Published in final edited form as:

Food Chem Toxicol. 2017 December ; 110: 408–417. doi:10.1016/j.fct.2017.10.038.

APROBA-Plus: A probabilistic tool to evaluate and express uncertainty in hazard characterization and exposure assessment of substances

Bas G.H. Bokkers^{1,*}, Marcel J. Mengelers¹, Martine I. Bakker¹, Weihsueh A. Chiu², and Wout Slob¹

¹National Institute for Public Health and the Environment (RIVM), Bilthoven, The Netherlands

²Department of Veterinary Integrative Biosciences, College of Veterinary Medicine and Biomedical Sciences, Texas A&M University, College Station, TX, USA

Abstract

To facilitate the application of probabilistic risk assessment, the WHO released the APROBA tool. This tool applies lognormal uncertainty distributions to the different aspects of the hazard characterization, resulting in a probabilistic health-based guidance value. The current paper describes an extension, APROBA-Plus, which combines the output from the probabilistic hazard characterization with the probabilistic exposure to rapidly characterize risk and its uncertainty. The uncertainty in exposure is graphically compared with the uncertainty in the target human dose, i.e. the dose that complies with the specified protection goals. APROBA-Plus is applied to several case studies, resulting in distinct outcomes and illustrating that APROBA-Plus could serve as a standard extension of routine risk assessments. By visualizing the uncertainties, APROBA-Plus provides a more transparent and informative outcome than the more usual deterministic approaches, so that risk managers can make better informed decisions. For example, APROBA-Plus can help in deciding whether risk-reducing measures are warranted or that a refined risk assessment would first be needed. If the latter, the tool can be used to prioritize possible refinements. APROBA-Plus may also be used to rank substances into different risk categories, based on potential health risks without being compromised by different levels of conservatism that may be associated with point estimates of risk.

Keywords

probabilistic risk assessment; communicating uncertainty; exposure assessment

INTRODUCTION

In current risk assessments, uncertainties are often taken into account by using conservative values, such as a high consumer exposure value, or an interspecies assessment factor assumed to be an upper bound estimate. The risk assessment then combines all those

*PO Box 1, 3720 BA, Bilthoven, The Netherlands, bas.bokkers@rivm.nl, T: +31-(0)30-274 2117.

conservative values, resulting in a final value, such as the ratio of exposure and health-based guidance value, as an indicator of the risk. If this so-called risk ratio, risk characterization ratio (RCR) or risk quotient (also called “hazard quotient”) is far below unity (i.e. exposure is much lower than the health-based guidance value), it is generally concluded that the health risk is negligible, and no further information is needed. If, however, the risk ratio is close to unity or even exceeds it, a clear answer on the magnitude of the health risk cannot be given. In some cases risk managers may decide to take mitigating measures, in other cases they may consider a higher tier assessment, in an attempt to make (one or more of) the conservative values in the assessment less conservative by finding better data.

Instead of making such risk management decisions based on a risk ratio relying on conservative point estimates it has been proposed to first (better) quantify the level of conservatism in the final risk outcome by quantitatively evaluating the uncertainties involved in a so-called probabilistic assessment (WHO-IPCS, 2014). Here, the conservative single values are replaced by uncertainty distributions resulting in an overall uncertainty distribution of the final risk outcome (Bokkers et al., 2009; Slob et al., 2014; van der Voet and Slob, 2007). There are two important benefits to this procedure. First, an overall uncertainty distribution of the final risk outcome gives insight in the level of conservativeness related to the final risk value (e.g. the risk ratio) as derived from the conservative single values (the usual approach). It might be found that the latter risk number was overly conservative (due to “compounding” of multiple conservative values). With a probabilistic assessment it is possible to select a more reasonable level of conservatism, and then evaluate if the protection goals are met with sufficient confidence. Thus, the probabilistic outcome might indicate that risk-reducing measures or additional data generation would not be needed after all. Second, if the probabilistic outcome is still inconclusive with regard to the health risk, the results of the probabilistic analysis may help in pointing out which aspects in the overall risk assessment have the highest contribution to the overall uncertainty. That insight indicates what type of additional information in the higher tier assessment would be potentially effective in terms of reducing uncertainty.

A fully probabilistic assessment consists of two steps. First, the uncertainties in each of the most important aspects of both the hazard characterization (e.g. dose-response or interspecies uncertainty) and exposure assessment (uncertainties in e.g. concentrations or food consumption behavior) are quantitatively reflected by a statistical distribution. Next, the overall uncertainty in the final outcome is evaluated by probabilistically combining all the uncertainty distributions. This approach leads to both an upper and a lower bound for the hazard characterization outcome, as well as for the exposure outcome, making the overall uncertainty in both outcomes visible. A further gain of a probabilistic assessment is that the protection goals (i.e., the magnitude of the effect in individuals and the fraction of the population protected from that magnitude of effect) as well as the level of conservatism (i.e. the probability of not underestimating the risk) are made explicit and can be tailored to the decision context. In this way, the risk manager may ask for assessments based on various defined protection goals or levels of conservatism.

An example of the methodology as described above is the ‘integrated probabilistic risk assessment’ (IPRA) approach, developed by van der Voet and Slob (2007). IPRA

probabilistically evaluates the uncertainty and variability in all aspects of a risk assessment considered relevant. The methodology is based on Monte Carlo sampling, which makes it flexible and generally applicable. However, IPRA is relatively time-consuming as it requires detailed (re)analysis of hazard and exposure data to obtain the required input distributions.

To facilitate the application of probabilistic analysis, the WHO International Programme on Chemical Safety (WHO-IPCS, 2014) recently released a Microsoft Excel tool providing for an approximate probabilistic hazard characterization, which is easy and quick in use (see: http://www.who.int/ipcs/methods/harmonization/areas/hazard_assessment/en/).

This tool, called APROBA, applies lognormal uncertainty distributions to the different aspects of the hazard assessment (e.g. point of departure, assessment factors), resulting in a confidence interval for the human dose associated with specified protection goals, as well as a probabilistic health-based guidance value. Both can be quickly calculated in a spreadsheet without needing Monte Carlo simulation. The probabilistic health-based guidance value can be compared with a deterministic exposure value (e.g. a mean, a percentile of the population, or an upper confidence bound of the mean/percentile). This approach, however, leads to an “unbalanced” risk assessment in which uncertainties are made visible in the hazard but not in the exposure. The uncertainty in the exposure estimate should be made visible just as well, because the exposure metric greatly varies in the level of conservatism among different assessments. To bridge this gap, we extended the APROBA tool (hence called APROBA-Plus, see Supplemental Material: APROBA_PLUS-V0.96_v0.12_TEMPLATE.xlsx), by including the option to insert a quick and approximate estimate of the exposure uncertainty. This uncertainty in exposure is then graphically compared with the probabilistic outcome from the hazard characterization, similarly to the RISK21 matrix (Embry et al., 2014; Pastoor et al., 2014). Thus, APROBA-Plus is a rapid screening tool with the advantage of making the approximate uncertainties in the overall risk assessment easily visible. The aim of this paper is to illustrate the potential applications and benefits of quantitatively expressing uncertain risk outcomes, rather than providing a detailed description of the methodology, which is already provided elsewhere (WHO-IPCS, 2014).

