, ataxic gait	10	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	AChE inhibitor
Carbofuran		0.3	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: Revised DAR 2008	AChE inhibition
Carbosulfan	Reduced motor activity	5	30	Acute neurotoxicity rat [redacted] Administration via gavage	Source: Revised DAR 2009	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Chlorpropham	Reduced motor activity	50	125	Acute dog [redacted]	Source: DAR 1999, JMPR 2005	Unknown
Chlorfenvinphos	AChE inhibition (brain, erythrocyte)	0.15	15	2-year rat (author not reported, JMPR 1994) Administration via gavage	Source: JMPR 1994 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Chlormequat	Ataxia	10	32	1-year dog [redacted] Administration via diet	Source: DAR 2007 Neurotoxicity studies not available ARfD: 0.09 mg/kg bw	Partial agonist of muscarinic and nicotinic acetylcholine receptor
Chlorpyrifos	Decreased motor activity, decreased grip performance	10	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1999	AChE inhibitor
Chlorpyrifos-methyl	–	75	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: RAR 2017	AChE inhibition
Clothianidin	Decreased activity, tremor	200	400	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003	Agonist of nAChR
Cyfluthrin	Choreoathetosis	1	2.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2006	Binding to VGSC
Cypermethrin	Decreased activity, decreased grip strength, splayed legs, landing foot splay, abnormal locomotion, lateral head movements	20	60	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR EC review report on cypermethrin (2005) considered.	Binding to VGSC
Deltamethrin	Ataxia, landing-foot splay, tremor	1	10	1-year dog [redacted] Administration via capsule	Source: DAR 1998 EC review report on deltamethrin (2002) considered	Binding to VGSC
Diazinon	Ataxic gait	2.5	150	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 EFSA conclusions on diazinon (2006) considered	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Dichlorvos	Ataxia	8	16	28-day rat [redacted] Administration via diet	Source: DAR 2003 Neurotoxicity studies not available	AChE inhibition
Dicofol	Ataxia	15	75	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2006	4 presumed MoA (Section 3.3.2)
Dieldrin	Increased motor activity, convulsions, tremors	1	5	3-days rat-mechanistic study [redacted] Administration via gavage	Source: PPR panel, 2007 Neurotoxicity studies not available	GABA-gated chlorine channel antagonist
Dimethoate	Ataxia, convulsions, reduced grip strength, reduced motor activity, tremor	20	200	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR	AChE inhibitor
Dinotefuran	Reduced motor activity	100	300	Acute single dose rabbit [redacted] Administration via gavage	Source: JMPR 2012	Agonist of nAChR
Emamectin benzoate	Tremor	5 (1)	10	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2008 EFSA conclusions on emamectin benzoate (2012) considered Additional UF of 5 due to small dose spacing and steep dose response curve, and possible acute effects in dogs at 5 mg/kg bw	GABA-gated chloride channel agonist
Endosulfan	Increased motor activity, tremor, reduced motor activity, convulsions, abnormal gait	3	6	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2001 and addendum	GABA-gated chloride channel blocker
Endrin	Convulsions	0.025	0.05	2-year dog [redacted] Administration via diet	Source: EHC 130, 1992 Neurotoxicity studies not available ADI: 0.0002 (JMPR 1994)	GABA-gated chloride channel antagonist
Esfenvalerate	Reduced grip strength	3.2	6.4	90-day neurotoxicity rat [redacted] Administration via diet	Source: JMPR 2002	Binding to VGSC

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Ethephon	Reduced motor activity	500	1,000	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	AChE inhibitor
Ethion	AChE inhibition (brain)	0.06	0.71	90-day dog [redacted] Administration via diet	Source: JMPR 1990 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Ethoprophos	Reduced motor activity, abnormal posture and gait	5	25	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	AChE inhibitor
Fenamiphos	Ataxia, muscle fasciculation	0.37	1.52	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003	AChE inhibitor
Fenitrothion	Increased motor activity, tremor, abnormal gait, hypoactivity, reduced grip strength	12.5	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003	AChE inhibitor
Fenpropathrin	Increased motor activity, tremor	15	30	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2012	Binding to VGSC
Fenthion	Reduced motor activity	1.04	49.05	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996, Addendum Tox 2001	AChE inhibitor
Fenvalerate	Ataxia, tremor	20	80	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2012	Binding to VGSC
Fipronil	Landing-foot splay	2.5	7.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	GABA-gated chloride channel blocker
Flufenacet	Ataxia, reduced motor activity	7.5	75	Acute neurotoxicity rat [redacted]	Not true NOAEL, but LOAEL divided by 10	Unknown
Fonofos	Increased motor activity, abnormal gait	4	7	Acute neurotoxicity rat (author not reported) Administration via gavage	Source: US EPA 1999 No reference values available	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Formetanate	Ataxia, tremor, reduced motor activity	1	10	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	AChE inhibitor
Fosthiazate	Ataxia, uncoordinated movements	5.4	26.8	28-day dog [redacted] Administration via capsule	Source: DAR 1998 Neurotoxicity studies not available	AChE inhibitor
Glufosinate	Hunched posture	100	500	Acute neurotoxicity rat [redacted]	EFSA 2005	Unknown
Heptachlor	Increased motor activity, convulsions	0.5	5	6-month rat [redacted] Administration via gavage	Source: JMPR 1970, CICADS 2006 Not true NOAEL, but LAOEL divided by 10. ADI: 0.0001 mg/kg (JMPR 1994) 14-day neurotoxicity rat [redacted]: No effect at 69 mg/kg bw per day (highest tested dose)	GABA-gated chloride channel antagonist
Imidacloprid	Tremor	23.5	45.4	90-day dog [redacted] Administration via diet	Source: DAR 2005	Agonist of nAChR
Indoxacarb	Decreased motor activity	50	100	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2000	Voltage-dependent sodium channel blocker
lambda-Cyhalothrin	Ataxia, tremor, convulsion	0.5	3.5	1-year dog [redacted] Administration via capsule Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996, JMPR 2007 EFSA conclusions on lambda-cyhalothrin (2014) considered	Binding to VGSC
Lindane	Increased forelimb grip strength	6	20	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2002 JMPR evaluation of lindane (2002) considered.	GABA-gated chloride channel antagonist
Malathion	Reduced ambulatory activity and total motor activities, reduced hindlimb resistance	1,000	2,000	Acute neurotoxicity rats [redacted] Administration via gavage	Source: DAR	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Mepiquat	Lateral position, spasm	32	95	3-month dogs [redacted] Administration via diet	Source : DAR 2005 EFSA conclusions on mepiquat (2008) considered	Activation of nicotinic and muscarinic acetylcholine receptors
Metaldehyde	Ataxia, reduced motility	30	75	1-year dog [redacted]	Source: DAR 2009 EFSA conclusions on metaldehyde (2010) considered.	GABA inhibitor (presumed)
Methamidophos	Increased or decreased muscle tone, abnormal gait, tremor, hypoactivity	1	3	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2000	AChE inhibitor
Methidathion	Tremor, ataxia	4	8	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 1997 addendum	AChE inhibitor
Methiocarb	Tremor	0.5	1.5	Developmental study rat [redacted] Administration via gavage	Source: DAR 2004 EFSA (2018) Neurotoxicity studies not available Same level as NOAEL for AchE inhibition	AChE inhibitor
Methomyl	Tremor	0.75	2	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR	AChE inhibitor
Milbemectin	Staggering gait, tremor	10	30	13-week dog [redacted] Administration via capsule	Source: DAR 2001, RAR, 2017	Glutamate-gated chloride (GluCl) allosteric modulator
Monocrotophos	Tremor	0.4	2.5	2-year dog [redacted] Administration via diet	Source: JMPR 1972 Neurotoxicity studies not available. NOAEL for AChE inhibition: 0.1 mg/kg bw	AChE inhibitor
Omethoate (metabolite of dimethoate)	Increased motor activity, tremor, abnormal gait, reduced grip strength	0.35	5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 (dimethoate)	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Oxamyl (IC)	Ataxia, hunched posture, landing-foot splay, reduced motor activity, tremor	0.1	0.75	Acute neurotoxicity [redacted] Administration via gavage	Source: DAR 2003 EFSA conclusions on oxamyl (2006) considered	AChE inhibitor
Oxydemeton-methyl	Increased or decreased muscle tone	1.88	3.75	90-day dog [redacted] Administration via diet	Source: DAR 2004 Neurotoxicity studies not available NOAEL for AChE inhibition: 0.2 mg/kg bw	AChE inhibitor
Parathion	Tremor	1.75	5.6	90-day rat [redacted] Administration via diet	Source: ECCO 2001, JMPR 1995 Neurotoxicity studies not available NOAEL for AChE inhibition: 0.25 mg/kg bw	AChE inhibitor
Parathion-methyl	Tremor	1.69	5.9	90-day rat [redacted] Administration via diet	Source: DAR 2001 Neurotoxicity studies not available NOAEL for AChE inhibition: 0.25 mg/kg bw	AChE inhibitor
Penflufen	Reduced motor activity	50	100	Acute neurotoxicity rat [redacted]	EFSA (2012)	Unknown
Permethrin	Abnormal gait, tremor	150	300	Single dose rat [redacted] Administration via gavage	Source: DAR 1998, JMPR 1999	Binding to VGSC
Phenthoate	AChE inhibition (erythrocytes)	0.29	0.87	2-year dog [redacted] Administration via diet	Source: JMPR 1980 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate.	AChE inhibitor
Phosalone	Tremor, hypoactivity	25	60	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 EFSA (2006)	AChE inhibitor
Phosmet	Ataxia, tremor	22.5	–	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 EFSA (2006)	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Phoxim	Convulsions	5	25	90-day dog [redacted] Administration via diet	Source: JECFA 1999 Neurotoxicity studies not available NOAEL for AChE inhibition: 1.3 mg/kg bw	AChE inhibitor
Pirimicarb	Hunched posture	10	40	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003 EFSA (2005)	AChE inhibitor
Pirimiphos-methyl	Convulsions, reduced grip strength	150	1,500	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003	AChE inhibitor
Profenofos	Ataxia, abnormal gait, tremors, altered posture	190	380	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2007	AChE inhibitor
Pymetrozine	Decreased motor activity	12.5	125	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1998 (and addendum 1999) Not true NOAEL, but LOAEL divided by 10	Chordotonal organ TRPV channel modulator (presumed)
Pyrazophos	Increased or decreased muscle tone, weakness, abnormal gait	0.45	8	6-month dog [redacted] Administration via diet	Source: DAR 1998 Neurotoxicity studies not available NOAEL for AChE inhibition: 0.05 mg/kg bw	AChE inhibitor
Pyrethrins	Tremor	20	63	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2007, 2012 EFSA (2013)	Binding to VGSC
Pyridate	Prostration, tremor, hunched posture, hypoactivity	40	80	90-day dog [redacted]	Source: RAR 2013	Unknown
Spirotetramat	Reduced motor activity	100	200	Acute neurotoxicity rat [redacted]	Source: DAR 2012 EFSA (2013)	Unknown
Sulfoxaflor	Reduced motor activity	25	75	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2012 EFSA (2014)	Nicotinergic AChR partial agonist

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Tau-fluvalinate	Altered posture	10	60	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2007	Binding to VGSC
Tefluthrin	Tremor	0.5	1.5	90-day dog [redacted] Administration via capsule	Source: DAR 2006 EFSA (2010)	Binding to VGSC
Tembotrione	Reduced motor activity	200	500	Acute neurotoxicity rat [redacted]	Source: DAR 2012 EFSA (2013)	Unknown
Tetramethrin	Increased motor activity: tremor	31	63	6-month dog [redacted] Administration via diet	Source: WHO 1990 Neurotoxicity studies not available No reference values available	Binding to VGSC
Thiacloprid	Reduced motor activity	3.1	11	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2000, 2017 EC review report on thiacloprid (2004) considered	agonist of nAChR
Thiamethoxam	Impairment of gait, increased forelimb grip strength, hypoactivity	100	500	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2002 EC review report on thiamethoxam (2007) considered	agonist of nAChR
Thiodicarb	Reduced motor activity, ataxia, tremor	5	20	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003	AChE inhibitor
Thiram	Reduced grip strength	5	150	Acute neurotoxicity rat [redacted]	Source: DAR 2003	Neurotoxic effect might be due to the metabolite CS_2 (presumed)
Tolclofos-methyl	Decreased motor activity	200	700	Acute neurotoxicity rat [redacted] Administration via gavage	Source: RAR 2016	AChE inhibitor
Triadimefon	Increased motor activity, abnormal gait	2	35	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2004 JMPR evaluations on triademefon (2004) considered	Inhibition of dopamine transporter (presumed)

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Tri-allate	flat footed appearance/decreased motor activity	60	300	Acute neurotoxicity [redacted]	Source: DAR 2007 EFSA conclusions on tri-allate (2009) considered	Unknown
Triazophos	AChE inhibition (erythrocytes)	0.012	0.13	1-year dog [redacted]	Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Trichlorfon	Increased or decreased muscle tone	10	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2002	AChE inhibitor
zeta-Cypermethrin	Ataxia, tremor, convulsions, staggered gait, splayed hindlimbs	10	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 and addendum 2005 EFSA conclusions on zeta-cypermethrin (2008) considered	Binding to VGSC
Ziram	Ataxia, hunched posture	15	300	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1998	Neurotoxic effect might be due to the metabolite CS_2 (presumed)

CAG: cumulative assessment group; AS: active substance; NOEL: no observed effect level; LOEL: lowest observed effect level; NOAEL: no observed adverse effect level; LOAEL: lowest observed adverse effect level; bw: body weight; GABA: gamma-aminobutyric acid; AChE: acetylcholinesterase; VGSC: voltage-gated sodium channel; nAChR: nicotinic acetylcholine receptor; MoA: mode of action.

