Environmental Toxicology and Chemistry—Volume 40, Number 11—pp. 2978–2989, 2021
Received: 2 August 2021 | Accepted: 7 September 2021

Critical Perspectives

Assessing the Risks to Bats from Plant Protection Products: A Review of the Recent European Food Safety Authority Statement Regarding Toxicity and Exposure Routes

A.C. Brooks,[a,*] J. Nopper,[b] A. Weyers,[c] H. Crosland,[a] M. Foudoulakis,[d] S. Haaf,[e] M. Hackett,[a] and A. Lawrence[a]

[a]Cambridge Environmental Assessments, Cambridge, UK
[b]BASF, Limburgerhof, Germany
[c]CropScience Division, Bayer, Monheim, Germany
[d]Corteva™ Agriscience, Halandri, Greece
[e]ADAMA Deutschland, Cologne, Germany

Abstract: Wild birds and mammals that feed in agricultural habitats are potentially exposed to pesticides through various routes. Until recently, it has been implicitly assumed that the existing European Union risk assessment scheme for birds and mammals also covered bats (Chiroptera). However, recent publications raised concerns and, in 2019, a scientific statement was published by the European Food Safety Authority (EFSA) that concluded that bats were not adequately covered by the current risk assessment scheme. We review the evidence presented and assumptions made in the EFSA bat statement relating to toxicity, bioaccumulation, and exposure pathways (oral, dermal, and inhalation), in terms of their relevance for bats potentially foraging in agricultural areas in the European Union; we highlight where uncertainties remain and how these could be addressed. Based on our review, it is clear that there is still much uncertainty with regard to the appropriateness of the assumptions made in the EFSA bat statement. Significantly more information needs to be gathered to answer fundamental questions regarding bat behavior in agricultural landscapes, together with the relative sensitivity of bats to pesticide exposure. Given the current critical information gaps, it is recommended that quantitative risk assessments for bats not be performed for pesticides until more robust, reliable, and relevant data are available. The risk to bats can then be compared with that for birds and ground-dwelling mammals, to determine the protectiveness of the existing scheme and thus whether a bat scenario is indeed required and under what circumstances. *Environ Toxicol Chem* 2021;40:2978–2989. © 2021 Cambridge Environmental Assessments, part of RSK ADAS Ltd. *Environmental Toxicology and Chemistry* published by Wiley Periodicals LLC on behalf of SETAC.

Keywords: Bats; Chiroptera; Pesticides; Risk assessment; Toxicity; Exposure

INTRODUCTION

Wild birds and mammals that feed in agricultural habitats are potentially exposed to pesticides through various routes (Brooks et al., 2017). The risks to wildlife from exposure to pesticides are assessed in the European Union under the current regulation concerning the placing of plant protection products on the market (European Commission [EC], 2009). The guidance for assessing the risks to birds and mammals from potential exposure to pesticides in the European Union was published by the European Food Safety Authority (EFSA) in 2009 and is currently under review.

Until recently, it has been implicitly assumed that the EFSA (2009) risk assessment scheme for birds and mammals also covered the potential risks to bats (Chiroptera). However, recent publications (see Stahlschmidt & Brühl, 2012) raised concerns regarding the protectiveness of the current guidance for bats. Given the ecology and characteristics of bats, and the protected status of all 53 European bat species, a review of the protectiveness was considered "vital" by the EFSA's Panel on Plant Protection Products and their Residues (PPR). In 2019, the Panel published a scientific statement regarding the coverage of bats by the current risk

* Address correspondence to amy.brooks@cea-res.co.uk
Published online 12 October 2021 9 September 2021 in Wiley Online Library (wileyonlinelibrary.com).
DOI: 10.1002/etc.5209

assessment framework for birds and ground-dwelling mammals (ESFA, 2019). Hereafter referred to as the "EFSA bat statement," the statement concluded that "bats would not be adequately covered by the current risk assessment scheme" and that there is a need to develop a new scheme for bats (EFSA, 2019).

The EFSA bat statement reviewed the available ecological knowledge base for bat behavior and ecology to identify the potential exposure of bats inhabiting or foraging in areas where pesticides might be applied. The potential risks from oral, dermal, and inhalation exposure were discussed. Although specific risks from dermal and inhalation exposure are in practice currently not assessed for birds and ground-dwelling mammals, it was considered that the characteristics of bats make them potentially more vulnerable to such exposures. The potential risk from simultaneous exposure to all exposure routes was also highlighted in the EFSA bat statement.

Our aim was to review the evidence presented in the EFSA bat statement relating to toxicity, bioaccumulation, and exposure pathways, in terms of its relevance for bats potentially foraging in agricultural areas in the European Union, and the likely protectiveness of the existing bird and mammal risk assessment scheme, highlighting where uncertainties remain and how these could be addressed.

TOXICITY

The EFSA bat statement aimed to determine whether the toxicological sensitivity of bats was comparable to that of birds and ground-dwelling mammals. The key question was whether the standard terrestrial vertebrate data set already collected for pesticide authorizations, comprising effects on bird (usually quail or duck species) and mammal (usually rodents or lagomorphs) species tested in laboratory conditions, is suitable for predicting effects on bats.

The comparison of toxicological sensitivity of bats compared with ground-dwelling mammals and birds in the EFSA bat statement was based on a literature review. The resulting toxicity values obtained for bats were compared with those available for mice, rats, and birds from public literature, regulatory documents (e.g., EFSA conclusions), or databases (e.g., the Hazardous Substances Data Base [HSDB], the Extension Toxicology Network [EXTOXNET] InfoBase, the US National Institute for Occupational Safety and Health [NIOSH] database, the Toxicology Data Network [TOXNET]). The bat data identified were primarily from oral exposure studies ($n = 8$), but some dermal ($n = 4$) and dietary ($n = 1$) studies were also available. Overall, only 13 toxicity endpoints for 12 substances were identified for bats. A more recent review (Torquetti et al., 2020) identified a total of 37 active substances or their metabolites that had been studied in relation to bats over the last 60 years. The authors concluded that most studies were observational and represented only 5% of bat species. There was little information available to assess the effects of pesticides on bat populations, representing a gap in scientific research.

In agreement with findings in Torquetti et al. (2020), only limited information on bat sensitivity to pesticides was likewise identified in the EFSA bat statement. The results obtained were inconsistent, with examples of bats being more, less, or equally sensitive compared with ground-dwelling mammals. For example, the acute toxicity of methyl-parathion to big brown bat (*Eptesicus fuscus*) was lower (median lethal dose [LD50] = 372 mg/kg body wt) than for mice (LD50 = 44 mg/kg body wt; Clark, 1986). Conversely, the short-term dietary toxicity of deltamethrin to fruit bats (no-observed-effect level [NOEL] = 0.03 mg/kg body wt/day) was reported to be higher compared with ground-dwelling mammals and birds (toxicity endpoints above 1 mg/kg body wt/day; Oliveira et al., 2017). For chlorpyrifos (Eidels et al., 2016) and imidacloprid (Hsiao et al., 2016), comparable effects on bats and ground-dwelling mammals were observed. It was not always possible to determine the relative sensitivity of bats based on the data available; for example, in a study with DDT (Hurley & Fenton, 1980), no body weight measurements were taken, and the test substance was simply dusted on the back of isolated bats. Thus the exposure dose experienced by the bats (in terms of mg/kg body wt) is uncertain. Thus, comparison with known exposure doses in ground-dwelling mammals is of limited value.