We first illustrate the use of APROBA-Plus in three (food related) case studies, one of which (nivalenol) is fully reported in the main text. For the next two illustrations, only the final results are shown in the main text (with full report in the Supplemental Material). These three cases illustrate the three main types of possible outcomes of an APROBA-Plus analysis. In addition, we explored the usefulness of the tool using 16 substances drawn from the Dutch Quality Programme for Agricultural Products (van Dooren et al., 1995), which includes pesticides, metals, dioxins/PCBs, flame retardants and veterinary drugs. Based on the outcomes of the tested substances we discuss the potential use of APROBA-Plus in the risk assessment process in general. In addition, we show how the tool could be used for the purpose of prioritization or ranking of chemicals.

METHODS

The target human dose

WHO-IPCS (2014) defines the target human dose as the (true but unknown) human dose corresponding to two preset protection goals: a magnitude (M) of the effect in individuals and the fraction (incidence goal, I) of the population not being protected from that magnitude of effect. This target human dose is denoted as HD_M^I , where specific values of M and I can be used. For instance, HD_{05}^{01} represents the dose where 1% of the population would be subject to an effect of 5% (or more) reduction in red blood cells (if red blood cells was the critical effect). Put another way, 99% of the population would be subject to less than 5% RBC reduction. The target human dose is not known exactly, and its estimate may be presented as an uncertainty range from the lower to the upper confidence limit. The lower confidence limit of the target human dose is defined by IPCS as the probabilistic health-based guidance value.

APROBA

In APROBA the individual uncertainties in each aspect of the hazard characterization are combined by assuming that all uncertainties (e.g. in dose-response data, interspecies differences, intraspecies differences) can be described by independent lognormal probability distributions. The approach is termed Approximate PROBABilistic Analysis, as some of the uncertainties might be better quantified by slightly different distributions, which would require the more time-consuming Monte Carlo sampling in a fully probabilistic risk analysis. The approximate approach based on lognormal distributions is much quicker and easier to apply, and simulations indicate that the resulting confidence bounds are a reasonable approximation to those obtained in the fully probabilistic approach (WHO-IPCS, 2014). The final outcome of APROBA is a confidence interval for the target human dose, the HD_M^I , represented by a confidence interval.

A user guide for APROBA can be found in WHO-IPCS (2014). This document also provides an extensive discussion of the methodology of probabilistic hazard characterization, including a justification of the default uncertainty distributions. For a more comprehensive discussion of probabilistic hazard characterization see Chiu and Slob (2015).

APROBA-Plus

APROBA-Plus is an extension of APROBA with an additional worksheet for entering the information on exposure in terms of an uncertainty range. This may be a confidence interval on some exposure characteristic (often a given exposure percentile of the human population), as reported in the literature, or an uncertainty range based on expert judgment. This will be further discussed in the next two sections.

In the same additional worksheet, the probabilistic results from both the hazard characterization and the exposure assessment are combined in a single graph, providing a visualization of the potential health risks. The y-axis of the plot represents the target human dose, the x-axis the exposure in the highly exposed human subpopulation (e.g. 95th percentile). The (approximate) confidence intervals for these two parameters in both

directions are plotted as a cross in the plot, with an ellipse around it. The latter approximately reflects the two-dimensional confidence region for the combination of the target human dose and the exposure percentile.

Types of available exposure data

The exposure of individuals in a population varies, e.g. because of varying habits in consumption or product use. Hence, the exposure of a population may be characterized by a statistical distribution. Depending on the available information and method used, the estimate of the exposure (as well as its variability among individuals) may be less or more uncertain.

Low tier exposure estimates usually are not very realistic estimates of exposure. They are designed to overestimate the true exposure of (most of) the population. For example, the exposure of a (possibly hypothetical) extreme individual is estimated, or, the highest value from all available exposure data is selected. Low tier assessments generally provide only one value for the entire population without further quantitative specification of the uncertainties. Higher tier exposure estimates are typically based on relatively rich datasets, with adequate statistical analysis. The exposure may then be reported as one or several percentiles of the observed exposure distribution, for example the 50th, 95th or 99th percentiles. In addition, statistical methods can provide confidence intervals around each of the reported exposure percentiles.

Entering exposure information into APROBA-Plus

In practice, the user of APROBA-Plus will use available, existing exposure assessments, which might be a lower or a higher tier assessment for the chemical considered. When the user of APROBA-Plus is of the opinion that the available low tier exposure assessment was not sufficiently conservative or that the available higher tier exposure assessment did not include particular uncertainties, then extra uncertainty ranges can be set by expert judgement. A detailed overview on the possible uncertainties encountered in (existing) exposure assessments can be found elsewhere (e.g. EFSA, 2006; Kettler et al., 2015; WHO-IPCS, 2008). This additional uncertainty will be plotted distinguishably from the uncertainty as reported in the available exposure assessment (see next section).

Obviously, the analyst should ensure that the exposure is entered in the same unit as used for the point of departure (PoD) in the hazard characterization sheet, to allow for a meaningful comparison between human dose and exposure.

Currently, four different exposure estimates (e.g. related to different subpopulations or data sources) can be entered into the exposure table of APROBA-Plus. Each of them requires a brief label, e.g. indicating the (sub)population and the associated percentage of the population the exposure information relates to. This will be used in the legend of the final plot. The exposure is characterized by its lower and upper confidence limit (LCL and UCL), representing the uncertainty for the exposure percentile considered. Although the percentage associated with that percentile needs to be entered in the sheet (in the cells below "INPUTS"), the coverage related to the uncertainty in terms of confidence intervals (e.g. 90%- or 95%-CI) are in many cases not exactly known (they may not be reported in the

available exposure assessment used). Preferably, the confidence level associated with the exposure interval should be similar to that used for the HD_M^I ("probabilistic coverage goal" in the first sheet). The assessor should ensure that the exposure confidence interval reported in the assessment document provides sufficient coverage to be able to conclude whether the protection goals are met or not in the final outcome of APROBA-Plus. Therefore, the assessor needs to make a judgement on the coverage of the reported values of the exposure (interval), and, if this coverage is considered too low, expand the interval based on expert judgment to reach the required coverage. Clearly, the coverage cannot be precisely quantified, and it needs to be assumed that it has approximate coverage, or a coverage range (EFSA, in prep.; Mastrandrea et al., 2010).