Table B.2: CAG on functional effects on sensory division: toxicological characterisation of ASs to be considered in acute exposure/risk assessments

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Abamectin	Reduced splay reflex	0.5	1.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2005, addendum 2007 EFSA conclusions on abamectin (2008) considered	GABA-gated chloride channel agonist
Acephate	–	74.2	–	49-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2002	AChE inhibitor
Acetamiprid	–	100	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2001	Agonist of nAChR
Acrinathrin	–	37.5	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2007	Binding to VGSC
Aldicarb	Decreased reactivity: tail pinch response	0.1	0.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996	AChE inhibitor
alpha-cypermethrin	Righting reflex	20	40	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2000	Binding to VGSC
Amitraz	hyperreactivity	12	50	90-day rat [redacted] Administration via gavage	Source: DAR Neurotoxicity studies not available	Partial agonist of presynaptic α2-adrenergic receptor
Azinphos-ethyl	AChE inhibition (erythrocyte)	0.0125	0.025	90-day dog [redacted] Administration via diet	Source: JMPR 1973 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Azinphos-methyl	Decreased reactivity: righting reflex (air drop)	2	6	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996, Addendum 6, 2000 (neurotoxicity)	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Benfuracarb	Decreased reactivity: analgesic reflex (nociception response)	2	20	28-day rat [redacted] Administration via gavage	Source: DAR 2004 Acute neurotoxicity study not available	Known AChE inhibitor
beta-Cyfluthrin	Decreased touch responses, tail pinch response and impaired righting.	2	10	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2006, DAR 1996	Binding to VGSC
beta-cypermethrin	Decreased reactivity: hypoactivity, tail pinch response	100	500	Acute neurotoxicity rat [redacted]	Source: DAR 2013	Known Binding to VGSC
Bifenthrin	–	75	–	Acute neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	Known Binding to VGSC
Cadusafos	–	40	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR, addendum 2005	AChE inhibitor
Carbaryl	Impaired toe pinch	10	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	AChE inhibitor
Carbofuran	–	0.3	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: Revised DAR 2008	AChE inhibition
Carbosulfan	Slot tail pinch response	5	30	Acute neurotoxicity rat [redacted] Administration via gavage	Source: Revised DAR 2009	AChE inhibitor
Chlorfenvinphos	AChE inhibition (brain, erythrocyte)	0.15	15	2-year rat (author not reported, JMPR 1994) Administration via gavage	Source: JMPR 1994 Neurotoxicity studies not available NOAEL for AchE inhibition used as surrogate	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Chlormequat	Decreased reflex response	50	62.5	90-day dog [redacted] Administration via det	Source: DAR 2007	Partial agonist of muscarinic and nicotinic acetylcholine receptor
Chlorpyrifos	–	100	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 1999	AChE inhibitor
Chlorpyrifos-methyl	–	75	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: RAR 2017	AChE inhibition
Clothianidin	Pin-point constriction	200	400	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003	Agonist of nAChR
Cyfluthrin	–	7.5	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: JMPR 2006	Binding to VGSC
Cypermethrin	–	60	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR	Binding to VGSC
Deltamethrin	Absent approach response, absent touch response, absent startle and tail pinch responses, altered air righting reflex	15	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: Source: DAR 1998, addendum	Binding to VGSC
Diazinon	Decreased tail pinch response, impaired righting reflex	300	600	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	AChE inhibitor
Dicamba	Abnormal righting reflex. Increased tail flick latency time	30	300	Acute neurotoxicity rat [redacted] Administration via gavage	Source: revised DAR Not true NOAEL, but LOAEL divided by 10	Unknown

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Dichlorvos	AChE inhibition (erythrocytes)	0.1	1.5	13-week rat [redacted]	Source: DAR 2003 (addendum) Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Dicofol	Decreased reactivity-hypoactivity	75	350	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2006	4 presumed MoA (Section 3.3.2)
Dieldrin		0.3		Virtual NOAEL corresponding to the ARfD of 0.003 mg/kg bw/d derived by the PPR Panel and considered as protective for all endpoints.	Source: PPR panel, 2007 Neurotoxicity studies not available	GABA-gated chlorine channel antagonist
Dimethoate	Absence of pupil response	2	20	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR	AChE inhibitor
Dinotefuran	–	1,500	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: JMPR 2012	Agonist of nAChR
Emamectin benzoate	–	82.2 (16.4)	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2008 EFSA (2012) Additional UF of 5 due to small dose spacing and steep dose response curve, and possible acute effects in dogs at 5 mg/kg bw	GABA-gated chloride channel agonist
Endosulfan	Hyperreactivity	3	6	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2001 and addendum	GABA-gated chloride channel blocker
Endrin (IC)	Hyperreactivity	0.05	1.0	2-year rat [redacted] Administration via diet	Source: EHC 130, 1992 Neurotoxicity studies not available ADI: 0.0002 (JMPR 1994)	GABA-gated chloride channel antagonist

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Esfenvalerate	Increased reaction to touch	20	80	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2002 Approved.	Binding to VGSC
Ethephon	–	2,000	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2004	AChE inhibitor
Ethion	AChE inhibition (brain)	0.06	0.71	90-day dog [redacted] Administration via diet	Source: JMPR 1990 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Ethoprophos	–	25	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2004	AChE inhibitor
Fenamiphos	–	2.31	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2003	AChE inhibitor
Fenitrothion	Decreased reactivity: tail pinch response, righting reflex (air drop)	12.5	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003	AChE inhibitor
Fenpropathrin	–	30	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: JMPR 2012	Binding to VGSC
Fenthion	Impaired righting reflex, increased touch response	1.04	49.05	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996, Addendum Tox 2001	AChE inhibitor
Fenvalerate	–	360	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: JMPR 2012	Binding to VGSC

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Fipronil	Approach response, tail pinch response, air righting reflex	5	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	GABA-gated chloride channel blocker
Fonofos	Decreased reactivity: patellar reflex	4	7	Acute neurotoxicity rat (author not reported) Administration via gavage	Source: US EPA 1999 No reference values available	AChE inhibitor
Formetanate	Diminished reaction to tail pinch test, abnormal response to visual placing test, auditory startle response	1	10	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	AChE inhibitor
Fosthiazate	AChE inhibition (brain)	0.54	5.4	90-day dog [redacted] Administration via capsule	Source: DAR 1998 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Halosulfuron methyl	Decreased reactivity: righting reflex (air drop)	600	2,000	Acute neurotoxicity rat [redacted]	Source: DAR 2007	Unknown
Heptachlor	Hyperreactivity	2	7	14-day neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 1970, CICADS 2006 ADI: 0.0001 mg/kg (JMPR 1994)	GABA-gated chloride channel antagonist
Imidacloprid	–	307	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2005	Agonist of nAChR
Indoxacarb	–	100	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2000	Voltage-dependent sodium channel blocker
Lambda-cyhalothrin	More energetic response to touch	1	10	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR Not true NOAEL, but LOAEL divided by 10	Binding to VGSC

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Lindane		6			Neurotoxicity studies not available NOAEL of 6 mg/kg bw/d used by JMPR (2002) to establish the ARfD used as surrogate	
Malathion	–	2,000	–	Acute neurotoxicity rats [redacted] Administration via gavage Highest tested dose	Source: DAR	AChE inhibitor
Mepiquat	Lack of pupillary reflex	300	1,200	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2005	Activation of nicotinic and muscarinic acetylcholine receptors
Metaldehyde	Reduced righting reflex, reduced toe/tail pinch response	150	250	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR	Presumed GABA inhibitor
Methamidophos	–	9	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2000	AChE inhibitor
Methidathion	Decreased reactivity: analgesic reflex (nociception response)	4	5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 1997 addendum	AChE inhibitor
Methiocarb	AChE inhibition (erythrocytes)	0.5	2	28-day rat [redacted] Administration via gavage	Source: DAR 2004 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Methomyl	–	2	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Milbemectin	–	160	–	Acute neurotoxicity rat [redacted] Administration via gavage	Source: RAR 2017	GABA-gated chloride channel agonist
Monocrotophos	AChE inhibition (brain, erythrocytes)	0.1	0.3	Single dose rat [redacted] Administration via gavage	Source: JMPR 1995, addendum Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Omethoate (metabolite of dimethoate)	Decreased reactivity: pupil response	0.35	5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 (dimethoate)	AChE inhibitor
Oxamyl (IC)	Righting reflex, tail pinch	0.1	0.751	Acute neurotoxicity [redacted] Administration via gavage	Source: DAR 2003	AChE inhibitor
Oxydemeton-methyl	AChE inhibition (brain, erythrocytes)	0.2	0.6	14-day rat [redacted] Administration via diet	Source: DAR 2004 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Parathion	AChE inhibition (brain)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: JMPR 1995 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Parathion-methyl	AChE inhibition (erythrocytes)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: DAR 2001 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Permethrin	Hyperreactivity	150	300	Single dose rat [redacted] Administration via gavage	Source: JMPR 1999	Binding to VGSC

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Phenthoate	AChE inhibition (erythrocytes)	0.29	0.87	2-year dog [redacted] Administration via diet	Source: JMPR 1980 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Phosalone	–	60	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2004	AChE inhibitor
Phosmet	–	22.5	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2004	AChE inhibitor
Phoxim	AChE inhibition (erythrocytes)	1.3	5	90-day dog [redacted] Administration via diet	Source: JECFA 1999 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Pirimicarb	–	110	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2003	AChE inhibitor
Pirimiphos-methyl	–	1,500	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2003	AChE inhibitor
Profenofos	–	380	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: JMPR 2007	AChE inhibitor
Pyrazophos	AChE inhibition (erythrocytes)	0.05	0.125	2-year dog [redacted] Administration via diet	Source: JMPR 1992 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Pyrethrins	–	200	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2007, 2012	Binding to VGSC
Sulfoxaflor	Decreased reactivity: touch response (handling reactivity)	75	750	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2012	Nicotinergic AChR partial agonist
Tau-fluvalinate	–	100	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2007	Binding to VGSC
Tebuconazole	Poor reflexes	10§	100	Acute mouse [redacted] Administration via gavage	Source: DAR 2008 Not true NOAEL, but LOAEL divided by 10	Unknown
Tefluthrin	–	10	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2006	Binding to VGSC
Tembotrione	Decreased reactivity: approach response	500	2,000	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2012	Unknown
Tetramethrin					Information insufficient No reference values available	Binding to VGSC
Thiacloprid	–	109	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2000, RAR, 2017	Agonist of nAChR
Thiamethoxam	Uncoordinated landing in the righting reflex	100	500	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2002 EC review report on thiamethoxam (2007) considered	Agonist of nAChR
Thiodicarb	Decreased reactivity: tail pinch response	5	20	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Thiram	Handling reactivity, approach response, startle response, air righting	5	150	Acute neurotoxicity rat [redacted]	Source: DAR 2003	Neurotoxic effect might be due to the metabolite CS_2 (presumed)
Trichlorfon	Decreased reactivity: righting reflex (air drop)	10	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2002	AChE inhibitor
Tolclofos-methyl	–	2,000	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: RAR 2016	AChE inhibitor
Triazophos	AChE inhibition (erythrocytes)	0.012	0.13	1-year dog [redacted] Administration via diet	Source: JMPR 2002 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
zeta-Cypermethrin	Righting reflex	50	250	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 and addendum 2005	Binding to VGSC

CAG: cumulative assessment group; AS: active substance; NOEL: no observed effect level; LOEL: lowest observed effect level; NOAEL: no observed adverse effect level; LOAEL: lowest observed adverse effect level; bw: body weight; GABA: gamma-aminobutyric acid; AChE: acetylcholinesterase; VGSC: voltage-gated sodium channel; nAChR: nicotinic acetylcholine receptor; MoA: mode of action.

Table B.3: CAG on functional effects on autonomic division: toxicological characterisation of ASs to be considered in acute exposure/risk assessments.