Based on the literature review results, the EFSA bat statement authors stated that "it is not possible to draw a conclusion on bat susceptibility to pesticides relative to other bird or mammal species, and there is probably no relationship between toxicity to bats and other mammals or birds on which a predictive assessment could be made." If it is not possible to base the risk assessment for bats on the available information for ground-dwelling mammals or birds, then this would suggest that additional testing would be required to confirm bat sensitivity. However, rather than recommending further direct toxicity testing for bats, the EFSA bat statement suggests the "development of an adverse outcome pathway approach, whenever possible." Some relevant ex vivo and in vitro approaches developed for mammals (Boitet et al., 2018; Hodroge et al., 2011) were discussed, and were suggested as the most appropriate pathway for determining bat susceptibility to pesticides. However, as there is no clear pattern of sensitivity observed in bats versus ground-dwelling mammals for pesticides, with only a limited initial database of in vivo toxicity testing, the ability to develop ex vivo or in vitro assays is currently limited. Therefore, the use of an adverse outcome pathway approach would still require the generation of robust in vivo studies to determine apical adverse outcomes and to calibrate any ex vivo and in vitro approaches developed for bats.

There are also insufficient toxicity data to determine whether there are differences in sensitivity between bat species. Although studies on bats identified in the literature review cover a range of species, only a single study/substance is presented. Therefore, the interspecies variability of bat sensitivity to individual pesticides is not included in the comparison and is also currently unknown. To address this issue, it may be useful to perform additional literature searches to include all organic chemicals, not just pesticides. Literature data could also be used to identify worst-case European bat species in terms of life history traits, ecology, and behavior in agricultural landscapes. These data could then be used to inform future testing and/or assessment strategies.

In summary, there are currently limited data available in the literature on bat sensitivity to pesticide exposure, mainly based on legacy compounds. Due to the scarcity of data and inconsistent methods used, there is not a clear indication as to whether bats are more or less sensitive than ground-dwelling mammals or birds. A number of different bat species were tested in each of the studies, and no standardized guideline was used to perform these studies. For mice, rats, and birds, the toxicity data were primarily obtained from regulatory studies. Therefore, the data being compared for bats and other terrestrial vertebrates cannot be considered equivalent. Until such time that a reliable estimate of relative sensitivity of bats compared with birds and ground-dwelling mammals is available, our recommendation is that no quantitative regulatory risk assessment should be undertaken.

BIOACCUMULATION

In terms of bioaccumulation, the focus in the EFSA bat statement was to investigate whether there is greater concern for bats than for birds or ground-dwelling mammals and, therefore, whether the existing secondary poisoning risk assessment scheme within the EFSA (2009) is sufficiently protective. This concern predominantly arises due to the occurrence of hibernation and torpor (controlled reduction of body temperature and metabolic rate reducing energy expenditure) in bats. Bats accumulate stores of brown and white fats during active periods that are then metabolized and used as energy during hibernation and torpor as well as during arousal from these states. The EFSA bat statement postulates that high fat consumption during hibernation may expose bats to mobilized accumulated lipophilic active substances, potentially increasing the risk of direct mortality. For example, a study by Clark and Krynitsky (1983) investigating dichlorodiphenyldichloroethylene (DDE) observed high residues in brown fats, which were metabolized during hibernation and resulted in markedly increased DDE concentrations in the body. Although no toxic effects were identified in the study, Clark and Krynitsky (1983) questioned whether such effects may occur in the event of higher initial fat concentrations or for more toxic active substances. We do not consider it useful to refer to such legacy compounds for the assessment of current pesticide chemistry, with problematic compounds such as these now being screened out during the mandatory assessment of persistence, bioaccumulation, and toxicity (PBT).

Substances that are bioaccumulative would today be screened out in the PBT assessment, which uses fish bioconcentration data to protect all freshwater, marine, and terrestrial wildlife groups. Recommendations provided by the EFSA (2009) are then applied to assess the risk of secondary poisoning for any remaining substances with the potential to bioaccumulate (log potential octanol–water partition coefficient [P_{OW}] > 3), using fish-eating and earthworm-eating bird and mammal scenarios. Furthermore, the risk to any small mammal (including bats) can also be evaluated using the information on adsorption, distribution, metabolism, and elimination from the mammalian toxicology studies undertaken in the process of pesticide registration as well as from the metabolism studies with livestock (EFSA, 2009).

In summary, some evidence exists that bioaccumulation occurs in bats, predominantly based on legacy compounds, but there are currently insufficient data available to robustly compare the extent or kinetics of bioaccumulation observed in bats with that of other mammals and birds. The potential for food-chain transfer to bats could be further investigated to determine whether this is already covered by the fish-eating and earthworm-eating bird and ground-dwelling mammal risk assessments.

EXPOSURE ROUTES

Under the current risk assessment framework for birds and ground-dwelling mammals (EFSA, 2009), the routes of exposure that are routinely assessed are dietary intake and drinking water. These are also relevant exposure routes for bats, which may feed on insect prey that contains pesticide residues and drink from water sources within agricultural areas. A number of additional exposure routes are proposed within the EFSA bat statement (Figure 1). The risk to bats from oral exposure via grooming and via milk (by pups) has been highlighted as a potential concern, and it has been proposed that the risks to bats via dermal and inhalation exposure should be assessed. The evidence presented in the EFSA bat statement for each of these exposure routes is reviewed below.

Oral exposure

Food intake rates. The food-intake rate (FIR) is a key driver in determining the extent of dietary exposure, and therefore the data used for FIR calculation is critical. The EFSA bat statement attempts to estimate bat-specific values based on consumption of contaminated food, to calculate shortcut values that are directly compared with those in the EFSA statement (2009).

For the purposes of a bat-specific FIR calculation, the authors of the EFSA bat statement have used data from two studies—one on *Myotis lucifugus* (Kurta & Kunz, 1988) and the other on *Plecotus auritus* (McLean & Speakman, 1999). *Myotis lucifugus* is an endangered species of insectivorous microbat found in North America, and *P. auritus* is a small Eurasian insectivorous species of bat found across Europe. The daily energy requirement and weight of lactating *M. lucifugus* was chosen for use in the FIR/body weight calculation, because the data from that study was stated to be more reliable and the species more representative of European bat species based on their size. The relevance of the approach taken is considered in detail below.

The method for calculating daily energy expenditure (DEE) values (and from there, FIR values) is outlined in EFSA (2009). The underlying data were derived from a project carried out by

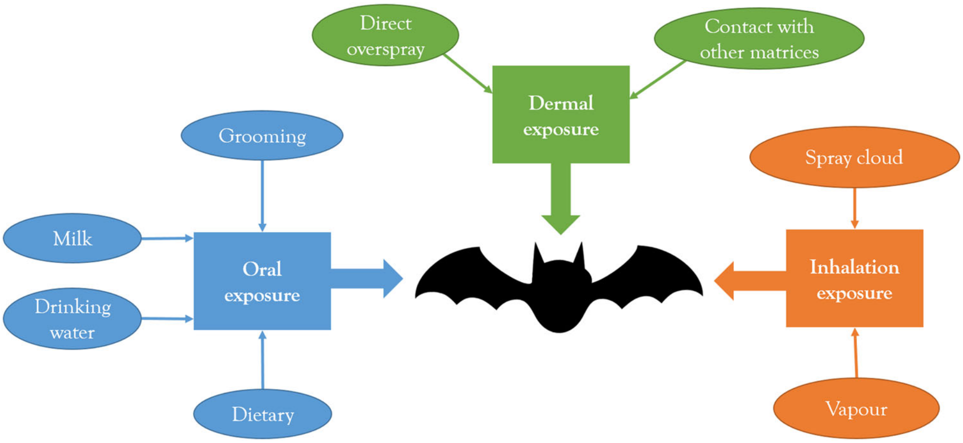

FIGURE 1: All potential exposure routes by which bats could encounter pesticides in agricultural landscapes, as proposed in the EFSA bat statement (ESFA, 2019). Note that not all routes will be applicable to the same individual, for example, either milk or the default dietary, but not both.

the Department for Environment, Food and Rural Affairs (DEFRA, 2007), consolidating 608 values for 115 species from 117 studies to estimate values for mammal DEEs.