Comparison of hazard and exposure

Once the inputs for evaluating the hazard and the exposure uncertainties have been entered in APROBA-Plus the uncertainty range for the HD_M^I is graphically compared with the uncertainty range for the exposure.

Figure 1 shows an example of the final plot, with the exposure uncertainty on the x-axis and the HD_M^I uncertainty on the y-axis. The HD_M^I is depicted as a solid (vertical) line representing its confidence interval. The confidence interval of the exposure percentile is presented as a solid (horizontal) line representing data-based uncertainties, while the dashed extension indicates the additional uncertainties that were based on expert judgment (if applicable). The two lines intersect at the midpoint of the HD_M^I range and the midpoint of the (reported) exposure range (both on log-scale). The vertical line indicating the human exposure is plotted at the midpoint of the (reported) LCL and UCL rather than the additional (dashed) LCL and UCL. This is a matter of choice, but the ellipses will in general be similar in both options.

The overall uncertainty is represented by the ellipse that is drawn around the plotted confidence interval cross. This ellipse indicates where the combination of the *true* HD_M^I and *true* exposure characteristic might be, given the information available.

The values that were entered for the incidence goal in the target human dose (HD_M^I), as well as on the (approximate) percentage related to the exposure percentile are reported in the legends in this plot.

Here it should be noted that the y-axis relates to a subpopulation that is sensitive in terms of toxicokinetics and/or toxicodynamics, while the x-axis relates to a subpopulation that is highly exposed. Imagine the hypothetical situation of a complete lack of uncertainty, where we knew the $HD_M^{0.05}$ and the 95th exposure percentile (i.e. both related to a 5% subpopulation) exactly. If this 95th exposure percentile were greater than this $HD_M^{0.05}$, the percentile of the human population at risk would be smaller than 5%, as the most sensitive people are not necessarily the same people as those with highest exposure. In that sense, the results from APROBA-Plus will be conservative. Meanwhile, it is visualized that two subpopulations are considered by plotting both the HD_M^I and the exposure. This hypothetical example also shows that it is not useful to divide the uncertainty distribution for the exposure percentile by the uncertainty distribution of the HD_M^I , analogous to the

calculation of a risk ratio. The subpopulations in the numerator and denominator would not relate to the same groups of individuals, and the resulting ratio would only allow for a conservative estimate. However, the differences between both subpopulations may be overlooked and the main sources of uncertainty cannot be identified anymore, because a single metric is reported. The idea of taking a ratio of hazard and exposure values is only meaningful if this is done on the level of the individual, as it does not make sense to compare the critical dose of one individual with the exposure in another. A full IPRA (e.g. van der Voet and Slob, 2007) does allow for a probabilistic assessment on the individual level, by dividing the two underlying variability distributions by each other, while taking the uncertainty in those distributions into account.

These considerations are part of the reasons that APROBA-Plus shows both the uncertainty in the target human dose and the uncertainty in exposure separately, and compares them in a plot, rather than reduce them to a single dimension by division. Yet another reason is that in this way it is directly visible how much larger the uncertainty in one direction is compared to the other direction. If refinement of the risk assessment is needed, the graph provides visual guidance on what type of information (hazard, exposure, or both) is likely to provide the greatest improvement (Embry et al., 2014), as illustrated below. When it is considered more effective to reduce the uncertainty in the target human dose, the first (original APROBA) sheet includes further information on the relative contributions from the various aspects to the overall uncertainty in the HD_M^I .

Because of the reasons just discussed, and because of the fact that the exposure uncertainty entered in the spreadsheet usually represents a fairly rough estimate without a defined confidence level, APROBA-Plus does not draw the line where exposure equals the HD_M^I as that would suggest a level of precision that is not realistic. Instead, the distance to the identity line is indicated by different shades of green (exposure < HD_M^I) and red (exposure > HD_M^I). When the whole ellipse is clearly located in the green area of the plot, then the (true) exposure most likely is below the (true) HD_M^I , and the protection goals are most likely to be met (where “most likely” means close to the assumed coverage). When the ellipse is completely located in the red area, this indicates that the exposure most likely exceeds the HD_M^I , and the protection goals (in terms of M and I) would not be met. In case the ellipse is located in both the green and the red area a conclusive answer cannot be given, and various options may be considered. When most of the ellipse lies in the green area, the odds are that the exposure is smaller than the HD_M^I , but this cannot be stated with the assumed confidence level (as a small fraction of the ellipse lies in the red area). In that case a full probabilistic risk assessment (IPRA) might be considered. IPRA gives a more precise evaluation of uncertainties, while it avoids the conservative comparison of the sensitive subpopulation to the highly exposed subpopulation, as is in fact done in the APROBA-Plus plot. So, there is a possibility that after a full IPRA the protection goals turn out to be met (with the required confidence). However, a full IPRA will not dramatically change the results from APROBA-Plus, and when a substantial part of the ellipse is in the red area, a full IPRA would probably turn out to be inconclusive as well. In this case, it might be considered to obtain additional data right away to reduce the uncertainty either in the exposure or in the HD_M^I . Here, it may be taken into account which specific uncertainties

contribute most to the overall uncertainty; this information is shown by the APROBA-Plus output.

Datasets used

We first apply APROBA-Plus to three compounds purposely selected for illustrating three distinct possible outcomes. To further explore the applicability of APROBA-Plus for routine risk assessments in general, we randomly drew 40 substances from the Dutch Quality Programme of Agricultural Products database (van Dooren et al., 1995). Twenty pesticides and twenty substances from the subgroups metals, dioxins/PCBs, flame retardants and veterinary drugs were drawn, aiming at some variation in types of chemicals.

From these 40 substances 24 were excluded from analysis, for the reasons specified in the supplementary material (Table S11). Eight of the substances were part of a Common Assessment Group (CAG) and a risk assessment of the single substance was considered pointless. When data are available for the entire CAG, an assessment with APROBA-Plus may, under specific conditions, be possible, but this was considered beyond the scope of the present study. Four substances were not included for being accumulating or carcinogenic substances. Probably, APROBA-Plus could, under specific conditions, be used for accumulating substances, but this would require some more methodological scrutiny. The carcinogenic substances were excluded because we only used reported PoDs in the present study, while a probabilistic hazard characterization for carcinogens according to the IPCS methodology would require additional dose-response information (WHO-IPCS 2014; Chiu and Slob 2015). For one substance (magnesium) information on a point of departure for hazard characterization could not be found, while for nine substances information on exposure was completely lacking. In another two substances information on both data types was lacking. For the remaining 16 substances the available quantitative information was rather limited, but sufficient for applying APROBA-Plus.