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Abamectin	–	6	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2005, addendum 2007	GABA-gated chloride channel agonist
Acephate	–	74.2	–	49-day neurotoxicity study [redacted] Administration via diet Highest tested dose	Source: JMPR 2002	AChE inhibitor
Acetamiprid	Urination	10	30	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2001	Agonist of nAChR
Acrinathrin	Salivation	10	37.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2007	Known Binding to VGSC
Aldicarb	Salivation, lacrimation	0.1	0.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996	Known AChE inhibitor
Alpha-Cypermethrin	Salivation	4	10	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2000	Known Binding to VGSC
Amitraz	Hypoactivity	1	4	90-day dog [redacted] Administration via diet	Source: DAR Neurotoxicity studies not available, therefore the source of the ARfD (0.01 mg/kg bw/d, SCoFCAH 2003) is used as surrogate	Partial agonist of presynaptic α2-adrenergic receptor
Azinphos-ethyl	AChE inhibition (erythrocyte)	0.0125	0.025	90-day dog [redacted] Administration via diet	Source: JMPR 1973 Neurotoxicity studies not available NOAEL for AchE inhibition used as surrogate	AChE inhibitor
Azinphos-methyl	Salivation, lacrimation	2	6	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996, Addendum 6, 2000 (neurotoxicity)	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Benfuracarb	Lacrimation	2	20	28-day rat [redacted] Administration via gavage	Source: DAR 2004 Acute neurotoxicity study not available	AChE inhibitor
Beta-Cyfluthrin	Salivation	2	10	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2006, DAR 1996	Binding to VGSC
Beta-cypermethrin	Salivation	20	100	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2013	Binding to VGSC
Bifenthrin	–	75	–	Acute neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	Binding to VGSC
Cadusafos	Urination	25	40	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR, addendum 2005	AChE inhibitor
Carbaryl	Salivation, urination	10	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	AChE inhibitor
Carbofuran	–	0.3	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: Revised DAR 2008	AChE inhibition
Carbosulfan	–	30	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: Revised DAR 2009	AChE inhibition
Chlorfenvinphos	AChE inhibition (brain, erythrocyte)	0.15	15	2-year rat (author not reported, JMPR, 1994) Administration via gavage	Source: JMPR 1994 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Chlormequat	salivation	5	10	1-year dog [redacted] Administration via diet	Source: DAR 2007 Neurotoxicity studies not available	Partial agonist of muscarinic and nicotinic acetylcholine receptor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Chlorpropham	Salivation (accompanied by vomiting and retching)	125	625	14-day dog [redacted] Administration via capsule	Source: DAR 1999	Unknown
Chlorpyrifos	–	100	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 1999	AChE inhibitor
Chlorpyrifos-methyl	–	75	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: RAR 2017	AChE inhibition
Clothianidin	–	400	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2003	Agonist of nAChR
Cyfluthrin	Salivation	2.5	7.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2006	Binding to VGSC
Cypermethrin	Urination	20	60	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR	Binding to VGSC
Deltamethrin	Mydriasis	1	2.5	90-day dog [redacted] Administration via capsule	Source: Source: DAR 1998 EC Review report on deltamethrin (2002) considered	Binding to VGSC
Diazinon	Lacrimation, salivation	300	600	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	AChE inhibitor
Dichlorvos	AChE inhibition (erythrocytes)	0.1	1.5	13-week rat [redacted]	Source: DAR 2003 (addendum) Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Dicofol	Lacrimation, salivation	25	250	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2006 Not true NOAEL, but LOAEL divided by 10	4 presumed MoA (Section 3.3.2)
Dieldrin		0.3		Virtual NOAEL corresponding to the ARfD of 0.003 mg/kg bw/d derived by the PPR Panel and considered as protective for all endpoints.	Source: PPR panel, 2007 Neurotoxicity studies not available	GABA-gated chlorine channel antagonist
Dimethoate	Lacrimation, salivation	20	200	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR	AChE inhibitor
Dinotefuran	–	1,500	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: JMPR 2012	Agonist of nAChR
Emamectin benzoate	Salivation	2.74	27.4	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2008 Not true NOAEL, but LOAEL divided by 10	GABA-gated chloride channel agonist
Endosulfan	Salivation	3	6	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2001 and addendum	GABA-gated chloride channel blocker
Endrin	Convulsions	0.025	0.05	2-year dog [redacted] Administration via diet	Source: EHC 130, 1992 Neurotoxicity studies not available NOAEL for functional alteration of the motor division used as surrogate ADI: 0.0002 (JMPR 1994)	GABA-gated chloride channel antagonist
Esfenvalerate	Salivation	20	80	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2002	Binding to VGSC
Ethephon	Miosis	50	500	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 Not true NOAEL, but LOAEL divided by 10	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Ethion	AChE inhibition (brain)	0.06	0.71	90-day dog [redacted] Administration via diet	Source: JMPR 1990 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Ethoprophos	–	25	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2004	AChE inhibitor
Fenamiphos	Miosis, piloerection	1.52	2.31	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003	AChE inhibitor
Fenitrothion	Miosis, salivation	12.5	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003	AChE inhibitor
Fenpropathrin	–	30	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: JMPR 2012	Binding to VGSC
Fenthion	Autonomic signs (not specified)	1.04	49.05	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996, Addendum Tox 2001	AChE inhibitor
Fenvalerate	–	360	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: JMPR 2012	Binding to VGSC
Fipronil	Miosis	5	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	GABA-gated chloride channel blocker
Flufenacet	Urination	7.5	75	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1998 Not true NOAEL, but LOAEL divided by 10	unknown
Fonofos	Urination	4	7	Acute neurotoxicity rat (author not reported) Administration via gavage	Source: US EPA 1999 No reference values available	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Formetanate	Miosis	1	10	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	AChE inhibitor
Fosthiazate	AChE inhibition (brain)	0.54	5.4	90-day dog [redacted] Administration via capsule	Source: DAR 1998 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Heptachlor	–	69	–	14-day neurotoxicity rat Administration via gavage	Source: CICADS, 2006	GABA-gated chloride channel antagonist
Imidacloprid	–	307	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2005	Agonist of nAChR
Indoxacarb	–	100	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2000	Voltage-dependent sodium channel blocker
Lambda-cyhalothrin	Salivation, lacrimation	1	10	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR Not true NOAEL, but LOAEL divided by 10	Binding to VGSC
Lindane		6			Neurotoxicity studies not available NOAEL of 6 mg/kg bw/d used by JMPR (2002) to establish the ARfD used as surrogate	
Malathion	Salivation	500	1,000	Acute neurotoxicity rats [redacted] Administration via gavage	Source: DAR	AChE inhibitor
Mepiquat	–	1,200	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2005	Activation of nicotinic and muscarinic acetylcholine receptors

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Metaldehyde	Mydriasis	7.5	75	28-day dog [redacted] Administration via diet	Source: DAR Not true NOAEL, but LOAEL divided by 10	Presumed GABA inhibitor
Methamidophos	Urination, lacrimation	1	3	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2000	AChE inhibitor
Methidathion	Lacrimation, salivation	4	5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 1997 addendum	AChE inhibitor
Methiocarb	AChE inhibition (erythrocytes)	0.5	2	28-day rat [redacted] Administration via gavage	Source: DAR 2004 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Methomyl	Lacrimation, salivation	0.75	2	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR	AChE inhibitor
Milbemectin	Salivation (accompanied by vomiting)	3	10	13-week dog [redacted] Administration via capsule	Source: DAR 2001	Glutamate-gated chloride (GluCl) allosteric modulator
Monocrotophos	AChE inhibition (brain, erythrocytes)	0.1	0.3	Single dose rat [redacted] Administration via gavage	Source: JMPR 1995, addendum Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Omethoate (metabolite of dimethoate)	–	5	–	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 (dimethoate)	AChE inhibitor
Oxamyl (IC)	Salivation, urination	0.1	0.75	Acute neurotoxicity [redacted] Administration via gavage	Source: DAR 2003 EFSA (2006)	AChE inhibitor
Oxydemeton-methyl	Salivation	1.88	3.75	90-day dog [redacted] Administration via diet	Source: DAR 2004 Neurotoxicity studies not available NOAEL for AChE inhibition: 0.2 mg/kg bw	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Parathion	AChE inhibition (brain)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: JMPR 1995 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Parathion-methyl	AChE inhibition (erythrocytes)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: DAR 2001 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Permethrin	Urination	250	500	28-day neurotoxicity rat [redacted] Administration via diet	Source: JMPR 1999	
Phenthoate	AChE inhibition (erythrocytes)	0.29	0.87	2-year dog [redacted] Administration via diet	Source: JMPR 1980 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Phosalone	–	60	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2004	AChE inhibitor
Phosmet	Salivation	9	36	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004	AChE inhibitor
Phoxim	Salivation	5	25	90-day dog [redacted]	Source: JECFA 1999 Neurotoxicity studies not available NOAEL for AChE inhibition: 1.3 mg/kg bw/d	AChE inhibitor
Pirimicarb	–	110	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2003	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Pirimiphos-methyl	–	1,500	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2003	AChE inhibitor
Profenofos	Lacrimation, miosis	190	380	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2007	AChE inhibitor
Pyrazophos	AChE inhibition (erythrocytes)	0.05	0.125	2-year dog [redacted] Administration via diet	Source: JMPR 1992 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Pyrethrins	–	200	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2007, 2012	Binding to VGSC
Pyridate	Mydriasis, salivation	60	80	90-day dog [redacted] Administration via capsule	Source: DAR	Unknown
Sulfoxaflor	Lacrimation	75	750	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2012	Nicotinergic AChR partial agonist
Tau-fluvalinate	Salivation	10	60	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2007	Binding to VGSC
Tebuconazole	Salivation	250	500	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2006	Unknown
Tefluthrin	–	10	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2006	Binding to VGSC
Tetramethrin					Information insufficient No reference values available	Binding to VGSC

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Thiacloprid	Mydriasis	2.2	22	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2000, RAR, 2017	Agonist of nAChR
Thiametoxam	Lacrimation	500	1,500	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2002	Agonist of nAChR
Thiodicarb	Salivation	5	20	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003	AChE inhibitor
Thiram	Urination	5	150	Acute neurotoxicity rat [redacted]	Source: DAR 2003	Neurotoxic effect might be due to the metabolite CS_2 (presumed)
Tolclofos-methyl	–	2,000	–	Acute neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: RAR 2016	AChE inhibitor
Tri-allate	Lacrimation, salivation	400	500	Acute neurotoxicity rat [redacted]	Source: DAR 2007	Unknown
Triazophos	AChE inhibition (erythrocytes)	0.012	0.13	1-year dog [redacted] Administration via diet	Source: JMPR 2002 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Trichlorfon	Salivation	50	200	Acute neurotoxicity rat [redacted] Administration via gavage	Source: JMPR 2002	AChE inhibitor
Zeta-cypermethrin	Lacrimation, salivation	50	250	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 and addendum 2005	Binding to VGSC

CAG: cumulative assessment group; AS: active substance; NOEL: no observed effect level; LOEL: lowest observed effect level; NOAEL: no observed adverse effect level; LOAEL: lowest observed adverse effect level; bw: body weight; GABA: gamma-aminobutyric acid; AChE: acetylcholinesterase; VGSC: voltage-gated sodium channel; nAChR: nicotinic acetylcholine receptor; MoA: mode of action.

Table B.4: CAG on brain and/or erythrocyte acetylcholinesterase inhibition: toxicological characterisation of ASs to be considered in acute exposure/risk assessments

