TITLE

Interpretation of the Margin of Exposure for Genotoxic Carcinogens – Elicitation of Expert Knowledge about the Form of the Dose Response Curve at Human Relevant Exposures

AUTHORS

Alan Boobis[1], Villie Flari[2], John Paul Gosling[2,3], Andy Hart[2], Peter Craig[4], Lesley Rushton[5], Ehi Idahosa-Taylor[5]

[1] Centre for Pharmacology and Therapeutics, Department of Medicine, Imperial College London, UK, Hammersmith Campus, Ducane Road, London W12 0NN

[2] Food and Environment Research Agency, Sand Hutton, YO41 1LZ, York, UK

[3] Department of Statistics, University of Leeds, Leeds, LS2 9JT, UK (since Sep 2011)

[4] Department of Mathematical Sciences, University of Durham, South Road, Durham DH1 3LE, England

[5] Department of Epidemiology and Biostatistics and School of Public Health and MRC-HPA Centre for Environment and Health, Imperial College London, St Mary's Campus, Norfolk Place, London W2 3PG

ABSTRACT

The general approach to risk assessment of genotoxic carcinogens has been to advise reduction of exposure to "as low as reasonably achievable/practicable" (ALARA/P). However, whilst this remains the preferred risk management option, it does not provide guidance on the urgency or extent of risk management actions necessary. To address this, the "Margin of Exposure" (MOE) approach has been proposed. The MOE is the ratio between the point of departure for carcinogenesis and estimated human exposure. However, interpretation of the MOE requires implicit or explicit consideration of the shape of the dose-response curve at human relevant exposures. In a structured elicitation exercise, we captured expert opinion on available scientific evidence for low dose-response relationships for genotoxic carcinogens. This allowed assessment of: available evidence for the nature of dose-response relationships at low levels of exposure; the generality of judgments about such dose-response relationships; uncertainties affecting judgments on the nature of such dose-response relationships; and whether this last should differ for different classes of genotoxic carcinogens. Elicitation results reflected the variability in experts' views on the form of the dose-response curve for low dose exposure and major sources of uncertainty and most importantly, query the rigour of the assumption of a linear relationship.

Highlights

- Experts are reluctant to express views on the dose-response curve for genotoxic carcinogens

- The POD can be extrapolated first from high to low dose in animals and then to humans, or vice versa
- Expert judgement was that the dose-response curve is highly non-linear at human relevant exposures
- Stochastic events and the distribution of susceptibilities will contribute, to an unknown extent
- Interpretation of the MOE requires consideration of MOA, species differences and human variability

Keywords

Dose–response function; expert judgment; genotoxic carcinogens; low dose extrapolation; margins of exposure; risk assessment

1. Introduction

Many chemicals (and other agents, such as radiation) that cause cancer by a genotoxic mechanism do so by mutating critical oncogenes. As cancer is a disease of clonal (single cell) origin, it has been argued that a single mutation in a single cell would be sufficient to give rise to cancer, and, as a consequence, there may be no threshold concentration for a genotoxic carcinogen such that exposure below that will not cause mutations. Hence, until relatively recently, risk assessment of such compounds took one of two forms. Either (1) risk assessment stops with identification of the hazard: the tumours caused by a compound could reasonably arise as a result of its genotoxicity, or (2) the experimental carcinogenicity data are extrapolated to a risk considered to be of low or negligible concern, typically 1 in 10^5 or 1 in 10^6, the corresponding exposure being known as the "virtually safe dose". In the former, the information is often translated into a risk management strategy of reducing exposure to levels that are "as low as reasonably achievable/practicable" (ALARA/P). However, the output of the risk assessment does not provide a clear basis for deciding on the urgency or extent of risk management action nor does it enable any prioritisation of competing hazards (EFSA, 2005; O'Brien et al., 2006; Benford et al., 2010). In the latter approach, a decision needs to be taken as to the form of the dose-response relationship below that tested experimentally. In the most recent update to the Cancer Guidelines of the US EPA, it was concluded that the shape of the dose-response curve at human relevant exposures cannot be assumed to be any given shape on the basis of current knowledge, and, hence, a plausible worst case would be to assume linear extrapolation from experimental data (EPA, 2005).

Low dose linear extrapolation has been criticised due to the considerable uncertainty about the shape of the dose-response curve at human relevant exposures (Figure 1) (Williams et al., 2005; EFSA 2005). In addition, risk management on the basis of population incidence requires a policy decision as to what risk is considered acceptable at a virtually safe dose and can lead to difficulties in risk communication. To overcome these concerns about low dose extrapolation whilst enabling risk assessors to provide information to risk managers to assist in judgements on the overall level of concern and in prioritisation of competing hazards, several bodies have proposed use of the margin of exposure (MOE). The MOE is the ratio of the point of departure (PoD), typically the Benchmark Dose – Lower Confidence Limit ($BMDL_{10}$) for a tumourigenic response in animals, against estimated human exposure; often, the estimation of exposure uses plausible worst case assumptions regarding exposure by likely routes, e.g. oral, inhalation, from anticipated uses and/or environmental levels, for a genotoxic carcinogen. Interpretation of the MOE requires consideration of the uncertainty and variability that underlie intra- and inter-species differences. Values of MOE that are equal to or greater than 10,000 have been considered to indicate "low concern", although different organisations (e.g. European Food Safety Authority; Committee on Carcinogenicity / Committee on Mutagenicity;

JEFCA: Joint FAO/WHO Expert Committee on Food Additives) may employ different expressions of risk.