ILLUSTRATIVE CASE STUDY: NIVALENOL

In 2013 EFSA assessed the risk of dietary nivalenol exposure (EFSA, 2013d). This assessment will be used to illustrate how to enter the inputs in APROBA-Plus.

Hazard characterization

Table 1 shows the APROBA Excel sheet (first part in APROBA-Plus) requiring inputs regarding the (critical) endpoint, the type of study in which the critical effect was observed and the protection goals defining the target human dose (HD_M^I). All grey cells need to be filled out. For some parameters common (default) values are suggested (see third column). For example, based on the input of “rat” as test species, the tool suggests 0.40 kg as a reasonable default. The user may use the tool’s suggestion or enter another value if more specific information is available. For an exhaustive description of the hazard characterization tables (Tables 1 to 4) and guidance on selecting the appropriate input, the reader is referred to WHO-IPCS (2014).

EFSA identified the decreased white blood cell (WBC) count in an oral 90-day rat study as the critical effect of nivalenol. The BMD05 90%-confidence interval (i.e. the BMDL and

BMDU) provided by EFSA ranges from 0.35 to 0.76 mg/kg bw/day. In Table 1, the protection goals (M and I) are set to 5% and 1%, respectively. The coverage (WHO-IPCS, 2014), reflecting the probability that the probabilistic reference dose (RfD) is small enough (i.e., smaller than the true HD_M^I), is set to 95%. Note that the coverage is in fact a one-sided confidence level.

In their assessment EFSA applied an assessment factor of 100 for inter- and intra-species differences. In addition, they decided to use two additional assessment factors: a factor of 2 for extrapolation from subchronic to chronic study duration, and a factor of 1.5 due to the limitations of the data that were available on reproductive and developmental toxicity of nivalenol. Applying this overall deterministic assessment factor of 300 to the BMDL results in a deterministic RfD of 1.2 µg/kg bw/day (see last row in Table 1).

In Table 2, the probabilistic inputs related to various hazard characterization aspects are listed. In accordance with EFSA, adjustments are made to account for inter- and intraspecies differences, subchronic to chronic extrapolation and limited data on reproductive and developmental toxicity. For interspecies, intraspecies and duration extrapolation APROBA provides data-based default uncertainty distributions. Given the lack of chemical-specific information, these defaults are applied. Note that the default distribution for exposure duration in APROBA assumes larger potential differences between chronic and subchronic BMDs than the factor of two used in the EFSA opinion (namely, ranging from 0.50 to 8.0, with 90% confidence; see (WHO-IPCS, 2014)). For the limited reproductive and developmental toxicity data EFSA applied an assessment factor of 1.5, which is considered here as the upper (95%) confidence limit. The latter is interpreted as follows: the (true) BMD for the most sensitive reproductive effect might be a factor of 1.5 lower than the subchronic BMD, but it is unlikely to be even lower than that. On the other hand, it can be hypothesized that the reprotoxic BMD is higher than the subchronic BMD. In that case, the critical effect remains decreased WBC count as observed in the subchronic study and the BMD confidence interval would not need any adjustment for data limitations. Therefore, the value of 1.0 will be used as the lower (5%) confidence limit of this adjustment factor, allowing the adjustment factor to be smaller than one, with small probability. In APROBA this cannot be avoided, as only (non-truncated) lognormal distributions are allowed. However, the consequences for the final confidence interval of the HD_M^I will be minimal in this case, given the relatively small value of the upper bound of 1.5.

Table 3 shows the output provided by APROBA based on the input in Tables 1 and 2. Here, the lower and upper bounds of the target human dose (HD_M^I) follow from probabilistically combining the uncertainty distributions for the PoD and all applied adjustment and extrapolation factors, as entered in Table 2. The resulting human dose related to a 5% decrease in WBC count in 1% of the population (HD_{05}^{01}) lies between 0.46 and 51 µg/kg bw/day (with 90% confidence). The uncertainty in this HD_M^I is a factor of 110 ($= 51/0.46$). The lower confidence bound of the HD_M^I , i.e. 0.46 µg/kg bw/day, is designated as the probabilistic RfD, reflecting the human dose at which, with 95% coverage (confidence), 1% of the population would have a decreased WBC count of 5%. Or, put another way, the human dose where 99% of the population would be subject to a smaller than 5% decrease in WBCs.

The contribution of each hazard characterization aspect to the overall uncertainty in the HD_M^I is listed in Table 4. It shows that the uncertainties related to intraspecies differences and those to exposure duration contribute most, while the PoD (in this case BMD) uncertainty is only minor.

Exposure assessment

In the EFSA opinion on nivalenol the exposure is given for multiple subpopulations, ranging from infants to the very elderly. For each of these subpopulations EFSA reports the mean and 95th percentile dietary exposures as lower and upper bounds across various countries. In Table 5 the selected inputs related to the exposure are listed (the 95th percentiles of the pertinent populations). The lower bound of the lowest country and the upper bound of the highest country are considered to very likely cover the actual 95th percentile of the total (European) population, and are taken as the overall uncertainty range of exposure (LCL and UCL) in the Excel sheet. Two subpopulations are selected: adults and toddlers. The adults are selected because this is the largest subgroup, and the toddlers because they have the highest exposure.

As mentioned above, the analyst may include additional uncertainties, for instance uncertainties that were not taken into account in the values taken from the literature, or when particular reported values are considered to be not conservative enough (see *expert opinion on limits* in Table 5). In the EFSA report several uncertainties are listed considering the exposure assessment, which are not accounted for in the exposure assessment. To cover these uncertainties in the current analysis it is assumed that the LCL and UCL could be up to a factor two lower and higher than the reported LCL and UCL, respectively. Entering a deterministic value (in the last column of Table 5) is optional. When a value is entered here, the deterministic exposure will be plotted against the deterministic RfD (triangle and diamond symbols in Figure 2) to illustrate the conservatism of the deterministic risk ratios. In this case of nivalenol, the reported upper bound was considered a reasonable (worst-case) deterministic exposure.

In Figure 2 the HD_M^I of nivalenol is graphically compared with the exposure in the two subpopulations (adults and toddlers), by plotting the uncertainty range of the HD_M^I on the y-axis and the uncertainty range of the exposure (95th percentile) on the x-axis.

RESULTS AND DISCUSSION

The results for the three illustrative, food related, case studies nivalenol, tropane alkaloids and Allura Red are discussed in some detail, while results for the 16 additional substances will be discussed in a more global way. The intention is to illustrate how risk managers may benefit from presenting the outcomes of a risk assessment as uncertainty ranges rather than deterministic (conservative) point estimates. The underlying methods of performing a probabilistic risk assessment have been described and discussed elsewhere (e.g., van der Voet and Slob, 2007; WHO-IPCS, 2014).