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Acephate	AChE inhibition (brain)	2.5	5	Acute single dose rat [redacted] Administration via gavage	Source: JMPR 2002	AChE inhibition
Aldicarb	AChE inhibition (erythrocytes)	0.05	0.1	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996	AChE inhibition
Azinphos-ethyl	AChE inhibition (erythrocytes)	0.0125	0.025	90-day neurotoxicity dog [redacted] Administration via diet	Source: JMPR 1973 Long term NOAEL used as surrogate acute NOAEL	AChE inhibition
Azinphos-methyl	AChE inhibition (erythrocytes)	1	2	acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996, Addendum 6, 2000 (neurotox) ScoFCAH meeting March 2006 considered.	AChE inhibition
Benfuracarb	AChE inhibition (brain, erythrocytes)	1.81	9.4	28-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2004 EFSA (2009)	AChE inhibition
Cadusafos	AChE inhibition (erythrocytes)	0.23	0.46	28-day rat [redacted] Administration via diet	Source: DAR 2004 EFSA (2009)	AChE inhibition
Carbaryl	AChE inhibition (brain, erythrocytes)	1	10	90-day neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 EFSA (2006)	AChE inhibition
Carbofuran	AChE inhibition (brain)	0.015	0.03	Acute neurotoxicity rat [redacted] Administration via gavage	Source: revised DAR 2008	AChE inhibition
Carbosulfan	AChE inhibition (brain, erythrocytes)	0.5	5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: Revised DAR 2009 EFSA (2009)	AChE inhibition
Chlorfenvinphos	AChE inhibition (brain, erythrocytes)	0.15	15	2-year rat (author not reported, JMPR 1994) Administration via diet	Source: JMPR 1994 Long term NOAEL used as surrogate acute NOAEL. Supported by NOAEL of 0.18 mg/kg bw/d for brain AChE inhibition in the 28-day mouse (Tennekes, 1991)	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Chlorpyrifos	AChE inhibition (erythrocytes)	0.5	2.5	Comparative cholinesterase assay rat [redacted] Administration via gavage	Source: EFSA (2014)	AChE inhibition
Chlorpyrifos-methyl	AChE inhibition (erythrocytes)	7.5 (10)	75	Acute neurotoxicity rat [redacted] Administration via gavage	Source: RAR 2017 EC review report 2005 considered	AChE inhibition
Diazinon	AChE inhibition (erythrocytes)	2.5	25	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 EFSA (2006)	AChE inhibition
Dichlorvos	AChE inhibition (erythrocytes)	0.1	1.5	13-week rat [redacted] Administration via gavage	Source: DAR 2003 (addendum) EFSA (2006)	AChE inhibition
Dimethoate	AChE inhibition (erythrocytes)	1	2	Acute neurotoxicity rat [redacted] Administration via diet	Source: DAR 2004 EFSA (2006, 2013)	AChE inhibition
Ethephon	AChE inhibition (erythrocytes)	6	14	28-day cholinesterase inhibition study in dogs [redacted] Administration via diet	Source: DAR 2004 EFSA conclusions on ethephon (2008) considered	AChE inhibition
Ethion	AChE inhibition (brain)	0.06	0.71	90-day dog [redacted] Administration via diet	Source: JMPR 1990 Long term NOAEL used as surrogate acute NOAEL.	AChE inhibition
Ethoprophos	AChE inhibition (erythrocyte)	0.95	2	Single dose study [redacted] Administration via gavage	Source: DAR 2004, addendum EFSA (2006)	AChE inhibition
Fenamiphos	AChE inhibition (erythrocytes)	0.25	0.5	Acute oral dog [redacted] Administration via gavage	Source: DAR 2003 EFSA (2006)	AChE inhibition
Fenitrothion	AChE inhibition (brain, erythrocytes)	1.32	3.99	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2003 EFSA (2006)	AChE inhibition
Fenthion	AChE inhibition (erythrocytes)	1	50	Acute single dose rat study [redacted]	Source: JMPR 1997 JMPR evaluations on fenthion (1997) considered	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Fonofos	AChE inhibition (erythrocytes)	0.2	1	1-year dog [redacted] Administration via capsule	Source: EPA 1999 US evaluation not completed because voluntary withdrawal in 1999 Long term NOAEL used as surrogate acute NOAEL	AChE inhibition
Formetanate	AChE inhibition (brain, erythrocytes)	0.5	2	Acute oral cholinesterase activity study in female rats [redacted] Administration via gavage	Source: DAR 2004 EFSA (2006)	AChE inhibition
Fosthiazate	AChE inhibition	0.54	5.4	90-day dog [redacted] Administration via capsule	Source: DAR 1998 EC review report 2003 considered	AChE inhibition
Malathion	AChE inhibition (brain)	34	340	90-day rat [redacted] Administration via diet	Source: DAR	AChE inhibition
Methamidophos	AChE inhibition (brain, erythrocytes)	0.3	0.7	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2000 EC review report on methamidophos (2006) considered	AChE inhibition
Methidathion	AChE inhibition (brain, erythrocytes)	1	4	Acute neurotoxicity, rat [redacted] Administration via gavage	Source: JMPR 1997 addendum	AChE inhibition
Methiocarb	AChE inhibition (erythrocytes)	0.5	2	28-day rat [redacted] Administration via gavage	Source: DAR 2004 EFSA (2018)	AChE inhibition
Methomyl	AChE inhibition (brain, erythrocytes)	0.25	0.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR EFSA (2006 and 2009)	AChE inhibition
Monocrotophos	AChE inhibition (brain, erythrocytes)	0.1	0.3	Single dose rat [redacted] Administration via gavage	Source: JMPR 1995, addendum	AChE inhibition
Omethoate (metabolite of dimethoate)	AChE inhibition (brain)	0.25	0.35	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 (dimethoate)	AChE inhibition
Oxamyl (IC)	AChE inhibition (brain, erythrocytes)	0.1	0.75	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003 EFSA (2005)	AChE inhibition
Oxydemeton-methyl	AChE inhibition (brain, erythrocytes)	0.2	0.6	14-day rat [redacted] Administration via diet	Source: DAR 2004 EFSA (2006)	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Parathion	AChE inhibition (brain)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: JMPR 1995 Long term NOAEL used as surrogate acute NOAEL	AChE inhibition
Parathion-methyl	AChE inhibition (erythrocytes)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: DAR 2001 Long term NOAEL used as surrogate acute NOAEL	AChE inhibition
Phenthoate	AChE inhibition (erythrocytes)	0.29	0.87	2-year dog [redacted] Administration via diet	Source: JMPR 1980 Long term NOAEL used as surrogate acute NOAEL	AChE inhibition
Phosalone	AChE inhibition (brain, erythrocytes)	25	60	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 EFSA (2006)	AChE inhibition
Phosmet	AChE inhibition (brain, erythrocytes)	4.5	22.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 EFSA (2006)	AChE inhibition
Phoxim	AChE inhibition (erythrocytes)	1.3	5	90-day dog [redacted] Administration via diet	Source: JECFA 1999	AChE inhibition
Pirimicarb	AChE inhibition (brain, erythrocyte)	10	25	1-year dog [redacted] Administration via capsule	Source: DAR 2003	AChE inhibition
Pirimiphos-methyl	AChE inhibition (brain, erythrocytes)	15	150	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003 EFSA (2005)	AChE inhibition
Profenofos	AChE inhibition (erythrocytes)	0.5	25	Acute neurotoxicity rat [redacted] Administration via gavage	JMPR 2007	AChE inhibition
Pyrazophos	AChE inhibition (erythrocytes)	0.05	0.125	2-year dog [redacted] Administration via diet	Source: JMPR 1992 Long term NOAEL used as surrogate acute NOAEL	AChE inhibition
Thiodicarb	AChE inhibition (brain, erythrocytes)	0.5	5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: 2003 EFSA (2005) NOAEL derived from the LOAEL with an UF of 10	AChE inhibition
Tolclofos-methyl	AChE inhibition (erythrocytes)	14	564	9-month mouse [redacted] Administration via diet	Source: DAR 2003 EFSA (2017)	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Triazophos	AChE inhibition (erythrocytes)	0.012	0.13	1-year dog [redacted] Administration via diet	Source: JMPR 2002 Long term NOAEL used as surrogate acute NOAEL	AChE inhibition
Trichlorfon	AChE inhibition (erythrocytes)	10	50	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1998	AChE inhibition

CAG: cumulative assessment group; AS: active substance; NOEL: no observed effect level; LOEL: lowest observed effect level; NOAEL: no observed adverse effect level; LOAEL: lowest observed adverse effect level; bw: body weight; AChE: acetylcholinesterase; MoA: mode of action.

Table B.5: CAG on functional effects on motor division: toxicological characterisation of ASs to be considered in chronic exposure/risk assessments

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Abamectin	Ataxia, tremor	0.25	0.5	18-week dog [redacted] Administration via gavage	Source: DAR 2005 EFSA (2008)	GABA-gated chloride channel agonist
Acephate	–	74.2	–	49-day neurotoxicity study [redacted] Administration via diet Highest tested dose	Source: JMPR 2002	AChE inhibitor
Acetamiprid	Hunched posture	7.1	17.5	2-year rat [redacted] Administration via diet	Source: DAR 2001	Agonist of nAChR
Acrinathrin	Reduced grip strength	0.24	2.4	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2007 Not true NOAEL, but LOAEL divided by 10 EFSA (2013)	Binding to VGSC
Aldicarb	Decreased motor activity, tremor, ataxic gait, decreased hindlimb strength	0.1	0.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996	AChE inhibitor
Alpha-cypermethrin	Ataxia	2.3	6.8	90-day dog [redacted] Administration via diet	Source: DAR 2000 Chronic neurotoxicity studies not available	Binding to VGSC

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Amitraz	Reduced motor activity	0.25	1	2-year dog [redacted] Administration via diet	Source: DAR Neurotoxicity studies not available	Partial agonist of presynaptic α2-adrenergic receptor
Azinphos-ethyl	AChE inhibition (erythrocyte)	0.0125	0.025	90-day dog [redacted] Administration via diet	Source: JMPR 1973 Neurotoxicity studies not available NOAEL for AchE inhibition used as surrogate	AChE inhibitor
Azinphos-methyl	Increased reactivity, tremor, decreased forelimb grip strength	3.23	6.99	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 1996, Addendum 6, 2000 (neurotoxicity)	AChE inhibition
Benfuracarb	Increased motor activity, convulsions; ataxia	2.5	5	2-year dog [redacted] Administration via capsule	Source: DAR 2004	AChE inhibition
Beta-Cyfluthrin	Fore- and hindlimb strength	7.99	26.81	90-day neurotoxicity rat [redacted] Administration via diet	Source: JMPR 2006, DAR 1996	Binding to VGSC
Beta-cypermethrin	Ataxia, tremor, convulsions	1	3	1-year dog [redacted] Administration via capsule	Source: DAR 2013	Binding to VGSC
Bifenthrin	Increased motor activity, tremor	1.5	3.0	1-year dog [redacted] Administration via capsule	Source: DAR	Binding to VGSC
Bromide ion	Muscle strength: increased or decreased muscle tone	7.76	77.6	90-day rat [redacted] Administration via diet	Source: JMPR 1988 Not true NOAEL, but LOAEL divided by 10	Unknown
Cadusafos	AChE inhibition (erythrocytes)	0.045	0.22	2-year rat [redacted] Administration via diet	Source: DAR 2004 The NOAEL of 0.031 mg/kg bw/d in the 90-day neurotoxicity rat (Watt and Freeman, 2001) was disregarded due to the inappropriate dose spacing and incompatibility with the ARfD	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Carbaryl	Tremor, erythrocyte and brain AChE inhibition	1	10	90-day neurotoxicity study [redacted] Administration via gavage	Source: DAR 2004 EFSA (2006)	AChE inhibition
Carbofuran	Reduced grip strength, landing foot splay	34	67.5	90-day neurotoxicity rat [redacted] Administration via diet	Source: Revised DAR 2008	AChE inhibition
Carbosulfan	Decreased motor activity	1.4	78.9	90-day neurotoxicity rat [redacted] Administration via diet	Source: Revised DAR 2009	AChE inhibition
Carbetamide	Ataxia	30	300	90-day dog [redacted] Administration via capsule	Source: DAR 2005	Unknown
Chlorfenvinphos	AChE inhibition (brain, erythrocytes)	0.15	15	2-year rat (author not reported, JMPR 1994) Administration via diet	Source: JMPR 1994 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Chlormequat	Ataxia	10	32	1-year dog [redacted] Administration via diet	Source: DAR 2007 Neurotoxicity studies not available	Partial agonist of muscarinic and nicotinic acetylcholine receptor
Chlorpyrifos	Decreased motor activity	5	15	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 1999	AChE inhibitor
Chlorpyrifos-methyl	Muscle weakness	10	50	90-day dog [redacted] Administration via diet	Source: DAR 1997	AChE inhibition
Clothianidin	Reduced motor activity	35.8	52.3	30-day dog [redacted] Administration via diet	Source: DAR 2003	agonist of nAChR
Cyfluthrin	Ataxia	2.4	11	1-year dog [redacted] Administration via diet	Source: DAR 1996; addendum Chronic neurotoxicity studies not available	Binding to VGSC