The DEFRA (2007) report does not detail exactly which species were included in the DEE calculations, but it states that "'eutherian—other' includes all terrestrial mammals (e.g., bat, rat) requiring relatively moist habitats." This category, "eutherian—other," is the group whose values are presented in EFSA (2009) for the calculation of the mammal DEE. If bats are included in this data set, it is not clear why a new bat-specific approach was proposed in the EFSA bat statement. It is recommended that the data set used for the original EFSA (2009) calculations be further investigated to determine whether bats were included, and whether they fall within the range found for other mammalian species.

The DEFRA (2007) report does not state whether data for mammals were taken as basal metabolite rate, or what state the animals were in (e.g., pregnant or lactating). The worst-case was not selected for the FIR values, but instead a regression was fitted for the various mammal groupings, with "other eutherian mammals" considered the most appropriate for the risk assessment of ground-dwelling mammals. This is in contrast to the approach taken in the EFSA bat statement, in which the worst-case single-species data point for lactating bats was taken as the basis for FIR calculations. The EFSA bat statement authors are therefore comparing worst-case data (lactating females) for one species with data for many species that may not all be worst-case individuals (i.e., may not be lactating females), and thus this is not a like-for-like comparison. Furthermore, combining the FIR of lactating females with the smallest body weight is an unrealistically conservative combination, because lactating females will occur at the end of the season and will be heavier.

Although only two species were used to define FIR values (based on availability of data for pregnant and lactating females), the EFSA bat statement actually includes data on weight and energy consumption for 64 species of bats. For "other eutherians" in the DEFRA (2007) report, 46 mammal species were used. Therefore, there are sufficient data on bat species to calculate reliable values for the DEE equation. It may be possible to combine the bat data from DEFRA (2007) if this can be extracted. It is recommended that the resulting bat-specific DEE be assessed against the current ground-dwelling mammal DEEs to determine whether they are already protective of bats, as has been assumed until now.

An additional input into the calculation of FIR values is assimilation efficiency. The EFSA statement (2009) says that the assimilation efficiency of particular food items is group specific (i.e., mammals, passerine birds, etc.) as well as being specific to different food items. It is not explained in the EFSA bat statement whether values from EFSA (2009) have been used in the bat FIR calculations, or whether the assimilation efficiency for bats has been calculated using bat-specific data. The studies that were used to calculate the assimilation efficiency in EFSA (2009), Crocker et al. (2002), and Smit (2005) reference data on bats were grouped together with shrews ($n = 8$ for bats + shrews). It is recommended that further clarity be sought regarding the assimilation efficiency values used in the bat FIR calculations. It may be possible to obtain further data from the literature to calculate more specific assimilation efficiencies for European bat species.

Oral exposure via grooming. The EFSA bat statement proposes that self-grooming could potentially be a significant oral exposure route for bats that have been dermally exposed to pesticides. Although the EFSA bat statement asserts that self-grooming is common in bats, it is not clear how prevalent this behavior is across all bat species or how common it is in species linked to agricultural areas that might potentially be dermally exposed to pesticides. Reference is made to unpublished studies indicating that approximately 20% of roosting time was dedicated to self-grooming for Pipistrellus kuhlii and Hypsugo savii (cited as "L. Ancillotto and D. Russo, unpublished" in the EFSA bat statement). Other factors that would be important in determining exposure via grooming include the proportion of body surface groomed, the efficiency of grooming, and realistic estimates of dermal residue levels.

Social grooming within colonies may also result in oral exposure; however, based on the data cited in the EFSA bat statement, it seems that self-grooming is likely to be a greater potential source of exposure than social grooming, with approximately 20% of roosting time spent self-grooming versus approximately 7–8% of time spent socially grooming for *P. kuhlii* and *H. savii* (cited as "L. Ancillotto and D. Russo, unpublished" in the EFSA bat statement). It is recommended that further investigation be made into the prevalence of self and social grooming in European bat species that may potentially be exposed to pesticides in agricultural environments, to determine the relative importance of these exposure routes.

Although the EFSA bat statement authors consider that oral exposure via grooming is a relevant route of exposure, no method for estimating the level of exposure is provided. Although such a methodology could be developed, it is recommended that first the prevalence of grooming behavior in relevant bat species be investigated, and the methods for estimating dermal exposure first be improved (see later section on *Dermal exposure*).

Oral exposure of pups via milk. The EFSA bat statement assumes that residues of pesticides taken up by the adult are transferred to milk and subsequently to pups via the consumption of milk. This is true, but is also true for other mammals, for which the established guidance considers the chemical properties of the active substance or absorption, distribution, metabolism, and excretion (ADME) processes within the animal.

The calculation of pup exposure via milk in the EFSA bat statement uses a pup weight of 1.28 g for a newborn common pipistrelle (from Jones et al., 2009). Data from a study by Nagel and Disser (1990) were used to conclude that the oral exposure for newborn young is "equivalent to the adult exposure multiplied by 0.83 times the ratio of adult to young body weight." No further details are provided of how the value of 0.83 times was calculated. The abstract for Nagel and Disser (1990) states that residue concentrations in young were higher than those found in adults. The authors roughly estimated that the "quantity of chlorinated hydrocarbons transferred to the young amounts to 83% of the accumulated residues of total DDT in the adult females," which may be the source of the 0.83 conversion figure stated in the EFSA bat statement.

The study by Nagel and Disser (1990) focused on residues of chlorinated hydrocarbon pesticides in the common pipistrelle (females, males, and offspring). The chemicals used were hexachlorobenzene and lindane (substances currently not approved for use in the European Union); DDE (a breakdown product of DDT, not currently approved for use in the European Union); and three polychlorinated biphenyls (their use is currently highly restricted in the European Union). It is recommended that this issue be further investigated to determine how appropriate the value of 0.83 is for current pesticides.

A case study (Hofmann & Heise, 1991) of a poisoning incident of young bats in the field is also mentioned in the EFSA bat statement. This poisoning incident was attributed to applications of the organophosphate insecticide methamidophos and subsequent exposure via milk from adults consuming contaminated insects. This active substance is an acutely highly toxic organophosphate, not currently approved for the European market and therefore not relevant for the assessment of current pesticide chemistry.

The data referenced in the EFSA bat statement to support the case for oral exposure of pups via milk is therefore comprised of legacy chemicals and may not be appropriate for more current chemistry. No evidence was presented for transferral to milk for currently authorized pesticides.

The EFSA bat statement does not discuss nonlipophilic substances. It is possible that a threshold could be set based on chemical properties. The EFSA (2009) statement says that a log octanol–water partition coefficent (K_{OW}) ≥ 3 indicates a potential for bioaccumulation. Therefore, for consistency, the risk to pups from oral exposure via milk could be assessed only for chemicals with a log $K_{OW} \geq 3$, provided that there is evidence for relevance of this route of exposure above this threshold, or when residues are detected in the milk in lactating goat studies that are used to inform the human health risk assessment.

The evidence provided in the EFSA bat statement appears to be based on persistent and bioaccumulative substances, and no consideration is currently given to the ADME processes that will occur within bats. The chemical properties of a pesticide and the ADME processes within an animal govern the residues in milk. The EFSA bat statement does refer to metabolism studies in milk-producing livestock as a possible way to estimate exposure. Available data on residues in milk for pesticides that are currently authorized in the European Union could be examined, to help provide an estimation of the extent of transfer from adult female to milk, and subsequently to pups; for example, EFSA (2018a) provides an analysis of 582 samples of cow's milk for pesticide residues. All pesticide residues that were detected in cow's milk were from substances that are currently banned in the European Union. This suggests that exposure via residues in milk is unlikely for currently authorized pesticides in the European Union, although it is noted that the impact of processing milk for human consumption would need to be considered, as would any differences in milk properties between bats and cows, for example, fat content. It is recommended that further consideration be given to the influence of ADME processes on the likelihood of transferal of residues via milk, including the transfer of parent active versus metabolites because this may influence toxicity.