The three illustrative case studies

The results for the first three case studies are shown in Figure 2 (nivalenol) and Figure 3 (tropane alkaloids and Allura Red). The APROBA-Plus inputs for nivalenol were discussed above, those for the other two are reported in the Supplemental Material. These three graphs illustrate the three main types of possible outcomes from APROBA-Plus:

- For nivalenol (Figure 2) the uncertainty ellipse for the combination of target human dose and exposure is completely in the green area (where exposure is lower than the target human dose) and the conclusion would be that it is very likely that the protection goals are met.
- For the tropane alkaloids (Figure 3, left panel) the largest part of the uncertainty ellipse lies in the red area, with a smaller part in the green area. Thus, the odds are that the protection goals are not met, but it remains possible that they are. Performing a full IPRA will probably not lead to a dramatically different picture, and result in an inconclusive outcome as well, given the magnitude of the uncertainties indicated by APROBA-Plus. One option would be to re-analyze the dose-response data for the critical effect by the BMD approach rather than by the NOAEL, as this was found to be a major source of uncertainty in the estimated target human dose (see Table S4). If the dose-response data for the critical effect can be made available, this would be the easiest (cheapest) option for a refined assessment. However, whatever the effect of the BMD analysis, Figure S1 clearly shows that, in order to move the ellipse out of the red area, it is necessary to find better exposure data in this case, as the current uncertainty is large and based on expert judgment only, due to the lack of data.
- For Allura Red a small part of the uncertainty ellipse is located in (or close to) the red area. Note that this part combines highly exposed individuals with sensitive individuals, which do not necessarily need to be the same individuals. A full IPRA would take that into account, which might lead to a conclusive outcome (protection goals met) for this case. Therefore, performing a full IPRA would be a good option here. However, given the large uncertainty contribution of using a NOAEL in this case, it may be easier to first run a BMD analysis on the original dose-response data (if available). This may be expected to result in a smaller uncertainty in the vertical direction, with the possible consequence that the complete ellipse ends up in the green area. Yet, just as in the previous case, it should be kept in mind that the exposure uncertainty for one of the two subpopulations (adults) was based on expert judgment. For this reason, argumentation is needed that the risk assessment can be fully based on the other subpopulation (children).

In addition, these illustrative examples show that the conservatism of the deterministic risk ratios, which are indicated by the triangles and diamonds, varies. For example, for the tropane alkaloids the ellipses intrude much deeper into the red area than their corresponding deterministic ratios, implying that in this case the risk ratios might underestimate the true risk ratio. For nivalenol (Figure 2) the risk ratios are outside their respective ellipse, so in this case the risk ratios are conservative, and possibly more conservative than desirable.

These examples show that the deterministic risk ratio is not necessarily conservative, as usually assumed, even though they are in most cases (see plots for additional 16 compounds in Supplemental Material). Finally, the examples illustrate that prioritizing compounds based on deterministic risk ratios is highly unreliable, as some risk ratios are more conservative than others.

The additional 16 compounds

The underlying input information as used in the APROBA-Plus tables, a summary of the results and all ellipse-plots for each of the 16 additional substances can be found in Tables S14 and S15 and in Figure S3 in the Supplemental Material.

The critical toxicological studies were generally not available, and for this reason the information required for the hazard characterization was usually obtained from summaries of the toxicity studies. The following issues were encountered regarding the hazard characterization inputs of APROBA-Plus:

- For various substances (ametoctradin, bentazone, fludioxonil, lufenuron, fluroxypyr and chromium) the PoD related to both continuous and quantal endpoints (i.e. various endpoints resulted in the same NOAEL/LOAEL), or it was unclear if the critical endpoint was continuous or quantal. In those cases we always assumed that the critical endpoint was continuous as this tends to result in the lowest probabilistic reference dose.
- Sometimes the description of the biological nature of the critical effect was not available or not very specific (e.g. bentazone-*acute* and ametoctradin), which made interpretation of the results difficult.
- Body weights of the experimental animals were never available. For rat and mouse the default body weights provided by APROBA-Plus were used. A body weight of 11 kg was used for dogs.
- The 1-year dog studies were considered as subchronic and consequently an adjustment factor for subchronic-to-chronic duration extrapolation was applied.
- In APROBA-Plus a NOAEL based on continuous data is by default extrapolated to a BMD relating to 5% change in response. However, for some continuous endpoints a larger change in response may be considered relevant. This issue was encountered in the assessment of phoxim, where the critical effect was acetylcholinesterase activity (AChE) in brain. Decreases in AChE below 10% (U.S.) or below 20% (Europe) are usually considered nonadverse. Therefore, an additional adjustment factor (smaller than one) was applied in this case (see Table S13).

Except for chromium, exposure estimates were never reported as confidence intervals. Therefore, the reported point estimates were translated into the confidence limits (LCL and UCL) based on (our) expert opinion. We encountered the following situations:

- For several substances the reported exposure is based on food concentrations which are set to the maximum residue level (MRL), together with an assumed

(daily) food consumption pattern. According to limited (unpublished) data the measured long-term exposure can be up to a factor 100 lower than a calculated chronic exposure based on MRLs. On the other hand, the measured acute exposure can be up to a factor 100 higher compared to a calculated acute exposure based on MRLs. These factors were used in our evaluations of the uncertainties.

- Measured concentrations in food may be below LOQ. As a conservative assumption these concentrations below LOQ may be set to the value of LOQ to calculate the exposure. According to limited (unpublished) data chronic exposures based on the scenario in which concentrations below LOQ are set to zero could be up to a factor of 10 times lower compared to exposures based on the conservative scenario. This value was used in our evaluations of the uncertainties.
- In the available exposure assessments performed by EFSA a default (PRIMo) diet is generally used to derive the exposure. We assumed that the default diet could result in a conservative (i.e. up to ten times too high) estimate of a realistic high (e.g. 95th percentile) exposure.
- For some substances an acute exposure estimate was available while the relevant toxicological effect only occurs after repeated exposure. We assumed that the chronic exposure was at most a factor of 10 times lower than the reported acute exposure.
- In one specific case (chromium) the authors report confidence intervals with their exposure estimates, but at the same time they discussed some additional uncertainties about these values. The authors indicate that the exposure may be over- or underestimated. We assumed that the LCL and UCL is at most an arbitrary factor of 5.0 lower and higher than the reported values.

Table S14 (Supplemental Material) lists, for each substance, the uncertainties that were accounted for when setting the exposure limits by expert judgement.

The total number of analyses was 20, as for some of the 16 compounds both chronic and acute exposure was considered. Table S14 summarizes the results based on the final APROBA-Plus plots for each compound (see Figure S3 for the individual plots). The table indicates whether the protection goals are likely to be met for each substance tested. In case this is inconclusive, suggestions are made on possible next steps to get more conclusive results.