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Cypermethrin	Ataxia, tremor	5	15	1-year dog [redacted] Administration via capsule	Source: DAR Chronic neurotoxicity studies not available ADI: 0.05 mg/kg bw per day based on the 2-year rat study (EC Review Report 2005)	Binding to VGSC
Deltamethrin	Ataxia, landing-foot splay, tremor	1	10	1-year dog [redacted] Administration via capsule	Source: DAR EC review report on deltamethrin (2002) considered	Binding to VGSC
Desmedipham	Ataxia, reduced motor activity, tremor	52.5	168	1-year dog [redacted] Administration via diet	Source: DAR 2000	Unknown
Diazinon	Reduced forelimb and hindlimb grip strength	17	177	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2004	AChE inhibition
Dicamba	Reduced motor activity, tremor	50	300	13-week dog [redacted] Administration via capsule	Source: Revised DAR	Unknown
Dichlorvos	AChE inhibition (erythrocytes)	0.008	0.08	2-year dog [redacted]	Source: DAR 2003 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Dicofol	Reduced grip strength	0.3	5.6	90-day rat [redacted] Administration via gavage	Source: DAR 2006	4 presumed MoA (Section 3.3.2)
Dieldrin		0.025			Source: JMPR 1977 Neurotoxicity studies not available NOAEL in dog and rat which served as basis to the JMPR ADI used as surrogate	GABA-gated chlorine channel antagonist
Dimethoate	–	11.25	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Dinotefuran	Hypoactivity	400	3,806	90-day neurotoxicity rat [redacted] Administration via diet	Source: JMPR 2012	agonist of nAChR
Emamectin benzoate (IC)	Tremor	0.25 (0.05)	0.5	1-year dog [redacted] Administration via gavage	Source: DAR 2008 EFSA conclusions on endosulfan (2012) considered Additional UF of 5 due to small dose spacing and steep dose response curve	GABA-gated chloride channel agonist
Endosulfan	Tremor	0.57	2.3	1-year dog [redacted] Administration via diet	Source: DAR 2001 and addendum Chronic neurotoxicity study not available	GABA-gated chloride channel blocker
Endrin	Convulsions	0.025	0.05	2-year dog [redacted] Administration via diet	Source: EHC 130, 1992 Neurotoxicity studies not available ADI: 0.0002 (JMPR 1994)	GABA-gated chloride channel antagonist
Esfenvalerate	Reduced grip strength	3.2	6.4	90-day neurotoxicity rat [redacted] Administration via diet	Source: JMPR 2002	Binding to VGSC
Ethephon	–	400	–	90-day neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2004	AChE inhibitor
Ethion	AChE inhibition (brain)	0.06	0.71	90-day dog [redacted] Administration via diet	Source: JMPR 1990 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Ethoprophos	Reduced grip strength, reduced motor activity, tremor	2.65	27.11	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2004	AChE inhibition
Fenamiphos	Tremor	0.56	1.7	2-year rat [redacted] Administration via diet	Source: DAR 2003	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Fenitrothion	Tremor, reduced grip strength	4.85	17.6	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2003	AChE inhibitor
Fenpropathrin	Increased motor activity, tremor	3.1	7.7	1-year dog [redacted] Administration via diet	Source: JMPR 2012 JMPR conclusions on fenpropathrin considered	Binding to VGSC
Fenpropidin	Ataxia, paresis limbs, reduced motor activity	5	20	1-year dog [redacted] Administration via capsule	Source: DAR	Unknown
Fenpropimorph	Landing-foot splay	0.8	8.5	3-month neurotoxicity rat [redacted] Administration via diet	Source: DAR	Unknown
Fenvalerate	Increased motor activity	37.5	75	6-month dog [redacted]	Not approved. ADI: 0.0125 mg/kg bw per day (EMEA); ADI: 0.02 mg/kg bw per day (JMPR, 1986).	Binding to VGSC
Fenthion	Decreased activity	1.63	8.5	90-day neurotoxicity rat [redacted] Administration via diet	Source: JMPR 1995; Addendum Tox 2001	AChE inhibitor
Fipronil	Convulsions	0.019	0.059	2-year rat [redacted] Administration via capsule	Source: EFSA conclusions on fipronil (2006) considered	GABA-gated chloride channel blocker
Flufenacet	Deficits in stride width	1.14	27	1-year dog [redacted] Administration via diet	Source: DAR 1998	unknown
Fluquinconazole	Ataxia, hunched posture, tremor	0.44	4.77	2-year rat [redacted] Administration via diet	Source: DAR	Unknown
Fonofos	Tremor, abnormal gait	2.5	6.75	90-day neurotoxicity rat (author not reported, US EPA 1999) Administration via diet	Source: US EPA 1999 No reference values available	AChE inhibitor
Formetanate	Ataxia, head shaking	1.8	18	29-day dog [redacted] Administration via diet	Source: DAR 2004	AChE inhibitor
Fosthiazate	Ataxia	0.54	5.4	28-day dog [redacted] Administration via capsule	Source: DAR Neurotoxicity studies not available	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Glufosinate	Ataxia, convulsions, Hyperactivity followed by hypoactivity, tremor	4.5	8.4	1-year dog [redacted] Administration via diet	Source: DAR	Unknown
Heptachlor	Increased motor activity, convulsions	0.5	5	6-month rat [redacted] Administration via gavage	Source: JMPR 1970 Not true NOAEL, but LAOEL divided by 10. ADI: 0.0001 mg/kg (JMPR 1994) 14-day neurotoxicity rat [redacted]: No effect at 69 mg/kg bw/d (highest tested dose) 90-day neurotoxicity study not available.	GABA-gated chloride channel antagonist
Imidacloprid	Tremor	23.5	45.4	90-day dog [redacted] Administration via diet	Source: DAR 2005	Agonist of nAChR
Indoxacarb	Ataxia, hunched posture	2.6	14	18-month mouse [redacted] Administration via diet	Source: DAR 2000	Voltage-dependent sodium channel blocker
Isoxaflutole	Limited use of hindlimbs	20	500	2-year rat [redacted] Administration via diet	Source: DAR 1997	Unknown
Lambda-Cyhalothrin	Ataxia, convulsions, tremor	0.5	3.5	1-year dog [redacted] Administration via capsule	Source: DAR 1996 Chronic neurotoxicity studies not available	Binding to VGSC
Lindane	Increased motor activity, convulsions	6	24.3	2-year rat [redacted] Administration via diet	Source: JMPR 2002 Chronic neurotoxicity studies not available	GABA-gated chloride channel antagonist
Lufenuron	Convulsions	1.9	20	2-year rat [redacted] Administration via diet	Source: DAR 2006	Unknown
Malathion	–	1,486	–	90-day neurotoxicity are [redacted] Administration via diet Highest tested dose	Source: DAR	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Mancozeb	Paralysis	49	328	3-month rat [redacted]	Source: DAR 2000	presumed Neurotoxic effect might be due to the metabolite CS_2
Maneb	Paresis limbs	75	200	1-year dog [redacted]	Source: DAR 2000	presumed Neurotoxic effect might be due to the metabolite CS_2
Mepiquat	Convulsions, lateral position	32	95	3-month dog [redacted] Administration via diet	Source: DAR	Activation of nicotinic and muscarinic acetylcholine receptors
Methamidophos	AChE inhibition (brain, erythrocytes)	0.1	0.22	2-year rat [redacted] Administration via diet	Source: DAR 2000 EC review report on methamidophos (2006) considered 90-day neurotoxicity rat [redacted] with NOAEL at 0.067 mg/kg bw disregarded for reason of compatibility with the ADI	AChE inhibitor
Methidathion	Increased motor activity: tremor, hyperactivity	0.16	0.8	2-year rat [redacted] Administration via diet	Source: JMPR 1992 Chronic neurotoxicity studies not available	AChE inhibitor
Methiocarb	Muscle weakness, tremor	2.2	8.6	2-year dog [redacted] Administration via diet	Source: DAR 2004 Neurotoxicity studies not available	AChE inhibitor
Methomyl	Reduced fore- and hindlimb grip strength	9	95	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Metiram	Reduced grip strength	25.4	81.4	3-month rat [redacted]	Source: DAR 2000	Presumed Neurotoxic effect might be due to the metabolite CS_2
Milbemectin	–	59	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: RAR 2017	Glutamate-gated chloride (GluCl) allosteric modulator
Molinate	Ataxia	1.8	13	2-year rat [redacted] Administration via diet	Source: DAR	Presumed Inhibition of ALDH by molinate sulfone
Monocrotophos	AChE inhibition (brain, erythrocytes)	0.005	0.05	2-year rat [redacted] Administration via diet	Source: JMPR 1991 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Omethoate (metabolite of dimethoate)	Increased motor activity: tremor	0.3	2.9	2-year rat [redacted] Administration via drinking water	Source: DAR 2004 Chronic neurotoxicity studies not available	AChE inhibitor
Oxamyl	Ataxia, hunched posture, ptosis	1.69	15.3	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2003	AChE inhibitor
Oxasulfuron	Ataxia	1.3	11	1-year dog [redacted] Administration via diet	Source: DAR	Unknown
Oxydemeton-methyl	AChE inhibition (brain, erythrocytes)	0.027	0.224	2-year rat [redacted] Administration via diet	Source: DAR 2004 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Parathion	Tremor, abnormal gait	0.25	2.5	2-year rat [redacted] Administration via diet	Source: ECCO 2001, JMPR 1995 Neurotoxicity studies not available NOAEL for AChE inhibition: 0.25 mg/kg bw/d	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Parathion-methyl	Tremor, abnormal gait	0.25	2.5	2-year rat [redacted] Administration via diet	Source: DAR 2001 Neurotoxicity studies not available NOAEL for AChE inhibition: 0.25 mg/kg bw/d	AChE inhibitor
Permethrin	Increased motor activity, tremor	40	100	2-year rat [redacted] Administration via diet	Source: JMPR 1999	Binding to VGSC
Phenthoate	AChE inhibition (erythrocytes)	0.29	0.87	2-year dog [redacted] Administration via diet	Source: JMPR 1980 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Phosalone	Reduced grip strength	11.5	45.9	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2004	AChE inhibitor
Phosmet	–	9.4	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2004	AChE inhibitor
Phoxim	AChE inhibition (erythrocytes)	0.1	0.38	2-year dog [redacted] Administration via diet	Source: JECFA 1999 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Pirimicarb	Tremor	3.5	10	1-year dog [redacted] Administration via capsule	Source: DAR EFSA conclusions in pirimicarb (2005) considered	AChE inhibitor
Pirimiphos-methyl	Hunched posture	9	36	2-year mouse [redacted] Administration via diet	Source: DAR	AChE inhibitor
Profenofos	–	36	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2007	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Propineb	Ataxia (Hind-limb wheelbarrowing)	4.3	41	90-day dog [redacted] Administration via diet	Source: DAR 1996	Presumed Neurotoxic effect might be due to the metabolite CS2
Pyrazophos	Muscle strength: increased or decreased muscle tone, weakness; coordination: abnormal gait	0.45	8	6-month dog [redacted] Administration via diet	Source: DAR 1998 Neurotoxicity studies not available NOAEL for AChE inhibition: 0.05 mg/kg bw/d	AChE inhibitor
Pyrethrins	Ataxia, paresis limbs, tremor	30	86	56-day dog [redacted] Administration via diet	Source: DAR 2007 Chronic neurotoxicity studies not available	Binding to VGSC
Sulfoxaflor	–	94.9	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2012	Nicotinergic AChR partial agonist
tau-Fluvalinate	Choreoathetosis (Ruffling of body, pawing), transient hyperactivity followed by hypoactivity	0.5	1	2-year rat [redacted] Administration via gavage	Source: DAR EFSA (2018) Chronic neurotoxicity studies not available	Binding to VGSC
Tebuconazole	Reduced motor activity	100	300	4-week rat [redacted] Administration via gavage	Source: DAR 2006	Unknown
Tefluthrin	ataxia	0.5	2	1-year dog [redacted] Administration via capsule	Source: DAR EFSA (2010)	Binding to VGSC
Tembotrione	Reduced motor activity: hypoactivity	26.7	111	90-day dog [redacted] Administration via diet	Source: DAR 2012	Unknown
Tetraconazole	Hunched posture	17	65	28-day rat [redacted] Administration via diet	Source: DAR 2005	Unknown
Tetramethrin	Increased motor activity: tremor	31	63	6-month dog [redacted] Administration via diet	Source: WHO 1990 Neurotoxicity studies not available No reference values available	Binding to VGSC

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Thiacloprid	–	101	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2017	Agonist of nAChR
Thiametoxam	–	95.4	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2002	Agonist of nAChR
Thiodicarb	Increased motor activity	12.8	38.3	1-year dog [redacted] Administration via diet	Source: DAR 2003	AChE inhibitor
Thiram	Dragging of hind feet and tail, paralysis	5.3	20	18-month rat [redacted] Administration via diet	Source: DAR 1997 and addenda	Presumed Neurotoxic effect might be due to the metabolite CS_2
Tolclofos-methyl	–	735.7	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: RAR 2016	AChE inhibitor
Triadimefon	Increased motor activity: hyperactivity	3.4	54.6	90-day neurotoxicity rat [redacted] Administration via diet	Source: JMPR 2004	Inhibition of dopamine transporter (presumed)
Triadimenol (a metabolite of Triadimefon)	Increased motor activity	3.4	45	3-month neurotoxicity rat [redacted] Administration via diet	Source: DAR	Inhibition of dopamine transporter (presumed)
Tri-allate	Landing-foot splay, reduced grip strength	32.9	128.8	3-month neurotoxicity rat [redacted] Administration via diet	Source: DAR	Unknown
Triazophos	AChE inhibition (erythrocytes)	0.012	0.13	1-year dog [redacted] Administration via diet	Source: JMPR 2002 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Trichlorfon	–	168	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR addendum, 2005	AChE inhibitor
Zeta-Cypermethrin	Landing-foot splay, reduced motor activity	5	26	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR	Known Binding to VGSC

CAG: cumulative assessment group; AS: active substance; NOEL: no observed effect level; LOEL: lowest observed effect level; NOAEL: no observed adverse effect level; LOAEL: lowest observed adverse effect level; bw: body weight; GABA: gamma-aminobutyric acid; AChE: acetylcholinesterase; VGSC: voltage-gated sodium channel; nAChR: nicotinic acetylcholine receptor; MoA: mode of action.

Table B.6: CAG on functional effects on sensory division: toxicological characterisation of ASs to be considered in chronic exposure/risk assessments

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Abamectin	Decreased pupil reactivity	0.25	0.5	1-year dog [redacted] Administration via diet	Source: DAR 2005 EFSA (2008)	GABA-gated chloride channel agonist
Acephate	–	74.2	–	49-day neurotoxicity study [redacted] Administration via diet Highest tested dose	Source: JMPR 2002	AChE inhibitor
Acetamiprid	–	118	–	90-day neurotoxicity study [redacted] Administration via diet Highest tested dose	Source: DAR 2001	Agonist of nAChR
Acrinathrin	–	62.6	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2007	Binding to VGSC
Aldicarb	Decreased tail pinch response	0.1	0.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Alpha-cypermethrin		1.5			Chronic neurotoxicity studies not available NOAEL used to derive the ADI (1-year dog, EC review report on alpha-cypermethrin 2004) used as surrogate	Binding to VGSC
Amitraz	Hyperreactivity	12	50	90-day rat [redacted] Administration via gavage	Source: DAR Neurotoxicity studies not available	Partial agonist of presynaptic α2-adrenergic receptor
Azinphos-ethyl	AChE inhibition (erythrocyte)	0.0125	0.025	90-day dog [redacted] Administration via diet	Source: JMPR 1973 Neurotoxicity studies not available NOAEL for AchE inhibition used as surrogate	AChE inhibitor
Azinphos-methyl	Abnormal righting reflex	3.23	6.99	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 1996, Addendum 6, 2000 (neurotoxicity)	AChE inhibition
Benfuracarb	–	45.8	–	28-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2004	AChE inhibition
Beta-Cyfluthrin	–	26.81	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2006, DAR 1996	Binding to VGSC
Beta-cypermethrin	Hyperreactivity	0.82	8.2	90-day rat [redacted] Administration via diet	Source: DAR 2013 Not true NOAEL, but LOAEL divided by 10	Binding to VGSC
Bifenthrin	–	11.8	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	Binding to VGSC