Although the risk to ground-dwelling mammal offspring from exposure via milk is not currently explicitly assessed in EFSA (2009), any effects in pups resulting from exposure via milk would be covered by the toxicity endpoint from the multigeneration rat/mouse study. There is no quantitative evidence provided in the EFSA bat statement that oral exposure via milk is likely to pose a greater risk to bat pups than to offspring of ground-dwelling mammal species. Therefore, the risk assessment for ground-dwelling mammals may already be protective of the risk to bat pups. Further investigation is recommended in terms of evidence for pesticide exposure of bat pups versus exposure of ground-dwelling mammal offspring, together with rate of milk consumption by offspring, period over which offspring feed, and fat content of milk for bats and ground-dwelling mammals.

In summary, the EFSA bat statement aimed to determine whether dietary exposure of bats is within that estimated for birds and ground-dwelling mammals under EFSA (2009). The data used for food intake rate and assimilation efficiency should be further investigated, in terms of their relevance for European bat species in agricultural landscapes, and their comparability to parameters used for birds and ground-dwelling mammals. Further investigation should be made into the prevalence of grooming behaviors in European bat species. The input parameters for calculating exposure of pups via milk should be further investigated for relevance to bat species that are potentially foraging in agricultural areas treated with current pesticides. Available data on residues in milk should be further examined, to help estimate the transfer from adult female to pups and to further understand the role of ADME processes on residue transfer.

Dermal exposure

According to the EFSA bat statement, dermal exposure is the route that results in the highest magnitude of exposure to bats foraging in agricultural areas treated with pesticides, due to exposure to airborne spray droplets. An example of dermal exposure presented in the EFSA bat statement is the method used to cull vampire bats. Individual vampire bats are coated with anticoagulants and released back into the colony, resulting in exposure of others within the roost via grooming and contact exposure, and ultimately leading to their death (Johnson et al., 2014; Linhart et al., 1972). The bats in these studies were dermally exposed to anticoagulants by dissolving them in melted petroleum jelly and spreading the resulting solidified paste onto the back of the bat. This type of dermal exposure is likely to be far more severe than would occur as a result of a bat flying through a spray cloud.

The ESFA bat statement considers the risk to bats from dermal exposure to be higher than that of birds and other mammals due to their large and highly vascularized wing surface. Thus, there is the potential for dermal exposure and subsequent dermal penetration to occur for bats foraging in agricultural areas treated with plant protection products; however, it is the extent of that dermal exposure and penetration that needs to be ascertained. The key drivers of dermal exposure will thus be the extent that bats encounter dermal exposure, together with the ability of bat skin surfaces to absorb any chemicals that they come into contact with.

Direct overspray. The EFSA bat statement proposes that bats foraging in agricultural areas may encounter direct overspray by flying through the spray cloud. It states "There is no evidence to suggest that bats avoid machinery. Some species are even attracted to light…, and hawkers might be attracted to insects that are stirred up by machinery, as seen for other insectivorous predators in farmland …. Therefore, it cannot be ruled out that some bats will be attracted to spraying operations, and be exposed dermally." The primary foraging bout of a small hawker bat at dusk/early evening is assumed for the purposes of this scenario. However, the risks to bats from direct overspray are largely dependent on how bats behave and thus the extent to which they encounter it.

The EFSA bat statement assumes that bats will fly at 1 m behind the sprayer within the spray cloud either for their whole foraging bout (which is assumed to be 2 h), or will encounter the equivalent of 1 min of exposure by flying in and out of the spray cloud. These hypothetical scenarios have been proposed due to a lack of empirical data on how bats actually behave while crops are being sprayed. Although the EFSA bat statement acknowledges that it is unlikely that bats would fly 1 m behind a sprayer for 2 h, it is still included within the dermal exposure estimates. Given that some bats avoid flying while it is raining (see Voigt et al., 2011), it is likely that they would also avoid flying close to the sprayer to avoid becoming too wet.

Although the EFSA bat statement assumes that the direct overspray of bats will occur while they are flying at 1 m from the sprayer, the flight path of bats is likely to be significantly more complex than this. Bats can move both horizontally and vertically within the agricultural landscape and thus encounter both spatial and temporal gradients of air concentrations resulting from the spray cloud (Figure 2). A bat that is flying near the sprayer (position a in Figure 2) will experience a higher dermal exposure than one that is flying at the same height but further away from the sprayer in the residual spray cloud (position b or c in Figure 2). Similarly, a bat flying at a lower altitude (e.g., 1 m above the crop) will experience a higher dermal exposure than if it is are flying at a higher altitude (e.g., 10 m above the crop).

Bats are unlikely to remain at a constant height/distance away from the sprayer for the duration of their foraging bout, and thus the dermal exposure of bats will critically depend on the flight path that they take during the time that the spray cloud is present and on the movement of the spray cloud over time. This dynamic interaction between bat behavior and spray cloud movement is discussed further in the later section, *Estimating exposure from direct overspray*.

Direct overspray of bats will only potentially be an issue for applications of pesticides that are made when nocturnal bats are active. For example, some pesticides are required to be sprayed in the evening/dusk to avoid contact with foraging bees. For pesticides that do not require a bee label restriction, a mitigation option for bats may be to use a similar phrasing for spraying, for example, "Do not apply when bats may be active."

The EFSA bat statement refers to evidence that bats may be attracted to artificial lighting on sprayer equipment and farm buildings; however, the extent to which this applies to all European bat species that may be foraging in agricultural areas is unknown. A recent publication by EUROBATS (Voigt et al., 2018) extensively reviewed the available data for the response of bats to Artificial Light At Night (ALAN), highlighting that the response of bats to ALAN depends on the species, in terms of their size and foraging strategy. Therefore, it seems unlikely that all European bat species would be equally attracted to the activity of sprayers, with some species likely to actively avoid it. If the species that are attracted to sprayers are associated with particular regions, landscapes, or crop types, it may not

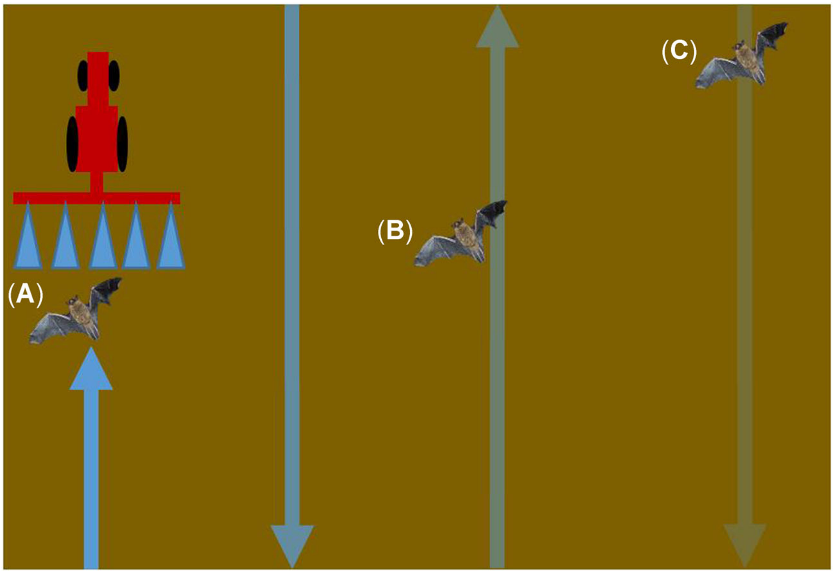

FIGURE 2: Graphic depicting the movement of bats within a crop that is being sprayed. The blue arrow indicates the direction of travel of the sprayer over time, starting travel from the top right hand side of the graphic. The transparency of the blue arrow indicates the relative air concentration resulting from the spray cloud, with the lowest concentration being at the top right hand side (paler blue) where spraying started and the highest being at the top left hand side (darker blue) where the sprayer is. See the text for description of dermal exposure of the bats in positions (**A**), (**B**), and (**C**).

be necessary to undertake a dermal risk assessment for all pesticide uses.