Twelve out of the 20 cases (very likely) meet the protection goals, assuming that the uncertainty is adequately evaluated. In five other analyses (bentazone-*acute*, colistin, dihydrostreptomycin, phoxim and ractopamine) the ellipses intrude the red area, but most of the ellipse is in the green area. The following next steps, or a combination thereof, may be considered in such situations.

- When the PoD was a NOAEL, and the PoD uncertainty was found to be non-negligible (as indicated in the APROBA-Plus table with relative contributions to

the overall uncertainty, see e.g. Table 4) a BMD analysis would be the easiest next step (if the dose-response data are available).

- Given that only a relatively small part of the ellipse lies in the red area, a full IPRA may be considered as another next step, as it more accurately integrates the hazard and exposure uncertainties, possibly resulting in a smaller overall uncertainty.
- If neither of these options provides a conclusive result, some of the uncertainties would need to be reduced by generating more or better data. Here, the table in APROBA-Plus summarizing the relative contributions of the various uncertainties may help in determining which uncertainties deserve first attention. In these five cases, the exposure uncertainties are relatively large, and, given that they are often largely based on expert judgment with limited data, generating exposure data would be an obvious option. In other cases, another toxicity study may also be an option for substantially reducing the overall uncertainty.

For three cases (captan-*acute*, mepiquat-chloride-*acute*, heptachlor) the ellipse did not intrude the red area but was close to it, while the effects involved were clearly adverse. In that case, the conclusion that the protection goals are met may be questioned, as the nature of the effects might call for stricter protection goals (lower *I* or lower *M* than the defaults used). One may consider to repeat the analysis with a lower incidence goal (changing *M* is not possible when NOAELs are used). This will shift the ellipse downwards, possibly into the red area. To illustrate the effect of lowering the incidence goal the analysis for captan (acute exposure) was repeated while setting an incidence goal of 0.01 and 0.0001%, i.e. 1 in 10,000 and 1 in a million. As Figure S4 shows, the entire ellipse shifts down while the uncertainty of the HD_M^I increases. The latter is a logical consequence of the fact that smaller incidences are increasingly more uncertain. Further, it should be kept in mind that in reality the uncertainties are even larger, as the outcomes of the analysis are increasingly more sensitive to the assumptions made (e.g. the lognormality approximation of the intraspecies variation). When the ellipse, after setting a lower incidence goal, turns out to enter the red area, the same options of further action may be considered as just discussed in the previous paragraph.

The 20 APROBA-Plus plots also show the deterministic RfDs plotted against the reported (deterministic) exposure values. In most of the 20 plots the deterministic point is close to the border of the ellipse. However, in some cases it is inside the ellipse, indicating that the deterministic assessment was not that conservative, and in some cases it is outside, and below or at the right side of the ellipse, indicating that the deterministic assessment was overly conservative. Figure 4 shows two examples where the deterministic point is close to the unity line, representing a risk ratio of close to one in both cases. Based on a deterministic assessment, the two compounds would be regarded as 'ties' in risk ranking. However, the ellipses show that the distance between target human dose and exposure percentile might in reality be much larger for novaluron (right panel) than for imidacloprid (left panel), as a large part of the ellipse is more remote from the light-colored diagonal area. Therefore, there would be less reason of concern for novaluron than for imidacloprid, and with the output from APROBA-Plus it actually is possible to rank both these compounds.

The APROBA-Plus plots for the 20 cases (see Figure S3) illustrate another use of APROBA-Plus: the uncertainty ellipses may be used to rank the risk of substances taking the following considerations into account:

- i. what fraction of the ellipse lies in the green/red area,
- ii. how remote is the left upper part of the ellipse from the light-colored diagonal (as illustrated in Fig. 4),
- iii. how adverse is the effect associated with the PoD (i.e. the nature of the effect plus the value of M).

Since the first two considerations are based on visual comparisons, it is of course needed that the plots use the same scales. In principle, ranking should be based on the combination of all three considerations, but it may be difficult to do so in a more or less consistent way. Possibly, some formal method could be developed in the future to support this, but for the time being a simplified approach might be considered. For example, one might first rank them according to the first consideration, then refine that ranking based on the second and third considerations.

It is re-emphasized that these three considerations should also be taken into account in any single risk assessment. This is an important extension to the approach of calculating a risk quotient based on values assumed to be conservative, where the level of conservatism is unknown, both in terms of coverage (first consideration) and in terms of the range of uncertainty in the actual distance between target human dose and exposure (second consideration). While the third consideration could equally well be taken into account in the deterministic risk quotient, there appears to be a tendency to focus on the value of the risk quotient and ignore the third consideration. In APROBA-Plus, the type of effect and the value of the associated effect size (M) is made visual in the final plot. Furthermore, the percentages of the subpopulations are made explicit. All these different dimensions are important in any risk assessment, and keeping the information on all those dimensions visible is better than aiming to reduce the multidimensional problem of risk assessment into one single dimension, as in a single risk quotient.

APROBA-Plus summarizes the results of the risk assessment as a plot of the (uncertainty in) the target human dose against the (uncertainty) in a particular exposure percentile. It might be tempting to draw the identity line in the plot, and check if the uncertainty ellipse intersects with that line. If it does not (and the ellipse is in the green area) the protection goal would be achieved. However, this is not an appropriate approach for various reasons. First, the fractions of the population relating to the target human dose and the exposure estimate may not be the same. Further, even if they are, the most sensitive people are not necessarily the highest exposed people. Therefore, a 95th percentile of the exposure distribution exceeding the target human dose for the 5% most sensitive individuals does not mean that 5% of the population is “at risk”. All that can be said is that it will be less than 5%, but not how much. Hence, the identity line cannot be interpreted in a way that is useful for risk managers. Second, quantification of uncertainties is dependent on the underlying data and/or expert judgment, i.e. the quantification of uncertainties itself is subject to (secondary) uncertainty (WHO-IPCS, 2014). Better evaluation of secondary uncertainties and developing

methodology for incorporating secondary uncertainties in risk assessment are desirable future developments, but beyond the scope of this paper. Nonetheless, it is clear that quantified uncertainties should always be considered as an approximation, making a sharp criterion like “Does the uncertainty ellipse intersect the identity line or not?” inappropriate.

Instead, the assessor should take a more global view on the results, and inspect the whole ellipse. When the other side of the ellipse is remote from the identity line, this indicates that the true target human dose could very well be far away from the exposure. If, however, the other side is close to the identity line as well, there should be more reason of concern. Finally, it is important to keep the specific target human dose into account: What type of effect and what associated effect size was it based on? For example, the same ellipse for a target human dose relating to mild liver effects does not have the same impact as when it related to 10% malformations as the underlying effect. For these reasons, APROBA-Plus does not draw the identity line in the final plot, but instead uses a light-colored range in that area.