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Cadusafos	–	27	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2004	AChE inhibition
Carbaryl	Decreased pupil size	10	30	90-day neurotoxicity study [redacted] Administration via gavage	Source: DAR 2004	AChE inhibition
Carbofuran	–	67.5	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: Revised DAR 2008	AChE inhibition
Carbosulfan	–	130.7	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: Revised DAR 2009	AChE inhibition
Chlorfenvinphos	AChE inhibition (brain, erythrocytes)	0.15	15	2-year rat (author not reported, JMPR 1994) Administration via diet	Source: JMPR 1994 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Chlormequat	Diminished reflex response	50	62.5	90-day dog [redacted] Administration via diet	Source: DAR 2007 Neurotoxicity studies not available	Known Partial agonist of muscarinic and nicotinic acetylcholine receptor
Chlorpyrifos	–	15	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 1999	AChE inhibition
Chlorpyrifos-methyl	–	250	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 1997	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Clothianidin	–	177	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2003	agonist of nAChR
Cyfluthrin		0.3			Chronic neurotoxicity studies not available NOAEL used to derive the ADI (pharmacological study in mice, EC review report on cyfluthrin 2002) used as surrogate	Binding to VGSC
Cymoxanil	Hyperreactivity	30	90	2-year rat [redacted] Administration via diet	Source: DAR	Unknown
Cypermethrin	Hypersensitivity to noise	5	15	1-year dog [redacted] Administration via capsule	Source: DAR Chronic neurotoxicity studies not available ADI: 0.05 mg/kg bw per day based on the 2-year rat study (EC Review Report 2005)	Binding to VGSC
Deltamethrin	Hypersensitivity to noise	4	14	90-day neurotoxicity rat [redacted]	Source: DAR addendum	Binding to VGSC
Diazinon	–	177	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2004	AChE inhibition
Dichlorvos	AChE inhibition (erythrocytes)	0.008	0.08	2-year dog [redacted]	Source: DAR 2003 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Dieldrin	Hyperreactivity	0.05	0.25	2-year rat [redacted] Administration via diet	Source: JMPR 1977 ADI (PTDI): 0.0001 mg/kg bw per day (JMPR 1994).	GABA-gated chlorine channel antagonist
Dimethoate	–	11.25	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Dinotefuran	–	3,413	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2012	Agonist of nAChR
Emamectin benzoate	Hyperreactivity	0.5 (0.1)	0.75	1-year dog [redacted] Administration via gavage	Source: DAR 2008 EFSA conclusions on emamectin benzoate (2012) considered Additional UF of 5 due to small dose spacing and steep dose response curve	GABA-gated chloride channel agonist
Endosulfan	Exaggerated auditory response (startle reflex)	0.57	2.3	1-year dog [redacted] Administration via diet	Source: DAR 2001 and addendum Chronic neurotoxicity study not available	GABA-gated chloride channel blocker
Endrin (IC)	Hyperreactivity	0.05	1	2-year rat [redacted] Administration via diet	Source: EHC 130, 1992 Neurotoxicity studies not available ADI (PTDI): 0.0002 (JMPR 1994)	GABA-gated chloride channel antagonist
Esfenvalerate	–	20.1	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2002	Binding to VGSC
Ethephon	–	400	–	90-day neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2004	AChE inhibitor
Ethion	AChE inhibition (brain)	0.06	0.71	90-day dog [redacted] Administration via diet	Source: JMPR 1990 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Ethoprophos	Negative air drop, pupillary responses, decreased analgesic reflex	2.65	27.11	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2004	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Fenamiphos	–	3.1	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2003	AChE inhibition
Fenitrothion	–	13.8	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2003	AChE inhibitor
Fenpropathrin	Uncoordinated righting reflex	15	50	90-day neurotoxicity rat [redacted] Administration via diet	Source: JMPR 2012	Binding to VGSC
Fenpropimorph	Retarded pupillary reflex	7.1	71	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR	Unknown
Fenthion	–	8.5	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 1995; Addendum Tox 2001	AChE inhibitor
Fenvalerate	Hyperreactivity	15	50	18-month mouse [redacted] Administration via diet	Source: JMPR 2012	Binding to VGSC
Fipronil	Hyperreactivity	0.2	2	1-year dog [redacted] Administration via capsule	Source: DAR 2004	GABA-gated chloride channel blocker
Flufenacet	Hypo-reactivity, reduced reaction to movement and sound, hyperreactivity	27	59	1-year dog [redacted] Administration via diet	Source: DAR 1998	Unknown
Fonofos	Decreased reactivity patellar reflex	2.5	6.75	90-day neurotoxicity rat (author not reported, US EPA 1999) Administration via diet	Source: US EPA 1999 No reference values available	AChE inhibitor
Formetanate	–	18.4	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2017	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Fosthiazate	AChE inhibition	0.42	2.36	2-year rat [redacted]	Source: DAR Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Glufosinate	Decrease in alertness and/or startle response	52.1	521	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR Not true NOAEL, but LOAEL divided by 10	Unknown
Heptachlor	Hyperreactivity	2	7	14-day neurotoxicity rat [redacted] Administration via gavage	Source: CICADS, 2006 90-day neurotoxicity study not available. Poor data. ADI (PTDI): 0.0001 mg/kg bw/d (JMPR 1994).	GABA-gated chloride channel antagonist
Imidacloprid	–	196	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2005	Agonist of nAChR
Indoxacarb	Hyperreactivity	2.6	14	18-month mouse [redacted] Administration via diet	Source: DAR 2000	Voltage-dependent sodium channel blocker
Lambda-cyhalothrin	Ataxia, convulsions, tremor	0.5	–	1-year dog [redacted] Administration via capsule	Chronic neurotoxicity studies not available NOAEL for functional alteration of the motor division used as surrogate ADI: 0.0025 mg/kg bw/d, based on multigeneration rat study (EFSA 2014)	Binding to VGSC
Lindane		0.47			Chronic neurotoxicity studies not available NOAEL used to derive the ADI (2-year rat, JMPR 2002) used as surrogate	GABA-gated chloride channel antagonist

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Malathion	–	1,486	–	90-day neurotoxicity are [redacted] Administration via diet Highest tested dose	Source: DAR	AChE inhibitor
Metaldehyde	No reaction to noise	30	90	1-year dog [redacted] Administration via diet	Source: DAR	Presumed GABA inhibitor
Methamidophos	–	4.26	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2000	AChE inhibitor
Mepiquat	–	517	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	Activation of nicotinic and muscarinic acetylcholine receptors
Methidathion	Hyperreactivity	0.16	1.72	2-year rat [redacted] Administration via diet	Source: JMPR 1992 Chronic neurotoxicity studies not available	AChE inhibition
Methiocarb	AChE inhibition (erythrocytes)	1.32	6.46	90-day dog [redacted] Administration via diet	Source: DAR 2004 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Methomyl	–	95	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	AChE inhibition
Milbemectin	–	59	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: RAR 2017	Glutamate-gated chloride (GluCl) allosteric modulator
Molinate	Patellar hyperreflexia, paraesthesia, proprioception deficit	10	50	1-year dog [redacted] Administration via capsule	Source: DAR	Presumed Inhibition of ALDH by molinate sulfone

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Monocrotophos	AChE inhibition (brain, erythrocytes)	0.005	0.05	2-year rat [redacted] Administration via diet	Source: JMPR 1991 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Omethoate (metabolite of dimethoate)	AChE inhibition (erythrocytes)	0.027	0.04	2-year rat [redacted]; supplementary 32-week rat [redacted]	Source: DAR 2004 (dimethoate) Chronic neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Oxamyl	Hyperreactivity, absent pupillary response	1.69	15.3	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2003	AChE inhibitor
Oxasulfuron	Hindlimb flexor reflex	83	425	2-year carcinogenicity rat [redacted] Administration via diet	Source: DAR	Unknown
Oxydemeton-methyl	AChE inhibition (brain, erythrocytes)	0.027	0.224	2-year rat [redacted] Administration via diet	Source: DAR 2004 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Parathion	AChE inhibition (brain)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: JMPR 1995 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Parathion-methyl	AChE inhibition (erythrocytes)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: DAR 2001 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Permethrin	Hypersensitivity	100	250	28-day neurotoxicity rat [redacted] Administration via diet	Source: JMPR 1999	Binding to VGSC

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Phenthoate	AChE inhibition (erythrocytes)	0.29	0.87	2-year dog [redacted] Administration via diet	Source: JMPR 1980 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Phosalone	–	45.9	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2004	AChE inhibitor
Phosmet	–	9.4	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2004	AChE inhibitor
Phoxim	AChE inhibition (erythrocytes)	0.1	0.38	2-year dog [redacted] Administration via diet	Source: JECFA 1999 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Pirimicarb	–	77.1	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	AChE inhibition
Pirimiphos-methyl	–	21.1	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	AChE inhibition
Profenofos	–	36	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2007	AChE inhibitor
Propineb	Sensory changes (proprioceptive deficit)	4.3	41.4	90-day dog [redacted] Administration via diet	Source: DAR 1996	Presumed Neurotoxic effect might be due to the metabolite CS2

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Pyrazophos	Decreased pupil reactivity	0.45	8	6-month dog [redacted] Administration via diet	Source: DAR 1998 Neurotoxicity studies not available NOAEL for AChE inhibition: 0.05 mg/kg bw/d	AChE inhibitor
Pyrethrins		4			Chronic neurotoxicity studies not available NOAEL used to derive the ADI (2-year rat, EFSA 2013) used as surrogate	Binding to VGSC
Sulcotrione	Decrease in proprioception, increased patellar reflex	300	600	16-week dog [redacted]; 90-day dog [redacted]	Source: DAR	Unknown
Sulfoxaflor	–	94.9	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2012	Nicotinergic AChR partial agonist
tau-Fluvalinate	Decreased responsiveness to sensory stimuli, increase in click response	2	6	8-day neurotoxicity rat [redacted] Administration via gavage	Source: DAR Chronic neurotoxicity studies not available ADI: 0.005 mg/kg bw/d, 2-year rat (EFSA, 2018)	Binding to VGSC
Tefluthrin	Increased response to sound	1.5	5.9	2-year carcinogenicity rat [redacted] Administration via diet	Source: DAR	Binding to VGSC
Tetramethrin					Neurotoxicity studies not available No reference values available	Binding to VGSC
Thiacloprid	–	101	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2017	Agonist of nAChR
Thiametoxam	–	95.4	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2002	Agonist of nAChR

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Thiodicarb	–	46	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2003	AChE inhibition
Tolclofos-methyl	–	735.7	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: RAR 2016	AChE inhibitor
Tri-allate	Increased alertness, impaired righting reflex	33	129	3-month neurotoxicity rat [redacted] Administration via diet	Source: DAR	Unknown
Triazophos	AChE inhibition (erythrocytes)	0.012	0.13	1-year dog [redacted] Administration via diet	Source: JMPR 2002 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Trichlorfon	–	168	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR addendum, 2005	AChE inhibitor
Zeta-cypermethrin	–	47	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	Known Binding to VGSC

CAG: cumulative assessment group; AS: active substance; NOEL: no observed effect level; LOEL: lowest observed effect level; NOAEL: no observed adverse effect level; LOAEL: lowest observed adverse effect level; bw: body weight; GABA: gamma-aminobutyric acid; AChE: acetylcholinesterase; VGSC: voltage-gated sodium channel; nAChR: nicotinic acetylcholine receptor; MoA: mode of action.

Table B.7: CAG on functional effects on autonomic division: toxicological characterisation of ASs to be considered in chronic exposure/risk assessments

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
2,4-D	Urination	75	150	1-year neurotoxicity rat [redacted] Administration via diet	Source: DAR 1997	Unknown
Abamectin	Salivation, mydriasis	0.25	0.5	18-week dog [redacted] Administration via gavage	Source: DAR 2005 EFSA conclusions on abamectin (2008) considered	GABA-gated chloride channel agonist
Acephate	–	74.2	–	49-day neurotoxicity study [redacted] Administration via diet Highest tested dose	Source: JMPR 2002	AChE inhibitor
Acetamiprid	–	118	–	90-day neurotoxicity study [redacted] Administration via diet Highest tested dose	Source: DAR 2001	Agonist of nAChR
Acrinathrin	Salivation	5	10	28-day rat [redacted] Administration via gavage	Source: DAR 2007	Binding to VGSC
Aldicarb	Salivation, lacrimation	0.1	0.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996	AChE inhibitor
Alpha-cypermethrin		1.5			Chronic neurotoxicity studies not available NOAEL used to derive the ADI (1-year dog, EC review report on alpha-cypermethrin 2004) used as surrogate	Binding to VGSC
Amitraz		0.3			Neurotoxicity studies not available NOAEL used to derive the ADI (SCoFCAH, 2003) used as surrogate	Partial agonist of presynaptic α2-adrenergic receptor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Azinphos-ethyl	AChE inhibition (erythrocyte)	0.0125	0.025	90-day dog [redacted] Administration via diet	Source: JMPR 1973 Neurotoxicity studies not available NOAEL for AchE inhibition used as surrogate	AChE inhibitor
Azinphos-methyl	–	6.99	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 1996, Addendum 6, 2000 (neurotoxicity)	AChE inhibition
Benfuracarb	Salivation	2.5	5	2-year dog [redacted] Administration via capsule	Source: DAR 2004	AChE inhibition
Beta-cyfluthrin	–	26.81	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2006, DAR 1996	Binding to VGSC
Beta-cypermethrin	Salivation	20	100	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2013 Chronic neurotoxicity studies not available	Binding to VGSC
Bifenthrin	–	11.8	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	Binding to VGSC
Cadusafos	–	27	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2004	AChE inhibition
Carbaryl	Salivation	10	30	90-day neurotoxicity study [redacted] Administration via gavage	Source: DAR 2004	AChE inhibition
Carbetamide	Salivation	150	300	28-day dog [redacted] Administration via capsule	Source: DAR 2005	Unknown
Carbofuran	–	67.5	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: Revised DAR 2008	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Carbosulfan	–	130.7	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: Revised DAR 2009	AChE inhibition
Chlorfenvinphos	AChE inhibition (brain, erythrocytes)	0.15	15	2-year rat (author not reported, JMPR 1994) Administration via diet	Source: JMPR 1994 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Chlormequat	Salivation	5	10	1-year dog [redacted] Administration via diet	Source: DAR 2007 Neurotoxicity studies not available	Partial agonist of muscarinic and nicotinic acetylcholine receptor
Chlorpyrifos	–	15	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 1999	AChE inhibition
Chlorpyrifos-methyl	–	250	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 1997	AChE inhibition
Clothianidin	Salivation	19.3	40.9	90-day dog [redacted] Administration via diet	Source: DAR 2003	Agonist of nAChR
Cyfluthin		0.3			Chronic neurotoxicity studies not available NOAEL used to derive the ADI (pharmacological study in mice, EC review report on cyfluthrin 2002) used as surrogate	Binding to VGSC
Cypermethrin	Salivation	6	20	1-year dog [redacted] Administration via diet	Source: DAR zeta-cypermethrin Chronic neurotoxicity studies not available ADI: 0.05 mg/kg bw per day based on the 2-year rat study (EC Review Report 2005)	Binding to VGSC