Estimating exposure from direct overspray. The EFSA bat statement proposes that the dermal exposure of bats (de) can be predicted by multiplying the volume of air that a bat passes through (v) by the proportion of pesticide coming into contact with the surface of the bat (i) and by the air concentration (ac) per unit body weight (bw; Equation 1).

$$de = v \times i \times \frac{ac}{bw} \quad (1)$$

The proportion of pesticide coming into contact with the bat surface (*i*) is assumed to be 1.0. This default assumption that the bat's wings would collect 100% of all dermally encountered residues, with none dripping off/being shaken off by the bat, is not plausible. Furthermore, it is assumed in the EFSA bat statement that 100% of these encountered residues will be absorbed across the wing membrane, which appears to be based on high permeability of the wings of fruit bats in terms of gas exchange; however, the relevance of gas exchange permeability compared with the absorption of liquids may be questionable. It also is not clear whether the morphology and properties of fruit bat wings are a suitable model for European bat species, most of which are not fruit bats. The assumption of 100% dermal penetration is more conservative than even for human health risk assessments. The default assumption for dermal penetration in humans is 10–75%, depending on the active substance content of the product, the log P_{OW}, and the molecular weight (EFSA, 2017).

In addition to the factors considered for human health risk assessments, there are other parameters that may be relevant in determining the extent of dermal penetration in bats. For example, dripping/shaking-off or evaporation may occur during bat flight that may reduce residues on the surface of the wing and thus reduce dermal penetration. There is some evidence that bat wings are coated in a waterproofing substance (see Sisk, 1957; Crowley & Hall, 1994), and this may result in increased dripping-off and thus reduced dermal exposure. Further investigation is recommended into the factors that could influence dermal penetration in bats, because it seems unlikely to be 100%, and thus *i* is likely to be less than 1.0.

The EFSA bat statement proposes that the volume of air that a bat would travel through (*v*) can be calculated by multiplying the front surface area of the bat (*sa*) by the speed of flight (*s*) and by the time spent in the spray cloud (*t*; Equation 2).

$$v = sa \times s \times t \quad (2)$$

The front surface area (*sa*) is calculated in the EFSA bat statement by assuming a full upward and downward wing stroke together with the area of the circle formed by the body diameter (Figure 3). The values used in the EFSA bat statement for the area formed by wing stroke (*wa*), body diameter (*bd*), and wing length (*wl*) to calculate the front surface area (*sa*) were either taken from the literature or estimated.

The EFSA bat statement then calculated the volume of air passed through by a foraging bat (*v*) for a small hawker and a large ground gleaner bat flying fast (6.7 m s) or slow (2.0 m s), and spending either 1 min or 2 h in the spray cloud. The input values for fast (Grodzinski et al., 2009) and slow (Kalko, 1995) flight speed (*s*) were taken from the literature for *Pipistrellus* species. The worst-case assumption regarding the time spent in the spray cloud (*t*) is based on the main foraging bout of a bat, such as a common pipistrelle, occurring entirely within the spray cloud, lasting up to 2 h (Bartonička et al., 2008). The

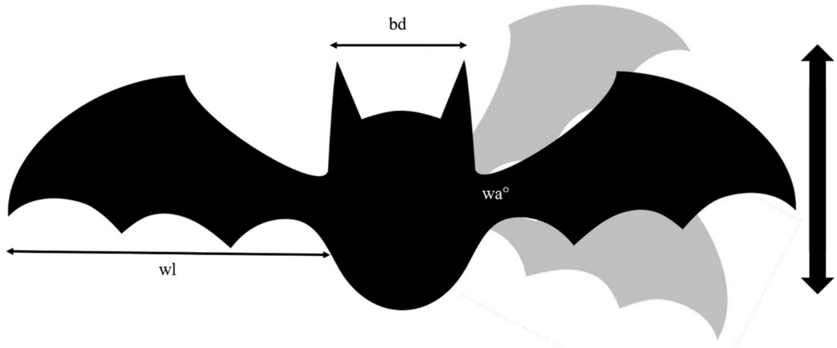

FIGURE 3: Scheme of surface area/air volume covered by bats flying through a spray cloud (wa = stroke angle of wing; wl = wing length; bd = body diameter). Based on the EFSA bat statement (ESFA, 2019).

relevance of the cited paper may be questionable given that the study focused on bats foraging in a floodplain forest, which may be of limited relevance for pesticide exposure scenarios. The EFSA bat statement acknowledges that spending 2 h within the spray cloud is not realistic, and thus an estimate that is considered to be "closer to real behaviour of bats" is also presented, with 1 min spent in the spray cloud and the remaining 119 min spent outside of the spray cloud. Further investigation into realistic durations spent in spray clouds in relevant habitats by relevant European bat species is recommended.

For the purposes of the direct overspray exposure calculations, it has been assumed that the bat is 1 m behind the sprayer for the duration of time spent in the spray cloud (*t*), being either 1 min or 2 h. In reality, the foraging path of bats is likely to be complex, in terms of distance from the sprayer in the vertical plane as well as the horizontal plane. Therefore, the combined assumption that a bat may spend 2 h at 1 m from the sprayer seems unrealistic. Even the least worst-case assumption of a bat spending 1 min at 1 m from the sprayer may still overestimate the exposure, for example, if bats actively avoid becoming too wet, or if bats actively avoid the noise and movement of machinery. Further investigation into bat behavior within pesticide-treated areas is therefore recommended.

Three models are presented in the EFSA bat statement that could be used to estimate the air concentration component of dermal exposure (*ac*): (1) the even distribution method, (2) the terrestrial investigation model (TIM), and (3) the drift area method. All are quite simplistic estimates of exposure that do not adequately take into account the temporal and spatial dynamics of the spray cloud itself. The EFSA bat statement acknowledges this, stating "although the pesticide concentrations in the air above the crop with time behind the sprayer could be calculated, this is also quite complex." Although modeling the predicted environmental concentration in air (PEC_{air}) is complex, it may be possible to use existing models or adapt those developed for other regulatory frameworks. The available options for modeling PEC_{air} for plant protection products were investigated by the FOCUS working group on pesticides in air (Kubiak et al., 2008); the report was finalized more than 10 years ago, and thus approaches may have since changed or new approaches may now be available. Although modeling the spatial and temporal dynamics of the spray cloud would be complex, this may be a useful higher tier refinement option and warrants further investigation, with simpler exposure models perhaps being reserved for screening stage risk assessments.

Dermal exposure estimates have been calculated within the EFSA bat statement for an application rate of 0.025 kg a.s./ha using all combinations of the three models for air concentration (even distribution, TIM, drift area), two bat species (hawker, ground gleaner), two flight speeds (slow, fast), and two durations of time spent in the spray cloud (1 min, 2 h). The resulting dermal exposure estimates varied enormously, being between 117 mg/kg body weight (slow, large bat spending 1 min in spray cloud using the drift area method) and 352 863 mg/kg body weight (fast, small bat spending 2 h in spray cloud using the even distribution method). This illustrates the importance of not only how the calculations are done, but also the input values used within the calculations. It also illustrates the importance of validating the results from exposure estimates against empirical field data; if the magnitudes of the predicted dermal exposure doses were occurring in reality, it would be expected that bat mortality would occur in the field and frequent sightings of dead bats would be reported by the public, which does not appear to be the case (see the UK Wildlife Incident Investigation Scheme; UK Health and Safety Executive, 1985).