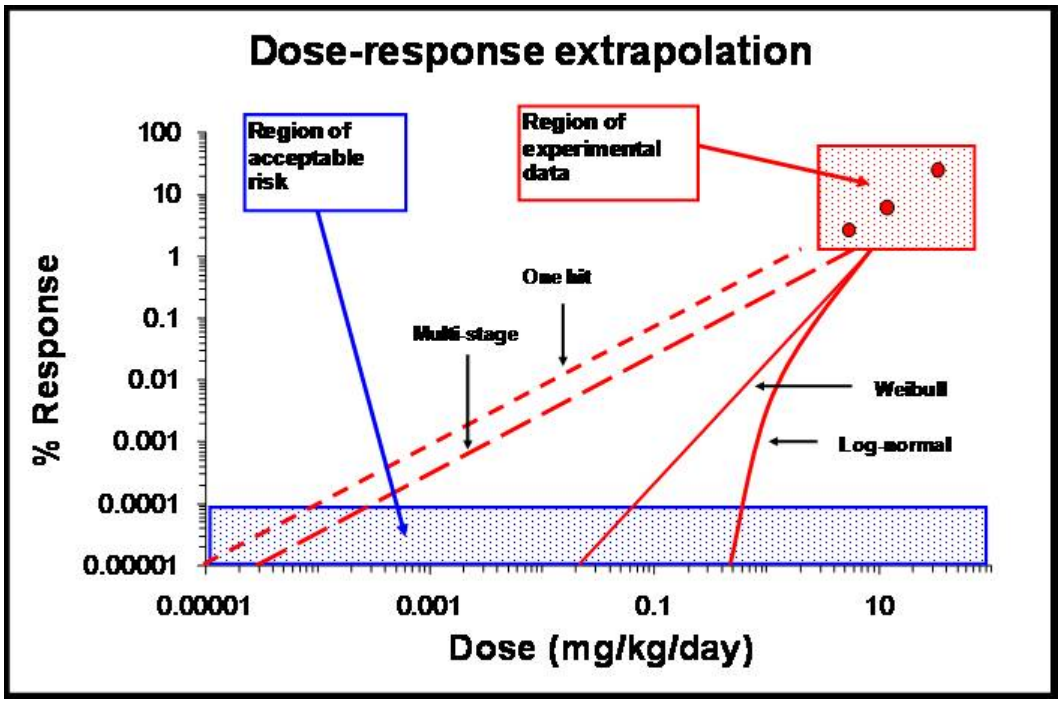


Figure 1: A few examples of possible dose-response curves for genotoxic carcinogens at human relevant exposures. The figure is reproduced and modified from the EFSA opinion on "Harmonised Approach for Risk Assessment of Substances Which are both Genotoxic and Carcinogenic Low dose extrapolation from animal carcinogenicity data using various models" (EFSA, 2005).

The nature of the dose-response curve at human relevant exposures is a key component of the risk assessment process for evaluating the potential adverse health effects of chemicals. A workshop held in Baltimore, Maryland, on 23–24 April 2007, sponsored by U.S. Environmental Protection Agency and Johns Hopkins Risk Sciences and Public Policy Institute, served as a starting point for the current exercise (White et al., 2009). At the 2007 workshop, a multidisciplinary group of experts reviewed the state of the science regarding low dose extrapolation modelling and its application in environmental health risk assessments. Participants identified discussion topics based on a literature review, which included examples for which human responses to ambient exposures have been extensively characterized for cancer and/or noncancer outcomes. Topics included the need for formalized approaches and criteria to assess the evidence for mode of action (MOA), the use of human versus animal data, the use of MOA information in biologically based models, and the implications of inter-individual variability, background disease processes, and background exposures in threshold versus non-threshold model choice. Participants recommended approaches that differ from current

practice for extrapolating high-dose animal data to low dose human exposures, including statistical approaches such as model averaging, categorical approaches for integrating information on MOA and inference-based models that explicitly consider uncertainty and inter-individual variability.

A rather large (theoretically infinite) number of dose-response models for genotoxic carcinogens exist; these include both linear and non-linear models (Bolt et al., 2004; Neumann, 2009; O'Brien et al, 2006; Swenberg et al, 2008; Williams et al., 2005). Most of the models used in dose-response analysis software (e.g. BMDS of the US EPA) are statistically-based (i.e. curve is based on goodness of fit considerations only) with no clear biological basis, e.g. Weibull, log-logistic models. Many additional models, based on different physiological considerations regarding the possible effects of genotoxic carcinogens, are possible, for example the multi-stage cancer model. What all of these models have in common, so far, is that they lack a detailed, transparent, rigorous scientific rationale to justify their consideration in risk assessment of genotoxic carcinogens at human relevant exposures. Also, some experts advocate the choice of a dose-response model on a "case by case" basis (Neumann, 2009; Swenberg et al, 2008).