The problems related to the identity line as just discussed at the same time hold for the risk quotient (risk characterization ratio). Therefore, presenting a distribution of the risk quotient (risk characterization ratio) is not a useful refinement, compared to plotting hazard against exposure. Instead, leaving the uncertainty in the target human dose and the uncertainty in the exposure estimate as separate information in the plot has the advantage that it can be directly seen which uncertainty is larger. Thus, a more informed decision can be made in how to best spend available resources, while taking the costs of generating various types of new data into account.

CONCLUSIONS

The results from the APROBA-Plus tool may be considered as an approximation of a fully integrated probabilistic risk assessment (IPRA). The strength of the APROBA-Plus tool is that, when the hazard and exposure results have been made available in a deterministic assessment, it takes very little time to fill out the APROBA-Plus input tables, after which a picture is instantly created that visualizes the uncertainty ranges of the target human dose and human exposure. As also noted previously (Embry et al., 2014), a tool such as APROBA-Plus may be very useful as a quick approach of quantitatively evaluating uncertainties, and could easily serve as a standard extension of routine risk assessments.

The three illustrative case studies and the application to another 16 compounds showed that APROBA provides more information than the current approach of comparing one exposure value to one health-based guidance value. For the 16 compounds the available quantitative information was rather limited, but sufficient for applying APROBA-Plus. It should be noted that limited information should be an impetus for performing a probabilistic risk assessment rather than avoiding it: the larger the uncertainties, the more important to evaluate them and take them into account. Only in situations of extreme lack of information, such as no PoD or no exposure information available at all, APROBA-Plus cannot be applied, but this equally holds for any non-probabilistic risk assessment. The examples illustrated how the APROBA-Plus outcomes may help in making rational decisions on next steps in the risk assessment of

a specific chemical when no conclusive answer could be given based on the information available at that point. In general, a first thing to consider as a next refinement step is performing a BMD analysis, and replace the NOAEL by a BMD estimate, as this will in general reduce the uncertainty in the target human dose (WHO-IPCS, 2014). Another option would be to perform a full IPRA, in particular when only a relatively small part of the ellipse enters the red area. And finally, one might consider to generate new data, in which case the results from APROBA-Plus are helpful by indicating which uncertainties are relatively large.

Another potential application of APROBA-Plus is to use the resulting visualizations of uncertainty to roughly rank substances based on the three considerations defined above. In doing so, not only the fraction of the ellipse entering the red area should be taken into account, but also the magnitude of the uncertainty as reflected by the size of the ellipse, and the severity of the critical effect. Such a ranking may help risk managers to focus on those chemicals that would need first attention. Once they are identified, it may be decided case-by-case what action would be most effective, taking into account all additional relevant information, including the costs for improving the probabilistic assessment, the costs for generating new data, and the costs (in a general sense) of specific mitigating measures.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

This work was funded by the Netherlands Food and Consumer Product Safety Authority. The authors would like to thank Myrna Kooi for gathering the information on hazard and exposure of the 40 randomly drawn substances.

List of abbreviations

AF	Assessment Factor
APROBA	Approximate probabilistic approach
BMD	Bench Mark Dose
BMDL	Lower confidence limit of the BMD
BMDU	Upper confidence limit of the BMD
BMR	Bench Mark Response
EFSA	European Food Safety Authority
IPCS	International Programme on Chemical Safety
IPRA	Integrated Probabilistic Risk Assessment
HD_M^I	Human dose with magnitude [<i>M</i>] of the effect in individuals and fraction [<i>I</i>] of population protected from that magnitude of effect
LCL	Lower Confidence Limit

LOQ	Limit of Quantification
NOAEL	No Observed Adverse Effect Level
PoD	Point of Departure
RfD	Reference Dose
TA	Tropane alkaloid
UCL	Upper Confidence Limit
WBC	White Blood Cell (count)
WHO	World Health Organization

References

- Bokkers, BGH., Bakker, MI., Boon, PE., Bos, P., Bosgra, S., Van der Heijden, GWAM., Janer, G., Slob, W., Van der Voet, H. The practicability of the integrated probabilistic risk assessment (IPRA) approach for substances in food. 2009. <http://www.rivm.nl/bibliotheek/rapporten/320121001.pdf>
- Chiu WA, Slob W. A Unified Probabilistic Framework for Dose-Response Assessment of Human Health Effects. *Environmental health perspectives*. 2015; 123:1241–1254. [PubMed: 26006063]
- EFSA. Guidance of the Scientific Committee on a request from EFSA related to Uncertainties in Dietary Exposure Assessment. Request No EFSA-Q-2004-019. *EFSA Journal*. 2006; 438:1–54.
- EFSA. Scientific Opinion on risks for animal and public health related to the presence of nivalenol in food and feed. *EFSA Journal*. 2013; 11:3262.
- EFSA. Revised DRAFT Guidance on Uncertainty in EFSA Scientific Assessment. Question number: EFSA-Q-2013-00738. in prep. Version of 21 March 2016.
- Embry MR, Bachman AN, Bell DR, Boobis AR, Cohen SM, Dellarco M, Dewhurst IC, Doerr NG, Hines RN, Moretto A, Pastoor TP, Phillips RD, Rowlands JC, Tanir JY, Wolf DC, Doe JE. Risk assessment in the 21st century: roadmap and matrix. *Crit Rev Toxicol*. 2014; 44(Suppl 3):6–16. [PubMed: 25070414]
- Kettler S, Kennedy M, McNamara C, Oberdorfer R, O'Mahony C, Schnabel J, Smith B, Sprong C, Faludi R, Tennant D. Assessing and reporting uncertainties in dietary exposure analysis: Mapping of uncertainties in a tiered approach. *Food Chem Toxicol*. 2015; 82:79–95. [PubMed: 25890086]
- Mastrandrea, MD., Field, CB., Stocker, TF., Edenhofer, O., Ebi, KL., Frame, DJ., Held, H., Kriegler, E., Mach, KJ., Matschoss, PR., Plattner, GK., Yohe, GW., Zwiers, FW. Guidance Note for Lead Authors of the IPCC Fifth Assessment Report on Consistent Treatment of Uncertainties. Intergovernmental Panel on Climate Change (IPCC). 2010. Available at <<http://www.ipcc.ch>>
- Pastoor TP, Bachman AN, Bell DR, Cohen SM, Dellarco M, Dewhurst IC, Doe JE, Doerr NG, Embry MR, Hines RN, Moretto A, Phillips RD, Rowlands JC, Tanir JY, Wolf DC, Boobis AR. A 21st century roadmap for human health risk assessment. *Crit Rev Toxicol*. 2014; 44(Suppl 3):1–5.
- Slob W, Bakker MI, Biesebeek JD, Bokkers BGH. Exploring the uncertainties in cancer risk assessment using the integrated probabilistic risk assessment (IPRA) approach. *Risk Anal*. 2014; 34:1401–1422. [PubMed: 24766324]
- van der Voet H, Slob W. Integration of probabilistic exposure assessment and probabilistic hazard characterization. *Risk Anal*. 2007; 27:351–371. [PubMed: 17511703]
- van Dooren, MMH., Boeijen, I., van Klaveren, JD., van Donkersgoed, G. Conversie van consumeerbare voedingsmiddelen naar primaire agrarische producten (Conversion of Consumed Foods into Raw Agricultural Commodities) (95.17). RIKILT-DLO; Wageningen: 1995. Available at www.rikilt.wur.nl
- WHO-IPCS. Uncertainty and data quality in exposure assessment. Part 1: Guidance document on characterizing and communicating uncertainty in exposure assessment; Part 2: Hallmarks of data