The risk from dermal exposure via direct overspray has been investigated elsewhere for other terrestrial vertebrates. The EFSA bat scientific opinion on pesticide risk assessment for reptiles and amphibians (EFSA, 2018b) assumes that the upper surface of animals residing in the crop would be directly oversprayed at the full application rate, with no crop interception and 100% absorption. Dermal exposure of humans to pesticides is assessed under Regulation (EC, 2009), in terms of the risk to workers, residents, and bystanders. In the United States, the US Environmental Protection Agency (USEPA) uses TIM to estimate the dermal exposure of birds, assuming the bird is directly oversprayed while being static within the crop. None of these existing approaches are considered directly applicable to bats and would require additional parameterization to become relevant. The most significant difference is that they all assume that the target is static, whereas the bat would potentially be flying in and out of the spray cloud and thus experiencing a gradient of exposure in space and time. The extent of dermal exposure of bats should therefore take into account the impact of a bat's flight path, which may be dependent on the species and feeding guild.

Dermal exposure from residues in other matrices. Several other sources for dermal exposure beyond direct overspray are discussed in the EFSA bat statement. Bats that forage via gleaning may be exposed to residues on leaves and soil while capturing their prey. Bats may be exposed to residues present on surfaces in their roosts. Clustering of bats within roosts may result in contact with residues present on the body surfaces of their neighbors that have been foraging in treated areas. No further detail is presented in the EFSA bat statement regarding the estimation of such routes of exposure or their relative importance. Further investigation is recommended into these other potential sources of dermal exposure, in terms of their occurrence in European bats that may be present in agricultural areas.

In summary, the EFSA bat statement aimed to determine whether dermal exposure of bats was a concern. The primary route of dermal exposure is considered to be via direct overspray while foraging within a crop that is being sprayed with pesticides, although there is much uncertainty as to whether this hypothetical exposure scenario actually occurs in reality. Several methods for calculating dermal exposure via direct overspray are presented within the EFSA bat statement; however, the underlying assumptions and input parameters require further scrutiny to determine their relevance to realistic exposure scenarios. Further investigation is recommended into the behavior of bats in agricultural areas in the European Union, to determine critical information regarding species present, attractiveness to ALAN, duration of time spent in treated areas, flight paths in terms of proximity to sprayer, and residual spray cloud. The proportion of pesticide applications made at times when bats would be actively foraging should be investigated, in terms of whether this is more prevalent for particular scenarios (e.g., crop type, product type, geographical regions, season, etc.). Further investigation is recommended into the factors that could influence dermal penetration in bats. Spray cloud dynamics over time and space should be further investigated.

Inhalation exposure

Inhalation exposure is not formally considered in the current bird and mammal risk assessment framework (EFSA, 2009), although it is mentioned in the evaluation of conservatism for the Tier I "long-term exposure" scenario. That analysis suggests that although nondietary exposure routes (such as dermal and inhalation) may increase overall exposure of birds and mammals to some degree, this will be limited to a short time after application and will be potentially relevant for only an acute risk assessment. The EFSA bat statement proposes that exposure of bats via inhalation should be assessed, but does not indicate whether this should be an acute or long-term/reproductive risk assessment.

Inhalation exposure routes. The EFSA bat statement focuses on inhalation of airborne spray droplets, and does not formally consider vapor/volatilized fractions (although this is highlighted in the EFSA bat statement for further consideration). As for dermal exposure, the EFSA bat statement provides exposure estimates for an example scenario for 1 min and for up to 2 h post application. It is interesting to note that an (unlikely worst-case) 2-h constant inhalation exposure would result in a similar level of exposure to that anticipated via the oral route. In reality, the duration for which droplets are airborne post spray would be highly scenario specific—influenced by formulation, spray pressure, nozzle type and droplet size spectrum, sprayer type (e.g., airblast, boom), and environmental conditions such as humidity, temperature, and wind speed. The assumptions in the EFSA bat statement could be made more realistic by gathering further information on application technology and bat foraging ecology.

The EFSA bat statement does not include exposure via vapor, but does mention this as a potential source of exposure. For volatile substances, volatilization may occur after application, leading to a later airborne fraction. The volatilized active substance will have different properties from spray droplets, and will be highly compound specific; therefore the resulting level of inhalation exposure may differ from that of airborne spray droplets.

Estimating inhalation exposure. The EFSA bat statement provides methods to estimate exposure via inhalation for bats, based on respiratory rates for three bat species, and assumptions made about pesticide air concentrations. The proposed model for calculating exposure via inhalation refers to concentration in air, and volume of air respired (breathing rate and volume over time per unit body wt) by flying bats (worst-case compared with roosting bats), leading to an estimate of exposure in mg/kg body weight:

$$\textit{Inhalation exposure} = \textit{Concentration in air} \times \textit{Volume respired}$$

Although this model makes sense in general, there are various assumptions/omissions warranting further investigation. The extent of uptake via the lung membrane is likely to be highly compound specific. This could be investigated using existing inhalation toxicity studies, when available.

The EFSA bat statement includes a factor that accounts for the fraction of droplets that may be inhaled, being 0.28 based on TIM (USEPA, 2015). This assumes that only smaller droplets will be inhaled, but the size range of droplets (and by implication the fraction that may be inhaled) will depend largely on the application scenario. Further investigation is recommended into the appropriateness of the value of 0.28, because this presumably relates to an assumed spray droplet size distribution, which may not be relevant for all spray scenarios.

The flight behavior of bats would also have a significant effect on potential exposure to inhalable spray droplets. The exposure estimates presented in the EFSA bat statement assume that bats would spend either 1 min or 2 h foraging directly behind the sprayer. For example, the even distribution method assumes that all the applied spray from a boom is airborne within 0–0.5 m above the ground, and that bats forage within this area. This is clearly unrealistic and suggests that any inhalation exposure estimates derived with this methodology are extreme worst‐case. More realistic information on the foraging behavior of bats in relation to agricultural sprayers would provide context to these assumptions.

A further consideration is that of avoidance, which is not mentioned in the EFSA bat statement in relation to inhalation exposure. This might be in the form of direct avoidance, because bats foraging in the treated area would likely be able to detect the spray solution via taste or smell. Inhalation exposure will also be mitigated, because although bats may forage at dusk, not all pesticides are applied at this time. When pesticides are applied in the daytime, while bats are roosting, the potential for exposure to spray droplets via inhalation would be significantly reduced. No scope for direct or indirect avoidance currently exists in the EFSA bat statement, and thus further investigation into such bat behavior is recommended.

In summary, the EFSA bat statement aimed to determine whether exposure of bats via inhalation was a concern. It was not stated whether the risks from inhalation exposure are over an acute or chronic time scale. The primary focus for inhalation exposure is uptake of spray droplets; however, exposure to volatile substances may occur outside of the spray application. The assumptions used for calculating inhalation exposure are very conservative, and it is recommended that further information be gathered to increase the realism of the exposure estimates, for example, application technologies, bat foraging behavior, absorbance by lung membrane, or spray droplet size distribution. The occurrence of avoidance behavior by bats, via smell or taste, should also be investigated.

Combined exposure via oral, dermal, and inhalation routes. Although there is no extensive discussion regarding the topic of combined exposure within the EFSA bat statement, it is stated that "it is reasonable to assume that the different exposure routes (oral, dermal and inhalation) are **additive** if bats are foraging in the field when a pesticide is applied in the evening," and that "it is important to highlight that any risk assessment scheme should consider the **total body burden from all exposure routes** as bats foraging in the field will be exposed to residues in insects and also via dermal and inhalation routes." There is no further detail regarding how to calculate combined exposure or how to undertake a combined risk assessment, which would be outside the scope of the current bird and mammal risk assessment framework (EFSA, 2009).