Most bodies advocating quantitative risk assessment, to identify a virtually safe dose of a genotoxic carcinogen, now recommend the use of low dose linear extrapolation (for example: USA, NL); the provision of a quantitative risk estimate is one major advantage of low dose linear extrapolation (European Commission, 2009).

In interpreting the level of concern represented by the margin of exposure, a number of issues can be considered, for example (a) that the point of departure is not equivalent to the NAEL (no adverse effect level), (b) uncertainties relevant to human variability in cell cycle control and DNA repair, and (c) uncertainties about the shape of the dose–response curve below the BMD and the dose level below which the cancer incidence is not increased (European Commission, 2009).

The present study concerns the elicitation of expert knowledge regarding the form of the dose-response curve for genotoxic carcinogens at human relevant exposures with the view to analysing this information for any implications for a level of concern for the MOE. It is part of a wider project[1] that is designed to provide guidance on the interpretation of the level of concern for the margins of exposure for genotoxic carcinogens.

[1] Funding source: Food Standards Agency, UK - project T01051 "Interpretation of Margins of Exposure approach for Genotoxic Carcinogens"

Expert judgment is being sought in a number of steps in risk assessment of genotoxic carcinogens (COC, 2004; IPCS, 2009) mainly because of the numerous gaps of knowledge and uncertainties that burden this area of risk assessment. Although it is widely recognised that expert judgment cannot replace data-driven studies, it is also widely accepted that in the face of incomplete knowledge elicitation of expert judgment is the only sound alternative for integrating available knowledge and, above all, for systematically characterising uncertainties (O'Hagan et al., 2006; Cooke, 2009). In particular, structured approaches to elicit expert judgment provide the necessary framework to ensure the coherent capture of experts' uncertainties and transparent documentation of experts' opinions.

The questions we desired to answer *via* the structured expert elicitation exercises of this study were:

- What are the known(s) and unknown factors underlying the different dose-response models for genotoxic carcinogens?
- What is the rigorous scientific rationale to support the choice among the different dose-response models and assumptions for genotoxic carcinogens?

2. Materials and methods

2.1 Expert elicitation process

The expert elicitation scheme was designed to capture information about the nature of the dose-response curve at human relevant exposures and was conducted over two phases. During the first phase, we elicited expert judgments remotely (*via* a structured online questionnaire) with a view to discussing and refining the elicited judgments in a follow-up experts' workshop (the second phase). The information we gathered during the first phase revealed that, in principle, the majority of participants were reluctant to provide detailed quantitative judgments on low dose extrapolation of genotoxic carcinogens. Nevertheless, the experts' inputs elicited during this phase were essential because they allowed us to prepare the framework for the second phase of elicitation (i.e. experts' workshop).

Different questionnaires were used in the two phases of the exercise. The questionnaires were custom designed and developed by the project team, and were aimed at facilitating the elicitation of quantitative information from experts. The questionnaire used in phase I was designed to prompt experts to provide the rationale and uncertainties behind their judgments (as this was an online exercise). Figure 2 was developed following analysis of the comments received during phase I, and was designed as a facilitation tool for focusing discussion of the rationale and uncertainties during the experts' workshop, i.e. in phase II. The questionnaire shown in Appendix II was used in this phase for elicitation of quantitative information.

2.1.1 First phase - online structured questionnaire

We identified a number of international experts involved in the risk assessment of genotoxic carcinogens or dose-response assessment of such compounds through their academic publications, employment in a regulatory body, e.g. US Environmental Protection Agency, UK Food Standards Agency, involvement in scientific advisory committees (both national and international), e.g. Joint FAO/WHO Expert Committee on Food Additives (JECFA), the EU Scientific Committee on Health and Environmental Risks (SCHER), UK Committee on Carcinogenicity (COC), and through their participation in collaborative activities relevant to the risk assessment of genotoxic carcinogens, e.g. WHO International Programme on Chemical Safety (IPCS), ILSI branches such as the Health and Environmental Sciences Institute and Research Foundation, European Centre for Ecotoxicology and Toxicology of Chemicals (ECETOC). Efforts were made to ensure geographical distribution and a range of scientific perspectives. In total, we invited 68 internationally renowned experts in the fields of toxicology and risk assessment of carcinogens to participate remotely in an elicitation exercise that was designed to elicit their judgments on low dose-response relationships for genotoxic carcinogens

in humans. The elicitation questionnaire[2] was sent to the experts who agreed in principle to participate; anonymity and confidentiality were guaranteed to the participants. This exercise was divided into four parts and invited experts to:

- Report personal attributes (e.g. professional affiliation, gender, age) and to declare any potential conflicts of interest, by completing a declaration of interest (Part I)
- Provide information about their expertise (Part II)
- Indicate their estimates and uncertainty regarding the carcinogenicity of chemicals at low levels of exposure (Part III); in particular experts were asked to provide quantitative estimates on:
    - Exposure that would lead to a certain number of extra cancer cases in the expert's own country
    - Baseline incidences of cancer cases in the expert's own country
    - Number of cancer cases for exposure of the general population and for occupational exposure
- Explain: a) the scientific rationale that would support the elicited opinions, and b) the scientific uncertainties that may burden the elicited opinions (Part IV); in particular the questionnaire included open questions for the experts to communicate:
    - Data gaps in the field of low dose extrapolation (LDE) for genotoxic carcinogens
    - The scientific rationale behind their judgments
    - The scientific uncertainties associated with their judgments

2.1.2 Second phase – experts' workshop

A one-and-a-half days experts' workshop was organised at the Food and Environment Research Agency (Fera), Sand Hutton, York (23rd – 24th of May 2011), in which 11 experts from Germany, Italy, Switzerland, UK and USA participated (Table 1). Two of these experts were also participants in the first phase of this study.

The goal of the workshop was to elicit expert opinion on the nature of the dose-response curve at human relevant exposures for genotoxic carcinogens (GCs), in particular on the following aspects:

- What evidence can be used to determine the nature of the dose-response curve at human relevant exposures?
- What judgments about the nature of the dose-response curve at human relevant exposures can be made for GCs in general?
- Does the form of the dose-response curve at human relevant exposures differ for different classes of GCs?

[2] The online questionnaire is available as supplementary material (Appendix I).

- What are the uncertainties associated with expert judgments?

The workshop comprised two plenary and two breakout group sessions. The initial plenary session was used to discuss two alternative approaches for thinking about the nature of the dose-response curve at human relevant exposures (Figure 2). These were identified after evaluating experts' judgments elicited during the first phase; one could think of interpreting the shape of the dose-response curve either after or before translating it from available experimental animal data to humans (i.e. approaches 1 and 2 respectively in figure 2).

The initial plenary session was followed by two breakout sessions. Participants were divided into two breakout groups, which were led by experts in the conduct of expert elicitation (details on participants of the breakout groups are shown in Table 1). The aim of the breakout sessions was to elicit quantitative estimates of exposure that could lead to a particular number of cancer cases per lifetime (i.e. 1 in 10,000, 1 in 100,000 and 1 in 1,000,000) for the general human population (approach 1 in figure 2) or the general rat population (approach 2 in figure 2). Experts were asked to indicate their most likely, minimum and maximum values as well as their 90% credible intervals (elicitation questions are shown in detail in Appendix II of the supplementary material), and they were familiarised with how to provide such quantitative estimates through a preceding brief training exercise. Before providing their actual judgments, experts discussed the issues involved in addressing the question such as the assumed dose, i.e. the group assumed that the question referred to lifetime exposure *via* the diet.[3] Experts were encouraged to exchange information so that they would consider all lines of evidence known to them[4]. Thereafter, individual opinions were elicited and consequently shared and discussed among the group; following the latter discussion, experts had the choice of changing their individual judgment if they so wished.

The two breakout groups addressed both routes of low dose interpretation, albeit in a different order. Breakout group 1 addressed approach 2 first, whereas breakout group 2 addressed approach 1 first. In order to ensure that all views would be captured, the breakout sessions were governed by certain rules to which all experts agreed in advance, e.g. equal time allocated to all experts; experts were urged to keep to time; respect everyone's opinions so that they would allow a balanced discussion based on

[3] Full text of the question addressed during the breakout groups: For a directly DNA reactive genotoxic carcinogen, given a point of departure (BMD_{10}) for rat dose response, please give us your judgements on the relative exposure that would cause an extra 1 in a million cases of cancer per lifetime for the population of (inbred) rats. Additional assumption: Lifetime exposure *via* the diet.

[4] The core information that experts shared in both breakout groups is shown in Appendix II in supplementary information, and it comprised published papers, non-published (at the time) specifics, and individual assumptions and/or views on major sources of uncertainties.

available evidence and rationale. The participants of the workshop were happy for their participation in the workshop to be published, but it was agreed that all workshop outputs would be communicated without attribution to individuals. All activities in the breakout groups were facilitated by project team members who did not have specific expertise in risk assessment of genotoxic carcinogens, therefore ensuring that discussions were not off-centre (i.e. balanced, as all participants were encouraged to voice their views, and without digressing from the issues of concern; see Table 1 for details of facilitators and participants of breakout groups).

During the second plenary session that followed the breakout sessions, the results of the breakout sessions were presented to all participants and discussed.