quality in chemical exposure assessment. 2008. WHO-IPCS harmonization project document no. 6 http://www.who.int/ipcs/publications/methods/harmonization/exposure_assessment.pdf?ua=1

WHO-IPCS. Guidance document on evaluating and expressing uncertainty in hazard characterization. 2014. WHO-IPCS harmonization project document no. 11 http://www.who.int/ipcs/methods/harmonization/areas/hazard_assessment/en/

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

Figure 1. Hypothetical example of an APROBA-Plus graph. In this example, the incidence goal related to the HD_M^I is 1%, and the exposure estimate relates to the 95th percentile of the adult population (see legend). See text for further explanation.

Figure 2.

Comparison of nivalenol's HD_M^I relating to an incidence goal of 1% and the 95th percentile exposure of toddlers (black, ellipse on right hand side) and adults (blue) in APROBA-Plus. The blue diamond and black triangle indicate the risk based on the deterministic RfD (of 1.2 µg/kg bw/day) and reported upper confidence limits of exposure of adults and toddlers respectively.

Figure 3.

Comparison of HD_M^I ($I=1\%$ and $M=5\%$) and exposure for the two other illustrative examples (tropane alkaloids, left panel, and Allura Red, right panel) in the APROBA-Plus analysis. In the right plot the two ellipses relate to two subpopulations, in the left plot to two different data sources. The blue diamonds and black triangles indicate the risk ratios of these compounds. For more details, see Supplemental Material.

Figure 4.

Illustration of the additional information from APROBA-Plus as compared to a deterministic risk assessment which are indicated by the marks (triangle, diamond). The deterministic risk ratio in both cases is close to unity (see diamonds) and would be regarded as “ties” in risk ranking. However, the ellipse for novaluron (right plot) extends further into the green area than for imidacloprid (left plot), implying that there is a considerable chance that the distance between exposure and target human dose is greater for novaluron than for imidacloprid. Thus, with the probabilistic approach risk ranking is possible in this case.

Table 1

APROBA(-Plus) inputs related to study, endpoint and protection goals of the hazard characterization of nivalenol.

DESCRIPTION	INPUTS	COMMON VALUE(S)
Endpoint	WBC	Case-specific
Data type	Continuous	Case-specific
Data route	Oral	Case-specific
Study type	Subchronic	Case-specific
Test species	Rat	Case-specific
Body weight test species (kg)	0.40	0.4
Human median body weight (kg)	60	60
Target BMR (= M , user input for BMDLs only)	5%	5%
Population incidence goal (= I)	1%	5%, 1%, 0.1%, 0.01%
Probabilistic coverage goal	95%	95%
PoD type	BMDL	Case-specific
PoD value	350	Case-specific
BMDU (User input for BMDLs only)	760	Case-specific
PoD units	µg/kg bw/day	mg/kg body weight per day
Deterministic overall AF (optional)	300	Case-specific
Deterministic RfD (calculated from other inputs)	1.2	Calculated

Table 2

Inputs related to adjustment, variability and uncertainty of the hazard characterization of nivalenol.

HAZARD CHARACTERIZATION ASPECT		INPUTS	PROVISIONAL VALUE(S)
PoD	LCL	350	Calculated from inputs
(Modelled BMD uncertainty)	UCL	760	Calculated from inputs
NOAEL to BMD	LCL	1.0	1.0
(NOAEL only)	UCL	1.0	1.0
Interspecies scaling	LCL	3.7	3.7
(Allometric for oral)	UCL	5.5	5.5
Interspecies TK & TD^a	LCL	0.33	0.33
(Remaining TK & TD ^a)	UCL	3.0	3.0
Duration extrapolation	LCL	0.50	0.50
	UCL	8.0	8.0
Intraspecies	LCL	2.2	2.2
	UCL	42	42
Other aspect #1	LCL	1.0	1.0
limited repro and dev tox data	UCL	1.5	1.0
Other aspect #2	LCL	1.0	1.0
(Description here)	UCL	1.0	1.0
Other aspect #3	LCL	1.0	1.0
(Description here)	UCL	1.0	1.0

^a toxicokinetics and toxicodynamics

Table 3

Approximate probabilistic analysis outputs of the APROBA hazard assessment of nivalenol.

Standard Confidence Interval			
Target Human Dose (HD_M^I)	LCL (P05)	0.46	µg/kg bw/day
	UCL (P95)	51	µg/kg bw/day
Degree of Uncertainty (Fold Range)			110
Probabilistic RfD 0.46	= Approximate probabilistic HD _M ^I at specified % confidence		
	= Estimate of dose (µg/kg bw/day) at which, with		
	95%	Confidence	
	1%	of the population will have	WBC
	of magnitude		5%

Table 4

Contribution of the various sources of uncertainty to the overall uncertainty in the APROBA hazard assessment of nivalenol.

UNCERTAINTY ANALYSES ASPECT	% contribution to overall uncertainty
PoD	3%
NOAEL to BMD	–
Interspecies scaling	1%
Interspecies TK & TD ^a	22%
Duration extrapolation	35%
Intraspecies	39%
Other aspect #1	1%
limited reproductive and developmental toxicity data	

^a toxicokinetics and toxicodynamics

Table 5

Inputs related to the APROBA-Plus exposure assessment of nivalenol

Enter available expo values, leave others empty									
Exposure unit: $\mu\text{g/kg bw/day}$									
DESCRIPTION	INPUTS	reported exposure		expert opinion on limits		Deterministic exposure			
		LCL	UCL	extra LCL	extra UCL	LCL	UCL	extra LCL	extra UCL
exposure #1	95% adults	0.0011	0.22	0.00055	0.45				0.22
exposure #2	95% toddlers	0.012	0.48	0.0060	0.97				0.48