The EFSA bat statement proposes that if a bat were actively foraging in a treated field during spray applications, then it could potentially be exposed via oral, dermal, and inhalation routes. However, the maximum internal exposure levels from each exposure route will not be reached at the same time. Dietary exposure will likely be the slowest, being processed through the digestive system, absorbed across the intestinal tract before entering the bloodstream, and transported to sites associated with toxicity. Inhalation exposure will likely be the quickest, because substances only need to be absorbed across the lung membrane before being transported in the bloodstream to the heart and then quickly circulated around the body. Dermal exposure will be somewhere in between, mainly being absorbed across the highly vascularized wing membrane and into the bloodstream. The ADME processes will also vary between the exposure routes, with dietary exposure via the intestinal tract first passing through the liver where detoxification processes can occur before being transported around the bat's body, further reducing the speed at which the maximum dietary exposure occurs. Therefore, because the peak exposure via each route will likely occur at different times within the bat, the total load at any given time is likely to be less than (maximum DDD_{oral}) + (maximum DDD_{dermal}) + (maximum $DDD_{inhalation}$), where DDD is daily dietary dose. Any assessment of combined exposure should therefore include consideration of the time course for internal exposure of the bat.

The route of exposure may also influence the type of effect observed. In regulatory toxicology data packages, test organisms are exposed via different routes (e.g., gavage, dietary, dermal, intravenous) depending on the study design and purpose, and these different exposure routes can affect the result observed even if the mg/kg body weight dose is the same. Therefore, if a combined exposure, or "total body burden," were calculated, it is unclear what toxicity endpoint this would be compared with. It would seem inappropriate to combine oral, dermal, and inhalation exposure and compare with a dietary toxicity endpoint, for example, particularly if it is known that the exposure route impacts the type or extent of the effect observed for that substance.

It is also unclear whether such a combined risk assessment is being proposed for an acute or chronic time scale. If the main concern is bats foraging in the field, then it would only be an acute exposure period, whereas the bat is eating insects and potentially flying through the spray cloud and experiencing dermal and inhalation exposure. However, it is possible that the EFSA bat statement authors are recommending a chronic risk assessment on the basis of short‐term exposure resulting in long‐term effects, although this is not the default approach for birds and mammals under the current risk assessment framework (EFSA, 2009).

DISCUSSION

The purpose of the present review was to evaluate the evidence used and assumptions made in the EFSA bat statement, regarding toxicity, bioaccumulation, and routes of exposure (oral, dermal, and inhalation). Furthermore, areas where further work is needed have been proposed, together with recommendations on a path forward to address the most important issues.

It is clear from our review that there are many unanswered questions in relation to the risk to bats from potential exposure to pesticides. The answers to these questions are fundamental to the integrity and realism of the resulting risk assessment framework. For example, there are currently insufficient toxicity data available to determine whether bats are more or less sensitive to pesticides compared with birds or ground-dwelling mammals. Therefore it is unclear whether existing toxicity endpoints in standard regulatory data packages are sufficiently protective. New methods are therefore required to estimate toxicity to bats. Similarly, it is unclear whether the risk to bats from bioaccumulation is comparable to the risk to birds and other mammals. In both cases, for toxicity and bioaccumulation, this is due to insufficient evidence for a conclusion in either direction.

In addition to there being insufficient information available regarding the effects and bioaccumulation of substances in bats, there is also a paucity of information regarding bat behavior in agricultural landscapes. Such information is critical in moving away from the currently proposed worst-case exposure assessment in the EFSA bat statement, to a more realistic, exposure-based approach. Clearly there is a hypothetical scenario in which bats could be actively flying while pesticides are being applied, but the real question should be how much this would actually occur in reality, that is, what is the real risk? And does this only occur in certain bat species, associated with certain crops, in certain geographic regions, and so on? And if bats do occur during pesticide applications, how do they behave during that time—do they fly directly behind the sprayer for prolonged periods of time, as proposed in the EFSA bat statement, or are they exhibiting a complex and dynamic flight path, moving in and out of an equally dynamic spray cloud?

There will likely be many scenarios for which the risks to bats are low, or are covered by the existing bird and mammal risk assessments, and therefore developing and undertaking separate complex, time-consuming, and potentially data-intensive risk assessment schemes for all pesticide uses for bats may be unnecessary. It would be better to construct a risk assessment framework based on realistic, sound science so that time is not wasted on low-risk chemicals, and only those with "real" potential risks to bats trigger the need for further consideration. This saves time and resources not only for industry companies attempting to seek authorizations for their pesticides, but also for the regulatory bodies that are tasked with evaluating all the submitted dossiers. It should be a principle of pesticide regulation to focus efforts where they are really required to achieve the protection goal.

CONCLUSIONS

Based on our review of the EFSA bat statement, it is clear that there is still much uncertainty with regard to the realism of the assumptions made, and therefore the conservativeness of a possible resulting risk assessment framework, for bats. Significantly more information needs to be gathered to answer fundamental questions, for example, which bat species are likely to be present in agricultural areas during spray applications, how they are behaving in terms of feeding preferences, flight paths and duration, and the dynamics of spray cloud behavior. The EFSA bat statement has taken a worst-case exposure-based approach to the risks to bats, and now it is important to assess how realistic this approach is for the target group for protection, that is, European bats feeding in agricultural landscapes. The usefulness of other parts of regulatory information for pesticides, particularly from the available data in the toxicology section, to inform a potential bat risk assessment needs to be explored further. Given the current critical information gaps, it is recommended that quantitative risk assessments for bats not be performed for pesticides until such time as more robust, reliable, and relevant data are collected to estimate the realistic risk to bats. Once such data are available, the risk to bats can then be compared with that for birds and ground-dwelling mammals, to determine the protectiveness of the existing scheme and thus whether a bat scenario is indeed required and under what circumstances.

Acknowledgment—The present review was funded by CropLife Europe (previously known as the European Crop Protection Association).

Author Contributions Statement—A.C. Brooks, H. Crosland, M. Hackett, and A. Lawrence: Preparation of a background review report on which the manuscript is based; A.C. Brooks, J. Nopper, and A. Weyers: Writing of the manuscript; H. Crosland, M. Foudoulakis, S. Haaf, M. Hackett, and A. Lawrence: Provision of technical and editorial assistance.

Data Availability Statement—All data referenced in the present review are available in the public domain. Data, associated metadata, and calculation tools are available from the corresponding author (amy.brooks@cea-res.co.uk).

REFERENCES

Bartonička, T., Řehák, Z., & Andreas, M. (2008). Diet composition and foraging activity of *Pipistrellus pygmaeus* in a floodplain forest. *Biologia*, *63*, 1–7.

Boitet, M., Hammed, A., Chatron, N., Debaux, J. V., Benoit, E., & Lattard, V. (2018). Elevated difenacoum metabolism is involved in the difenacoum-resistant phenotype observed in Berkshire rats homozygous for the L120Q mutation in the vitamin K epoxide reductase complex subunit 1 (Vkorc1) gene. *Pest Management Science*, *74*, 1328–1334.

Brooks, A. C., Fryer, M., Lawrence, A., Pascual, J., and Sharp, R. (2017) Reflections on bird and mammal risk assessment for plant protection products in the European Union: Past, present, and future. *Environmental Toxicology and Chemistry*, *36*, 565–575.

Clark, D. R. (1986). Toxicity of methyl parathion to bats: Mortality and coordination loss. *Environmental Toxicology and Chemistry*, *5*, 191–195.

Clark, D. R. Jr. & Krynitsky, A. J. (1983). DDE in brown and white fat of hibernating bats. *Environmental Pollution (Series A)*, *31*, 287–299.

Crocker, D., Hart, A., Gurney, J., & McCoy, C. (2002). *Methods for estimating daily food intake of wild birds and mammals* (Project No. PN0908). Central Science Laboratory.