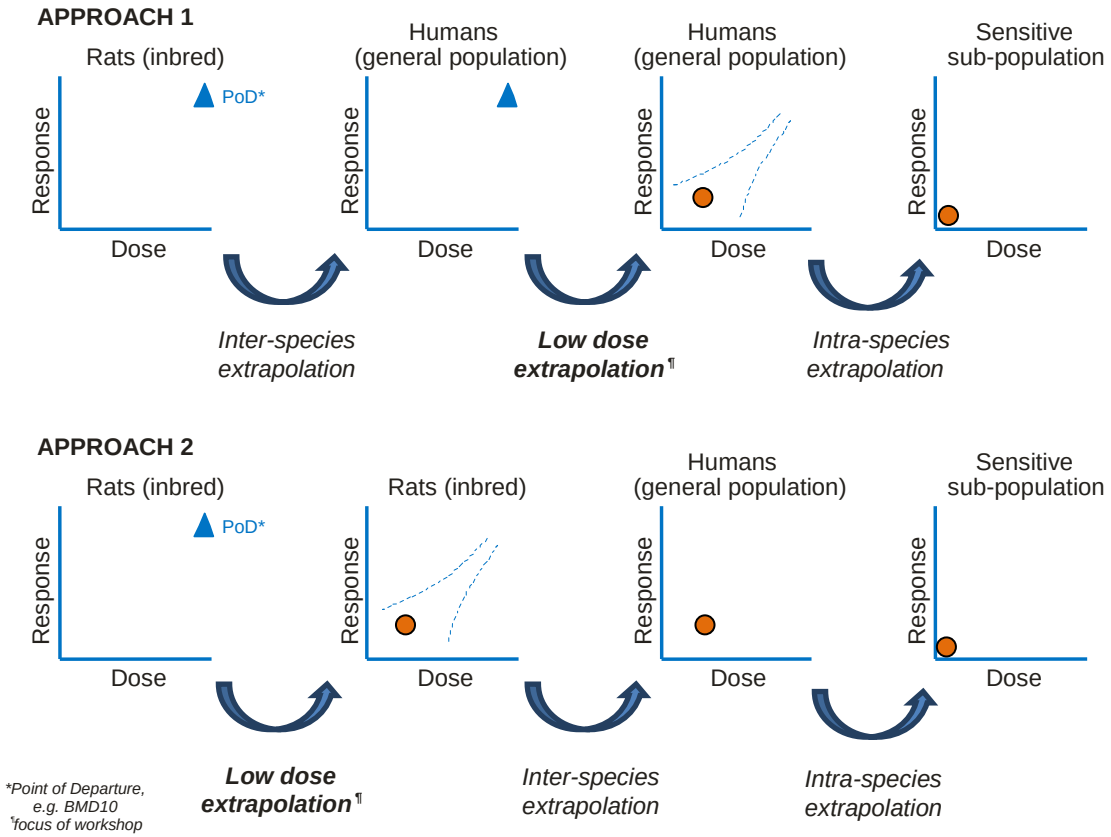


Figure 2: Illustration of possible sequence of steps when assessing risk of genotoxic carcinogens. In particular, one could think of interpreting the shape of the dose-response curve either before or after an inter-species extrapolation is performed. When designing the experts' workshop it was hypothesised that the choice of the approach chosen could affect the final assessment.

2.1.3 Analysis of results

Quantitative judgments elicited during the experts' workshop were treated individually although they are presented combined in graphs in the results section to facilitate comparison among the individual opinions.

Qualitative judgments elicited were analysed as follows: during a first step, categories of data (e.g. factors, uncertainties, assumptions) were formed, and, during the second step of analysis, individual data were grouped under the different categories. The approach we implemented to analyse experts' recorded views was exploratory, albeit based on a methodology we had developed previously (Flari et al., 2010). When all the original participants' opinions were taken into account, these were further grouped into a number of sub-categories; sub-categories depended heavily on the opinions expressed by the participants, i.e. the more diverse the opinions the larger the number of different sub-categories.

3. Results

In both phases of the elicitation study, it appeared that estimating the human exposure that could lead to a particular number of extra cancer cases is not easy. The main challenges when eliciting quantitative information relevant to the shape of the dose-response curve at human relevant exposures appeared to be related to (a) relevant historical views and established practices, (b) lack of appropriate key data, and (c) quantification of relevant uncertainties.

3.1 Biases for/against or familiarity with assumed linearity

18 out of the 68 invited experts agreed in principle to participate in the elicitation exercise included in the first phase of our work. Three of these experts cited concerns about delivering quantitative judgments on low dose extrapolation and withdrew from the exercise after they received the online questionnaire (all were from the EU). Although the sample number was small, there did seem to be greater willingness amongst respondents from the USA than from Europe to provide quantitative judgements, possibly reflecting wider acceptance of such an approach in their jurisdiction.
By the end of this phase, half of the experts who initially agreed to participate had sent responses (i.e. 9/18); the majority of the respondents were from USA (i.e. 6/9). The information elicited *via* the online structured questionnaire from these 9 experts was incomplete as they varied in their willingness to provide detailed quantitative judgments (Table 2); some experts did not provide any quantitative estimates for any of the unknown parameters (2/9), whereas most provided quantitative estimates for a subset of the unknown parameters (6/9). One expert provided all quantitative judgments for all unknown parameters.