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Deltamethrin	Mydriasis	1	2.5	90-day dog [redacted] Administration via diet	Source: DAR EC review report on deltamethrin considered	Binding to VGSC
Diazinon	–	177	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2004	AChE inhibition
Dicamba	Salivation	50	300	13-week dog [redacted] Administration via capsule	Source: Revised DAR year	Unknown
Dichlorvos	AChE inhibition (erythrocytes)	0.008	0.08	2-year dog [redacted]	Source: DAR 2003 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Dicofol	Salivation	3.31	9.78	90-day dog [redacted] Administration via diet	Source: DAR 2006	Unknown
Dieldrin		0.025			Source: JMPR 1977 Neurotoxicity studies not available NOAEL in dog and rat which served as basis to the JMPR ADI used as surrogate	GABA-gated chlorine channel antagonist
Dimethoate	–	11.25	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	AChE inhibitor
Dinotefuran	–	3,413	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2012	Agonist of nAChR

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Emamectin benzoate	Mydriasis	0.5 (0.1)	0.75	1-year dog [redacted] Administration via gavage	Source: DAR 2008 EFSA conclusions on emamectin benzoate (2012) considered Additional UF of 5 due to small dose spacing and steep dose response curve	GABA-gated chloride channel agonist
Endosulfan		0.6			Source: JMPR 1998 Chronic neurotoxicity study not available NOAEL in 2-year rat and 1-year dog which served as basis to the JMPR ADI used as surrogate	
Endrin		0.02			Neurotoxicity studies not available NOAEL based on the JMPR ADI (PTDI) of 0.0002 mg/kg bw/d (JMPR 1994)	GABA-gated chloride channel antagonist
Esfenvalerate	–	20.1	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2002	Binding to VGSC
Ethephon	–	400	–	90-day neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR 2004	AChE inhibitor
Ethion	AChE inhibition (brain)	0.06	0.71	90-day dog [redacted] Administration via diet	Source: JMPR 1990 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Ethoprophos	Salivation, lacrimation	2.65	27.11	90-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2004	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Fenamiphos	–	3.1	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2003	AChE inhibition
Fenitrothion	–	12.5	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2003	AChE inhibitor
Fenpropathrin	–	38	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2012	Binding to VGSC
Fenthion	–	8.5	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 1995; Addendum Tox 2001	AChE inhibitor
Fenvalerate	–	300	–	14-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2012 90-day neurotoxicity study not available	Binding to VGSC
Fipronil	–	8.9	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2004	GABA-gated chloride channel blocker
Fluquinconazole	Piloerection	1.73	8.81	28-day rat [redacted] Administration via diet	Source: DAR	Unknown
Fonofos	Urination	2.5	6.75	90-day neurotoxicity rat (author not reported, US EPA 1999) Administration via diet	Source: US EPA 1999 No reference values available	AChE inhibitor
Formetanate	–	18.4	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2017	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Fosthiazate	AChE inhibition	0.42	2.36	2-year rat [redacted]	Source: DAR Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Glufosinate	Trismus salivation	4.5	8.4	1-year dog [redacted] Administration via diet	Source: DAR	Unknown
Heptachlor	–	69	–	14-day neurotoxicity rat [redacted] Administration via gavage	Source: CICADS, 2006 90-day neurotoxicity study not available. Poor data. ADI (PTDI): 0.0001 mg/kg bw/d (JMPR 1994).	GABA-gated chloride channel antagonist
Imidacloprid	–	196	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2005	agonist of nAChR
Indoxacarb	Urination	2.6	14	18-month mouse [redacted] Administration via diet	Source: DAR 2000	Voltage-dependent sodium channel blocker
Lambda-Cyhalothrin	Piloerection	1.8	9.2	2-year mouse [redacted] Administration via diet	Source: DAR Chronic neurotoxicity studies not available ADI: 0.0025 mg/kg bw/d, based on multigeneration rat study (EFSA 2014)	Binding to VGSC
Lindane		0.47			Chronic neurotoxicity studies not available NOAEL used to derive the ADI (2-year rat, JMPR 2002) used as surrogate	GABA-gated chloride channel antagonist
Lufenuron	Salivation	7	30	1-year dog [redacted] Administration via diet	Source: DAR 2006	Unknown

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Malathion	–	1,486	–	90-day neurotoxicity are [redacted] Administration via diet Highest tested dose	Source: DAR	AChE inhibitor
Mepiquat	–	517	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	Activation of nicotinic and muscarinic acetylcholine receptors
Metaldehyde	Salivation	30	90	1-year dog [redacted] Administration via diet	Source: DAR	Presumed GABA inhibitor
Metamitron	Urination	10	50	28-day rat [redacted] Administration via diet	Source: DAR 2007	Unknown
Methamidophos (IC)	AChE inhibition (brain, erythrocytes)	0.1	0.22	2-year rat [redacted] Administration via diet	Source: DAR 2000 EC review report on methamidophos (2006) considered 90-day neurotoxicity rat [redacted] with NOAEL at 0.067 mg/kg bw disregarded for reason of compatibility with the ADI.	AChE inhibitor
Methidathion	AChE inhibition (brain, erythrocytes)	0.16	0.8	2-year rat [redacted] Administration via diet	Source: JMPR 1992 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Methiocarb	AChE inhibition (erythrocytes)	1.32	6.46	90-day dog [redacted] Administration via diet	Source: DAR 2004 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Methomyl	–	95	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Milbemectin		59	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: RAR 2017	Glutamate-gated chloride (GluCl) allosteric modulator
Molinate	Salivation	1	10	1-year dog [redacted] Administration via capsule	Source: DAR	Presumed Inhibition of ALDH by molinate sulfone
Monocrotophos	Salivation	0.4	2.5	2-year dog [redacted] Administration via diet	Source: JMPR 1972 Neurotoxicity studies not available ADI: 0.0006 mg/kg bw per day (JMPR 1995)	AChE inhibition
Omethoate (metabolite of dimethoate)	AChE inhibition (erythrocytes)	0.027	0.04	2-year rat [redacted]; supplementary 32-week rat [redacted]	Source: DAR 2004 (dimethoate) Chronic neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Oxamyl	–	15.3	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2003	AChE inhibitor
Oxydemeton-methyl	AChE inhibition (brain, erythrocytes)	0.027	0.224	2-year rat [redacted] Administration via diet	Source: DAR 2004 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Parathion	AChE inhibition (brain)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: JMPR 1995 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Parathion-methyl	AChE inhibition (erythrocytes)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: DAR 2001 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Permethrin	piloerection	100	250	28-day neurotoxicity rat [redacted] Administration via diet	Source: JMPR 1999 90-day neurotoxicity studies not available ADI: 0.05 mg/kg bw per day (JMPR 1999)	Binding to VGSC
Phenthoate	AChE inhibition (erythrocytes)	0.29	0.87	2-year dog [redacted] Administration via diet	Source: JMPR 1980 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Phosalone	–	45.9	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2004	AChE inhibitor
Phosmet	–	9.4	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2004	AChE inhibitor
Phoxim	AChE inhibition (erythrocytes)	0.1	0.38	2-year dog [redacted] Administration via diet	Source: JECFA 1999 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Pirimicarb	Urination (1) salivation (2)	10	25(1)/35(2)	90-day dog [redacted] (1), 1-year dog [redacted] (2)	Source: DAR	AChE inhibitor
Pirimifos-methyl	–	21.1	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Profenofos	–	36	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: JMPR 2007	AChE inhibitor
Pyrazophos	AChE inhibition (erythrocytes)	0.05	0.125	2-year dog [redacted] Administration via diet	Source: JMPR 1992 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibition
Pyrethrins		4			Chronic neurotoxicity studies not available NOAEL used to derive the ADI (2-year rat, EFSA 2013) used as surrogate	Binding to VGSC
Sulfoxaflor	–	94.9	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2012	Nicotinergic AChR partial agonist
tau-Fluvalinate	Salivation, lacrimation	0.5	1	2-year rat [redacted] Administration via gavage	Source: DAR Chronic neurotoxicity studies not available EFSA conclusion on tau-fluvalinate (2018) considered	Binding to VGSC
Tefluthrin	–	26.6	–	90-day neurotoxicity rat [redacted] Administration via gavage Highest tested dose	Source: DAR	Binding to VGSC
Tetramethrin					Neurotoxicity studies not available No reference values available	Binding to VGSC
Thiacloprid	–	101	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: RAR 2017	Agonist of nAChR

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Thiametoxam	–	95.4	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2002	Agonist of nAChR
Thiodicarb	–	46	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR 2003	AChE inhibition
Tolclofos-methyl	–	735.7	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: RAR 2016	AChE inhibitor
Triadimenol (metabolite of Triadimefon)	Piloerection	40	209	90-day rat [redacted] Administration via diet	Source DAR	Presumed Inhibition of dopamine transporter
Tri-allate	Lacrimation	33	129	3-month neurotoxicity rat [redacted] Administration via diet	Source: DAR	Unknown
Triazophos	AChE inhibition (erythrocytes)	0.012	0.13	1-year dog [redacted]) Administration via diet	Source: JMPR 2002 Neurotoxicity studies not available NOAEL for AChE inhibition used as surrogate	AChE inhibitor
Trichlorfon	–	168	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR addendum, 2005	AChE inhibitor
Zeta-cypermethrin	–	47	–	90-day neurotoxicity rat [redacted] Administration via diet Highest tested dose	Source: DAR	Known Binding to VGSC

CAG: cumulative assessment group; AS: active substance; NOEL: no observed effect level; LOEL: lowest observed effect level; NOAEL: no observed adverse effect level; LOAEL: lowest observed adverse effect level; bw: body weight; GABA: gamma-aminobutyric acid; AChE: acetylcholinesterase; VGSC: voltage-gated sodium channel; nAChR: nicotinic acetylcholine receptor; MoA: mode of action.

Table B.8: CAG on brain and/or erythrocyte acetylcholinesterase inhibition: toxicological characterisation of ASs to be considered in chronic exposure/risk assessments