Crowley, G. V., & Hall, L. S. (1994). Histological observations on the wing of the grey-headed flying-fox (*Pteropus-Poliocephalus*) (Chiroptera, Pteropodidae). *Australian Journal of Zoology*, *42*, 215–231.

Department for Environment, Food and Rural Affairs. (2007). *Improved estimates of daily food and water requirements for use in risk assessments* (Project Code PS2330).

Eidels, R. R., Sparks, D. W., Whitaker, J. O., & Sprague, C. A. (2016). Sub-lethal effects of chlorpyrifos on big brown bats (*Eptesicus fuscus*). *Archives of Environmental Contamination and Toxicology*, *71*, 322–335.

European Commission. (2009). Regulation (EC) No. 1107/2009 of the European Parliament and of the Council of 21 October 2009 concerning the placing of plant protection products on the market and repealing Council Directives 79/117/EEC and 91/414/EEC. *Official J Eur Union*, *L309*, 1–50.

European Food Safety Authority. (2009). Guidance document on risk assessment for birds and mammals on request from EFSA. *EFSA Journal*, *7*, 1438.

European Food Safety Authority. (2017). Guidance on dermal absorption. *EFSA Journal*, *15*(4873), 60.

European Food Safety Authority. (2018a). The 2016 European Union report on pesticide residues in food. *EFSA Journal*, *16*(5348), 139.

European Food Safety Authority. (2018b). Scientific opinion on the state of the science on pesticide risk assessment for amphibians and reptiles. *EFSA Journal*, *16*(5125), 301.

European Food Safety Authority. (2019). Scientific statement on the coverage of bats by the current pesticide risk assessment for birds and mammals. *EFSA Journal*, *17*(5758), 81.

Grodzinski, U., Spiegel, O., Korine, C., & Holderied, M. W. (2009). Context-dependent flight speed: Evidence for energetically optimal flight speed in the bat *Pipistrellus kuhlii*? *Journal of Animal Ecology*, *78*, 540–548.

Hodroge, A., Longin-Sauvageon, C., Fourel, I., Benoit, E., & Lattard, V. (2011). Biochemical characterization of spontaneous mutants of rat VKORC1 involved in the resistance to antivitamin K anticoagulants. *Archives of Biochemistry and Biophysics*, *515*, 14–20.

Hofmann, K., & Heise, G. (1991). Vergiftung junger Mausohren (*Myotis myotis*) durch Pflanzenschutzmittel. *Nyctalus*, *4*, 85–87.

Hsiao, C. J., Lin, C. L., Lin, T. Y., Wang, S. E., & Wu, C. H. (2016). Imidacloprid toxicity impairs spatial memory of echolocation bats through neural apoptosis in hippocampal CA1 and medial entorhinal cortex areas. *Neuroreport*, *27*, 462–468.

Hurley, S., & Fenton, M. B. (1980). Ineffectiveness of fenthion, zinc, phosphide, DDT and two ultrasonic rodent repellers for control of populations of little brown bats (*Myotis lucifugus*). *Bulletin of Environmental Contamination and Toxicology*, *25*, 503–507.

Johnson, N., Aréchiga-Ceballos, N., & Aguilar-Setien, A. (2014). Vampire bat rabies: Ecology, epidemiology and control. *Viruses*, *6*, 1911–1928.

Jones, K. E., Bielby, J., Cardillo, M., Fritz, S. A., O'Dell, J., Orme, C. D. L., Safi, K., Sechrest, W., Boakes, E. H., Carbone, C., Connolly, C., Cutts, M. J., Foster, J. K., Grenyer, R., Habib, M., Plaster, C. A., Price, S. A., Rigby, E. A., Rist, J., … Purvis, A. (2009). PanTHERIA: A species-level database of life history, ecology, and geography of extant and recently extinct mammals. *Ecology*, *90*, 2648.

Kalko, E. K. V. (1995). Insect pursuit, prey capture and echolocation in pipistrelle bats. *Animal Behavior*, *50*, 861–880.

Kubiak, R., Bürkle, L., Cousins, I., Hourdakis, A., Jarvis, T., Jene, B., Koch, W., Kreuger, J., Maier, W.-M., Millet, M., Reinert, W., Sweeney, P., Tournayre, J.-C., & Van den Berg, F. (2008). *Pesticides in air: Considerations for exposure assessment* (SANCO/10553/2006). FOCUS Working Group on Pesticides in Air (FOCUS Air Group). https://esdac.jrc.ec.europa.eu/public_path/projects_data/focus/air/docs/FOCUS_AIR_GROUP_REPORT-FINAL.pdf

Kurta, A., & Kunz, T. H. (1988). Roosting metabolic rate and body temperature of male little brown bats (*Myotis lucifugus*) in summer. *Journal of Mammalogy*, *69*, 645–651.

Linhart, S. B., Flores Crespo, R., & Mitchell, G. C. (1972). Control of vampire bats by topical application of an anticoagulant, chlorophacinone. *Boletin de la Oficina Sanitaria Panamericana*, *6*, 31–38.

McLean, J. A., & Speakman, J. R. (1999). Energy budgets of lactating and non-reproductive brown long-eared bats (*Plecotus auritus*) suggest females use compensation in lactation. *Functional Ecology*, *13*, 360–372.

Nagel, A., & Disser, J. (1990). Rückstände von Chlorkohlenwasserstoff-Pestiziden in einer Wochenstube der Zwergfledermaus (*Pipistrellus pipistrellus*). *Zeitschrift fur Säugetierkunde*, *55*, 217–225.

Oliveira, J. M., Losano, N. F., Condessa, S. S., de Freitas, R. M. P., Cardoso, S. A., Freitas, M. B., & de Oliveira, L. L. (2017). Exposure to deltamethrin induces oxidative stress and decreases of energy reserve in tissues of the neotropical fruit eating bat *Artibeus lituratus*. *Ecotoxicology and Environmental Safety*, *148*, 684–692.

Sisk, M. O. (1957). A study of the sudoriparous glands of the little brown bat, *Myotis lucifugus lucifugus*. *Journal of Morphology*, *101*, 425–455.

Smit, C. E. (2005). *Energy and moisture content and assimilation efficiency of bird and mammal food* (RIVM Report No. 601516013). RIVM. pp. 57–71.

Stahlschmidt, P., & Brühl, C. A. (2012). Bats at risk? Bat activity and insecticide residue analysis of food items in an apple orchard. *Environmental Toxicology and Chemistry*, *31*, 1556–1563.

Torquetti, C. G., Guimarães, A. T. B., & Soto-Blanco, B. (2020). Exposure to pesticides in bats. *Science of the Total Environment*, *755*, 142509.

UK Health and Safety Executive. (1985). *Wildlife Incident Investigation Scheme*. https://www.hse.gov.uk/pesticides/reducing-environmental-impact/wildlife/wildlife-incident-investigation-scheme.htm

US Environmental Protection Agency. (2015). *Technical description and user's guidance document for the terrestrial investigation model (TIM) Version 3.0 BETA*. Office of Pesticide Programs, Office of Chemical Safety and Pollution Prevention, U.S. Environmental Protection Agency.

Voigt, C. C., Azam, C., Dekker, J., Ferguson, J., Fritze, S., Gazaryan, M., Hölker, F., Jones, G., Leader, N., Lewanzik, D., Limpens, H. J. G. A., Mathews, F., Rydell, J., Schofield, H., Spoelstra, K., & Zagmajster, M. (2018). *Guidelines for consideration of bats in lighting projects* (EUROBATS Publication Series No. 8). UNEP/EUROBATS Secretariat. https://www.eurobats.org/sites/default/files/documents/publications/publication_series/WEB_DIN_A4_EUROBATS_08_ENGL_NVK_28022019.pdf

Voigt, C. C., Schneeberger, K., Voigt-Heucke, S. L., & Lewanzik, D. (2011). Rain increases the energy cost of bat flight. *Biology Letters*, *7*, 793–795.