For the second phase of the study, twenty-four experts in the field of risk assessment of genotoxic carcinogens were invited to participate in an experts' workshop with the view to discussing collected (*via* the online structured questionnaire) expert opinions about the functional form of the dose-response curves and the scientific rationale and uncertainties underlying these, and how results from the above could be used in the risk assessment of genotoxic carcinogens. 11/24 of these experts agreed to participate in the workshop, representing both EU and USA perspectives (Table 1).

Experts provided their uncertainty range for quantitative estimates of exposures that may lead to a specified number of additional cancer cases. Experts varied in their willingness to provide such estimates, albeit to a much lesser extent compared with the first phase of the elicitation when we tried to elicit these *via* the online structured questionnaire.

3.2 Timing of low dose extrapolation step during the risk assessment of genotoxic carcinogens: does it influence the quantification of uncertainties?

The two approaches identified in the analysis of the responses to the first phase of the study (i.e. *via* the online structured questionnaire), as described above, allowed us to structure the experts' workshop that followed in the second phase of this elicitation study. During the initial plenary session of the workshop, any questions the experts had regarding the alternative approaches to thinking about interpretation of the shape of the dose-response curve were clarified (figure 2). Also, the experts discussed the approaches in detail, and they agreed that each approach carried its own advantages and disadvantages for risk assessment of genotoxic carcinogens (Table 5). They indicated that each approach would make better use of different types of data, and they perceived approach 1 to be more familiar and easier to communicate to wider audiences. All experts agreed that the choice of approach would depend mainly on the availability of data and, thus, it would be best if one could decide on which approach to follow on a case-by-case basis (Table 5). Experts felt that, as a result of these identified differences the two approaches would allow different challenges and uncertainties to be highlighted.

3.3 Expert judgement on dose associated with minimal risk of cancer in humans

The quantitative information acquired during the online survey was too limited to perform any meaningful analysis of uncertainty. However, as mentioned above, experts were much more willing to provide quantitative estimates of their uncertainty during the workshop.

Irrespective of whether experts were considering approach 2 in figure 2 or approach 1 in figure 2, they considered that the dose-response curve at human relevant exposures was most likely to be sub-linear (Figures 3A and B). This was based on a number of lines of evidence, including general biological principles (see below), but it was generally agreed that definitive information in support of this view was lacking. A number of experts thought that the shape of the dose-response curve that would lead to a specified number of extra cancer cases would most likely be chemical specific (3/6 in breakout group 1 and 1/5 in breakout group 2). In addition, all experts considered that the least conservative best case (i.e. maximum exposure that would cause the specified increase in cancer incidence) would reflect a very steep decline in dose-response to a virtual threshold within one dose increment. In contrast, the majority of experts considered the most conservative case (i.e. minimum exposure) would reflect a linear decline, although the opinions varied from sub-linear to supra-linear (Figures 3A andB).

Experts in breakout group 2 were more reluctant to provide quantitative estimates: only one of the five experts provided all the required information (Figure 3B). Additionally, experts in this group felt that they could not provide any quantitative estimates of exposure that would lead to an extra 1 in

100,000 or 1 in 1,000,000 cancer cases because it would be extremely difficult to distinguish this from the background rates of cancer incidence (Figure 3B).

3.4 Lack of key data and experts' varied opinion on significance of underlying uncertainties

Regardless of the elicitation phase of the study, experts were more willing to cite the scientific uncertainties and the rationale behind their quantitative judgments or their lack of willingness to provide any quantitative estimates (Fig 3).

Uncertainties listed by the experts who participated in the first phase of the study are grouped according to the aspect of the low dose extrapolation step they concern; for most aspects of low dose extrapolation different experts mentioned different uncertainties as being most significant (Table 3A). Nevertheless, regardless of the phase of elicitation concerned, experts appeared to agree on the significance of uncertainties relevant to (a) modes of action, (b) species differences, (c) sensitive subpopulations, and (d) the stochastic nature of cancer in influencing the shape and form of the low dose curve.

During the workshop experts discussed a large number of different lines of evidence for interpreting the nature of the dose-response curve[5]; still, they were very uncertain about how justifiable or conservative is an MOE of 10,000 as the lower bound for a level of concern. A key issue discussed during the final plenary session was the extent to which the carcinogenic process is stochastic and the extent to which is reflects the distribution of individual susceptibilities. The former would lead to a linear dose-response relationship whilst the latter would lead to a threshold. It was concluded that it was not possible to determine the relative contribution made by these two aspects. Experts found it difficult to estimate the extent to which the various factors involved may overstate the risk, particularly as they did not feel able to quantify many aspects of the cancer process, e.g. DNA repair mechanisms.