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Acephate	AChE inhibition (brain, erythrocytes)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: JMPR 2002	AChE inhibitor
Aldicarb	AChE inhibition (erythrocytes)	0.05	0.1	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 1996	AChE inhibition
Azinphos-ethyl	AChE inhibition (erythrocyte)	0.0125	0.025	90-day dog [redacted] Administration via diet	Source: JMPR 1973	AChE inhibitor
Azinphos-methyl	AChE inhibition (erythrocytes)	0.16	0.74	1-year dog [redacted] Administration via diet	Source: DAR 1996, Addendum 6, 2000 (neurotox)	AChE inhibitor
Benfuracarb	AChE inhibition (erythrocytes)	1.81	9.4	28-day neurotoxicity rat [redacted] Administration via diet	Source: DAR 2004 EFSA (2009)	AChE inhibition
Cadusafos	AChE inhibition (erythrocytes)	0.045	0.22	2-year rat [redacted] Administration via diet	Source: DAR 2004 EFSA (2009)	AChE inhibitor
Carbaryl	AChE inhibition (brain, erythrocytes)	1	10	90-day neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2004 EFSA (2006)	AChE inhibition
Carbofuran	AChE inhibition (brain)	0.015	0.03	Acute neurotoxicity rat [redacted] Administration via gavage	Source: revised DAR 2008	AChE inhibition
Carbosulfan	AChE inhibition (brain, erythrocytes)	0.5	5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: Revised DAR 2009 EFSA conclusions on carbosulfan (2009) considered.	AChE inhibition
Chlorfenvinphos	AChE inhibition (brain, erythrocytes)	0.15	15	2-year rat (author not reported, JMPR 1994) Administration via diet	Source: JMPR 1994	AChE inhibitor
Chlorpyrifos	AChE inhibition (erythrocytes)	0.1	1	2-year rat [redacted] Administration via diet	Source: Addenda to the original Assessment Report (2013) EFSA conclusions on chlorpyriphos (2014) considered	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Chlorpyrifos-methyl	AChE inhibition (brain, erythrocytes)	1	50	2-year rat [redacted] Administration via diet	Source: DAR 1997 EC review report on chlorpyrifos-methyl (2005) considered	AChE inhibitor
Diazinon	AChE inhibition (brain, erythrocytes)	0.02	5.6	90-day dog [redacted] Administration via diet	Not approved EFSA conclusions on diazinon (2006) considered	AChE inhibitor
Dichlorvos	AChE inhibition (erythrocytes)	0.008	0.08	2-year dog [redacted]	Source: DAR 2003 EFSA conclusions on dichlorvos (2006) considered	AChE inhibitor
Dimethoate	AChE inhibition (brain, erythrocytes)	0.1	0.2	2-year rat [redacted] Administration via diet	Source: DAR 2004 EFSA conclusions on dimethoate (2006, 2013) considered ADI 0.001 (EFSA 2013) Overall NOAEL, combining reproduction, neurotoxicity and developmental neurotoxicity studies	AChE inhibitor
Ethephon	AChE inhibition (erythrocytes)	6	14	28-day cholinesterase inhibition study in dogs [redacted] Administration via diet	Source: DAR 2004 EFSA conclusions on ethephon (2008) considered	AChE inhibition
Ethion	AChE inhibition (brain)	0.06	0.71	90-day dog [redacted] Administration via diet	Source: JMPR 1990	AChE inhibition
Ethoprophos	AChE inhibition (brain)	0.04	2.4	2-year rat [redacted] Administration via diet	Source: DAR 2004 EFSA conclusions on ethoprophos (2006) considered.	AChE inhibition
Fenamiphos	AChE inhibition (brain, erythrocytes)	0.083	0.35	1-year dog [redacted] Administration via diet	Source: DAR 2003 EFSA conclusion on fenamiphos (2006) considered	AChE inhibition
Fenitrothion	AChE inhibition (brain, erythrocytes)	0.5	1.5	2-year rat [redacted] Administration via diet	Source: DAR 2003 EFSA conclusions on fenitrothion (2006) considered	AChE inhibition
Fenthion	AChE inhibition (erythrocytes)	0.05	0.23	1-year dog [redacted] Administration via diet	Source: DAR 1996	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Fonofos	AChE inhibition (erythrocytes)	0.2	1	1-year dog [redacted] Administration via capsule	Source: EPA 1999 US evaluation not completed because voluntary withdrawal in 1999	AChE inhibition
Formetanate	AChE inhibition (erythrocytes)	0.37	1.75	1-year dog [redacted] Administration via diet	Source: DAR 2004 EFSA conclusions on formetanate (2006) considered	AChE inhibition
Fosthiazate	AChE inhibition	0.42	2.36	2-year rat [redacted]	Source: DAR EC review report on fosthiazate (2003) considered.	AChE inhibitor
Malathion	AChE inhibition (erythrocyte)	17	35	2-year rat [redacted] and 2-year rat [redacted] combined Administration via diet	Source: DAR	AChE inhibitor
Methamidophos	AChE inhibition (brain, erythrocytes)	0.1	0.22	2-year rat [redacted] Administration via diet	Source: DAR 2000 EC review report on methamidophos (2006) considered	AChE inhibitor
Methidathion	AChE inhibition (brain, erythrocytes)	0.16	0.8	2-year rat [redacted] Administration via diet	Source: JMPR 1992	AChE inhibitor
Methiocarb	AChE inhibition (erythrocytes)	1.32	6.46	90-day dog [redacted] Administration via diet	Source: DAR 2004 EFSA conclusions on methiocarb (2006, 2018) considered	AChE inhibition
Methomyl	AChE inhibition (brain, erythrocytes)	0.25	0.5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR EFSA conclusions on methomyl (2006 and 2009) considered	AChE inhibition
Monocrotophos	AChE inhibition (brain, erythrocytes)	0.005	0.05	2-year rat [redacted] Administration via diet	Source: JMPR 1991	AChE inhibitor
Omethoate (IC) (metabolite of dimethoate)	AChE inhibition (erythrocytes)	0.027	0.04	2-year rat [redacted]; supplementary 32-week rat [redacted]	Source: DAR 2004 (dimethoate) EFSA conclusions on dimethoate (2013, 2018) considered.	AChE inhibitor
Oxamyl (IC)	AChE inhibition (brain, erythrocytes)	0.1	0.75	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003 EFSA conclusions on oxamyl (2005) considered	AChE inhibition

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Oxydemeton-methyl	AChE inhibition (brain, erythrocytes)	0.027	0.224	2-year rat [redacted] Administration via diet	Source: DAR 2004	AChE inhibitor
Parathion	AChE inhibition (brain)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: JMPR 1995	AChE inhibition
Parathion-methyl	AChE inhibition (erythrocytes)	0.25	2.5	2-year rat [redacted] Administration via diet	Source: DAR 2001	AChE inhibition
Phenthoate	AChE inhibition (erythrocytes)	0.29	0.87	2-year dog [redacted] Administration via diet	Source: JMPR 1980	AChE inhibitor
Phosalone	AChE inhibition (erythrocytes)	0.17	0.9	1-year dog [redacted] Administration via diet	Source: DAR 2004	AChE inhibition
Phosmet	AChE inhibition (brain)	1.1	1.8	2-year rat [redacted] Administration via diet	Source: DAR 2004 EFSA conclusions on phosmet (2006) considered	AChE inhibition
Phoxim	AChE inhibition (erythrocytes)	0.1	0.38	2-year dog [redacted] Administration via diet	Source: JECFA 1999	AChE inhibition
Pirimicarb	AChE inhibition (brain, erythrocytes)	10	25	1-year dog [redacted] Administration via capsule	Source: DAR 2003	AChE inhibition
Pirimiphos-methyl	AChE inhibition (brain)	0.4	2.1	2-year rat [redacted] Administration via diet	Source: DAR 2003 EFSA conclusions on pirimiphos-methyl (2005) considered.	AChE inhibition
Profenofos	AChE inhibition (erythrocytes)	0.017	0.56	2-year rat [redacted] Administration via diet	Source: JMPR 2007	AChE inhibitor
Pyrazophos	AChE inhibition (erythrocytes)	0.05	0.125	2-year dog [redacted] Administration via diet	Source: JMPR 1992	AChE inhibition
Thiodicarb	AChE inhibition (brain, erythrocytes)	0.5	5	Acute neurotoxicity rat [redacted] Administration via gavage	Source: DAR 2003 EFSA conclusions on thiodicarb (2005) considered. NOAEL derived from the LOAEL with an UF of 10.	AChE inhibition
Tolclofos-methyl	AChE inhibition (brain, erythrocytes)	6.9	34	2-year mouse [redacted] Administration via diet	Source: DAR 2003 EFSA conclusions on tolclofos-methyl (2017) considered	AChE inhibition
Triazophos	AChE inhibition (erythrocytes)	0.012	0.13	1-year dog [redacted] Administration via diet	Source: JMPR 2002	AChE inhibitor

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Trichlorfon	AChE inhibition (brain)	4.5	13.3	2-year rat [redacted] Administration via diet	Source: DAR 2004	AChE inhibitor

CAG: cumulative assessment group; AS: active substance; NOEL: no observed effect level; LOEL: lowest observed effect level; NOAEL: no observed adverse effect level; LOAEL: lowest observed adverse effect level; bw: body weight; AChE: acetylcholinesterase; MoA: mode of action.

Table B.9: CAG on functional effects on neuropathological end-points: toxicological characterisation of ASs to be considered in chronic exposure/risk assessments

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Chlorfenapyr	Myelin degeneration	14.8	27.6	90-day mouse [redacted]	Source: JMPR 1992	Unknown
Cymoxanil	Axonal degeneration, myelin degeneration	5	38	2-year rat [redacted]	Source: DAR	Unknown
Cypermethrin	Axonal degeneration (degeneration of trigeminus and increased galactosidase activity)	25	50	7-day rat [redacted]	Source: DAR	Unknown
Emamectin benzoate (IC)	Axonal degeneration, myelin degeneration	0.25	0.5	90-day dog [redacted]; 1-year dog [redacted]	Source: DAR 2008	Unknown
Fenpropidin	Myelin degeneration	5	20	1-year dog [redacted]	Source: DAR	Unknown
Flufenacet	Axonal degeneration	1.14	27	1-year dog [redacted]	Source: DAR 1998	Unknown
Indoxacarb	Axonal degeneration, neuronal degeneration/ necrosis	4	20	18-month mouse [redacted]	Source: DAR	Unknown
Isoxaflutole	Axonal degeneration, myelin degeneration	20	500	2-year rat [redacted]	Source: DAR 1997	Unknown
Lindane	Myelin degeneration	0.5	5	3-day rat [redacted]	Source: DAR Not true NOAEL, but LOAEL divided by 10	Unknown
Mancozeb	Myelin degeneration (myelin damage and Schwann cell proliferation)	8.2	49	3-month rat [redacted]	Source: DAR 2000	Unknown

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Molinate	Axonal degeneration, myelin degeneration	1.8	13	2-year rat [redacted]	Source: DAR	Presumed Sulfoxide metabolites can react with sulfhydryl groups of amino acids and proteins
Oxasulfuron	Axonal degeneration, myelin degeneration (secondary to axonal degeneration)	1.5	99	18-month mouse [redacted]	Source: DAR	Unknown
Quinoclamine	Myelin degeneration	3.82	40.2	18-month mouse [redacted]	Source: DAR	Unknown
tau-Fluvalinate	Axonal degeneration, myelin degeneration	1	10	7-day neurotoxicity rat [redacted]	Source: DAR Not true NOAEL, but LOAEL divided by 10	Unknown
Tembotrione	Neuronal degeneration/ necrosis	26.7	111	90-day dog [redacted]	Source: DAR 2012	Unknown
Thiram	Sciatic nerve lesions (not specified)	1.4	14	2-year rat [redacted]	Source: DAR 1997 + Addenda	Presumed Cross-linking of axonal proteins via reaction of the metabolite CS2 with axonal proteins
Tri-allate	Axonal degeneration, myelin degeneration	6.4	32	3-month neurotoxicity rat [redacted]	Source: DAR	Unknown
Trichlorfon	Myelin degeneration	31.2	168	90-day neurotoxicity rat [redacted]	Source: DAR addendum, 2005	Presumed Inhibition of neuropathy target esterase (NTE) and increased intracellular calcium

Active substance	Indicator of specific effect	NO(A)EL mg/kg bw	LO(A)EL mg/kg bw	Study	Remark	MoA
Ziram	Axonal degeneration	9	27	2-year rat [redacted]	Source: DAR 1998	Presumed Cross-linking of axonal proteins via reaction of the metabolite CS2 with axonal proteins

CAG: cumulative assessment group; AS: active substance; NOEL: no observed effect level; LOEL: lowest observed effect level; NOAEL: no observed adverse effect level; LOAEL: lowest observed adverse effect level; bw: body weight; MoA: mode of action.

Appendix C – Observed indicators of functional alterations of the motor, sensory and autonomic divisions of the nervous system in regulatory studies conducted with ASs belonging to chemical classes with a known neurotoxic MoA

Chemical classes/ Active substances	Observed indicators		
	Motor division	**Sensory division**	**Autonomic division**
***N*-methyl carbamate** insecticides	↑ motor activity, tremor, hyperactivity, ↓ motor activity, ataxia, muscle fasciculation, hunched posture Landing-foot splay, convulsions, muscle weakness, ptosis	↓ reactivity (tail pinch response, analgesic reflex (nociception response), righting reflex (air drop)), abnormal response to visual placing test, auditory startle response, ↓ Pupil response, hyperreactivity	Salivation, lacrimation, miosis, urination
Macrocyclic lactone insecticides	ataxia, tremor, ↑ motor activity, ↓ motor activity	↓ splay reflex, ↓ pupil reactivity, hyperreactivity	Salivation, mydriasis
Neonicotinoids insecticides	Tremor, ↓ motor activity, hunched posture, ataxia, convulsions, ↑ grip strength, hypoactivity	↑ reactivity, ↓ arousal, ↓ righting reflex	Urination, mydriasis, salivation
Organophosphorous insecticides	↓ motor activity, ↓ grip strength, hypoactivity, abnormal gait, ataxia, convulsions, tremor, ↑ motor activity, Hyperactivity, ↑ muscle strength, ↓ muscle strength, muscle weakness, hunched posture	↓ reactivity (righting reflex (air drop), tail pinch), ↓ Pupil response, ↓ analgesic reflex, hyperreactivity	Salivation, lacrimation, Miosis, urination, piloerection
Organochlorine insecticides	↑ motor activity, Convulsions, tremor, ↓ motor activity, abnormal gait	↑ reactivity, hyperreactivity	Salivation
Phenylpyrazole insecticides	landing-foot splay, convulsions	Approach response, ↓ tail pinch response, air righting reflex	Miosis
Pyrethrins and **pyrethroid** ester insecticides	↑ motor activity, ↓ motor activity, abnormal gait, ↓ grip strength, ataxia, tremor, convulsions, choreoathetosis, landing-foot splay, hunched posture, paresis limbs	↓ reactivity, ↓ touch responses, ↓ tail pinch response and impaired righting, ↑ sensitivity to noise, ↓ startle response, ↑ startle response, righting reflex	Salivation, urination, mydriasis, piloerection
Amitraz	↑ motor activity, convulsions	Hyperreactivity	
Chlormequat	ataxia	↓ reflex response	Salivation
Indoxacarb	ataxia, hunched posture landing-foot splay, ↓ grip strength, ↓ motor activity	hyperreactivity	Urination
Mepiquat	↓ motor activity, lateral posture, convulsions	↓ pupillary reflex	Salivation
Sulfoxaflor	↓ motor activity	↓ reactivity: touch response (handling reactivity)	Lacrimation

Appendix D – Effects on the motor division Uncertainty question 1 – evidence collection and grouping of ASs P

Appendix D can be found in the online version of this output ('Supporting information' section): https://doi.org/10.2903/j.efsa.2019.5800