4. Discussion

Experts who participated in the elicitation workshop were more willing to providing quantitative estimates of risk at low exposures compared with the experts who were invited to participate in the remote elicitation exercise. This may indicate that the particular task is too challenging to be realised remotely and under non-facilitated elicitation. Alternatively, expert judgement may be easier to express following sharing of knowledge and expertise with peers.

[5] Details on lines of evidence considered during the experts' workshop are available in Appendix III in supplementary information.

Regardless of the phase of the elicitation in our study it appeared that experts' judgements on low dose extrapolation stem, at least to some extent, from different philosophical perspectives that would not necessarily point to linearity. That is, experts hold a qualitative view of the nature of the dose-response relationship for genotoxic carcinogens, which depends only in part on objective scientific evidence, as the evidence base in incomplete.

A number of the participants felt that dose-response models could be employed mainly to explain the least and most conservative scenarios, whereas it is much more difficult to predict most likely dose that would lead to a particular number of extra cancer cases. However, eliciting the "*most likely*" judgement proved to be problematic; it is recognised that it might be wiser to ask for the median, described verbally as "*just as likely to be above as below*"; this in principle could help with other judgments as it introduces the concept of an indifference point which is what most approaches to quantification of subjective probabilities revolve around. One particularly useful outcome of the exercise was the variability within and between experts' quantitative judgments: why do experts' judgments differ as much? The results from the initial phase of our study indicated that at least part of experts' disagreement may arise from differences in a predetermined philosophical perspective they have when considering low dose extrapolation.

Two underlying conceptual theories for the relationship between low doses and cancer response were discussed at the workshop. The first is that the cancer process is purely stochastic and therefore there is a linear dose-response curve that would reflect effects on human health. There is some evidence in support of this view from studies of the dose-response to radionuclides (Gilbert, 2009) and to some chemicals such as certain alkylating agents (e.g. ethylnitrosourea; Gocke & Muller, 2009). The second is that the driver of inter-individual variability is the underlying susceptibility of each individual in the general human population. Hence, the dose-response relationship would then represent the distribution of susceptibility, reflecting the log-normal distribution that would arise from the multiplication of factors contributing to such susceptibility (Lutz, 1999). Log-normality is chosen to reflect multiplication of susceptibility factors (according to the central limit theorem); subsequently, susceptibility factors are effectively conditional probabilities of successive biological stages towards development of actual tumours and vary between individuals. However, this view is subject to criticism because of (a) statistical constraints, i.e. log-normality of the distribution is based on either assuming relative independence between all factors that contribute to susceptibility and/or that each of the contributing factors follows a log-normal distribution, and (b) lack of empirical data to support the view that the distribution must be of any given shape (Conolly et al., 2005).

The majority view expressed during the workshop was that, in reality, both mechanisms (i.e. (a) the cancer process is stochastic and (b) the cancer process is a reflection of underlying inter-individual susceptibility) are likely to be in operation, but that there is little or no information on the balance between them. Consequently, it would be pragmatic to start by assuming linearity and consider various models for departures from linearity depending on the particular chemical concerned, e.g. saturation of clearance, saturation of metabolic activation, induction of repair, saturation of repair, induction of cytotoxicity, cell division slow down, age effects, etc (Gocke & Muller, 2009; Lutz, 2009).

However, although experts may have considered a number of different philosophical perspectives, they appeared to agree that species differences, sensitive subpopulation/s, and most particularly the mode of action (and consequently extrapolation between chemicals) are amongst the most significant factors influencing the true form of the dose-response curve for genotoxic carcinogens. Experts also agreed that uncertainties associated with the above factors are numerous; for example, there is very little information on the levels of variability of susceptibility within the human population.

Additionally, experts recognised that although the dose-response curves for some chemical carcinogens have been extensively studied (e.g. 2-acetlaminofluorene (Farmer et al, 1980; Williams et al, 2000; Littlefield et al, 1980); dimethylnitrosamine (Peto et al, 1991); dibenzo[a,l]pyrene (Bailey et al, 2009); diethylnitrosamine (Peto et al, 1991; Williams et al, 2000)), it is still not possible to determine the nature of the relationship at human relevant exposures with confidence. For less studied chemicals, the lack of key evidence needed to make educated judgments about probable dose-response curves (i.e. knowledge of actual dose, other toxicity, tissue affected, degree of detoxification at a low dose, competing rates, etc) introduces an even higher level of uncertainty on the extrapolation step between chemicals.

Is the level of concern conservative? How close do experts think that it reflects true risk? The experts considered it was more likely than not that the dose-response curve at exposure levels of concern was non-linear. However, all found it difficult to reach generic conclusions on the contribution of each factor (e.g. cell repair mechanisms, inter-individual differences, differences in mode of action, tissue/s targets, etc) in the cancer process that might contribute to the nature of the dose-response relationship at such human relevant exposure levels. This inevitably has consequences on the interpretation and use of MOE values in the risk assessment of genotoxic carcinogens. During the workshop experts indicated that one may need to consider interpretation of the MOE of such chemicals on a case by case basis.

Both elicitation sessions in our study showed that experts consider mode of action, species' differences, and inter-individual variability as the most significant factors influencing the dose-response for genotoxic carcinogens. Our future work aims to elicit expert judgment on how to classify genotoxic carcinogens by taking into account these three factors, and which case studies would best represent classes/categories of genotoxic carcinogens to be evaluated further, i.e. elicit quantitative estimates on the form of the dose response curve for each identified classes/categories of genotoxic carcinogen, and evaluate systematically all uncertainties underlying experts' judgments, and their possible influence (i.e. negative or positive) on the shape of dose response curve/s.

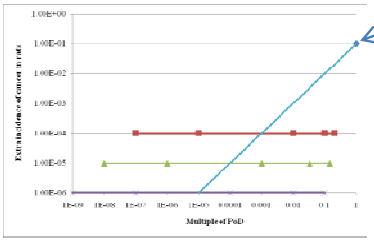
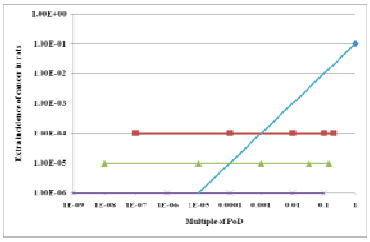
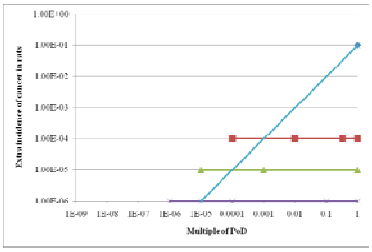
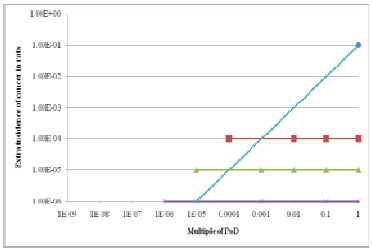
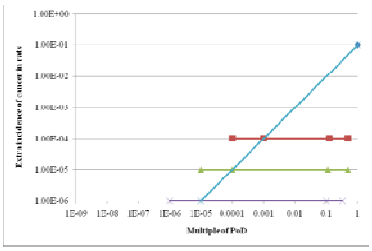
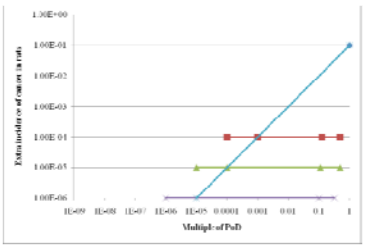
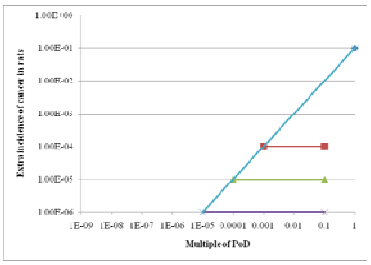
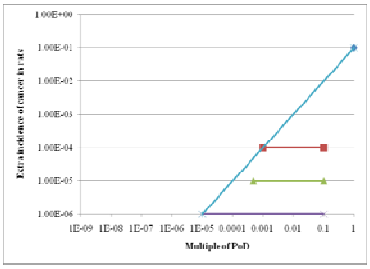
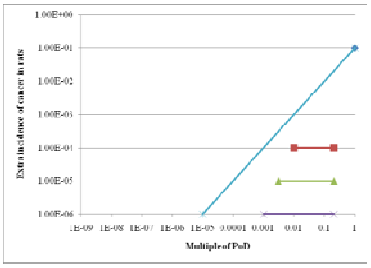
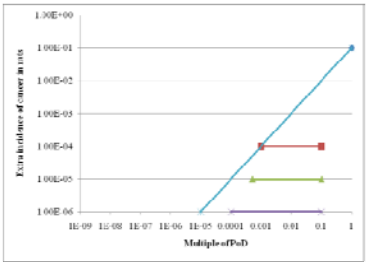
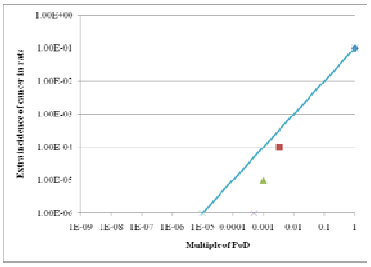
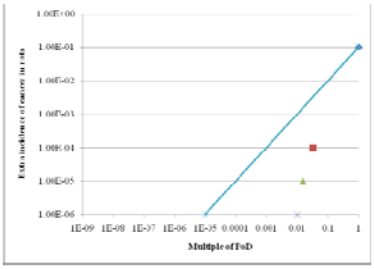
Approach 1		**Approach 2**
	PoD BG1 - Expert A All	
	BG1 - Expert B Approach 1: Min, Max, 90% CIs Approach 2: All	
	BG1 - Expert C Min, Max, 90% CI	
	BG1 - Expert D Min, Max	
	BG1 - Expert E Min, Max	
	BG1 - Expert F ML	

Figure 3A

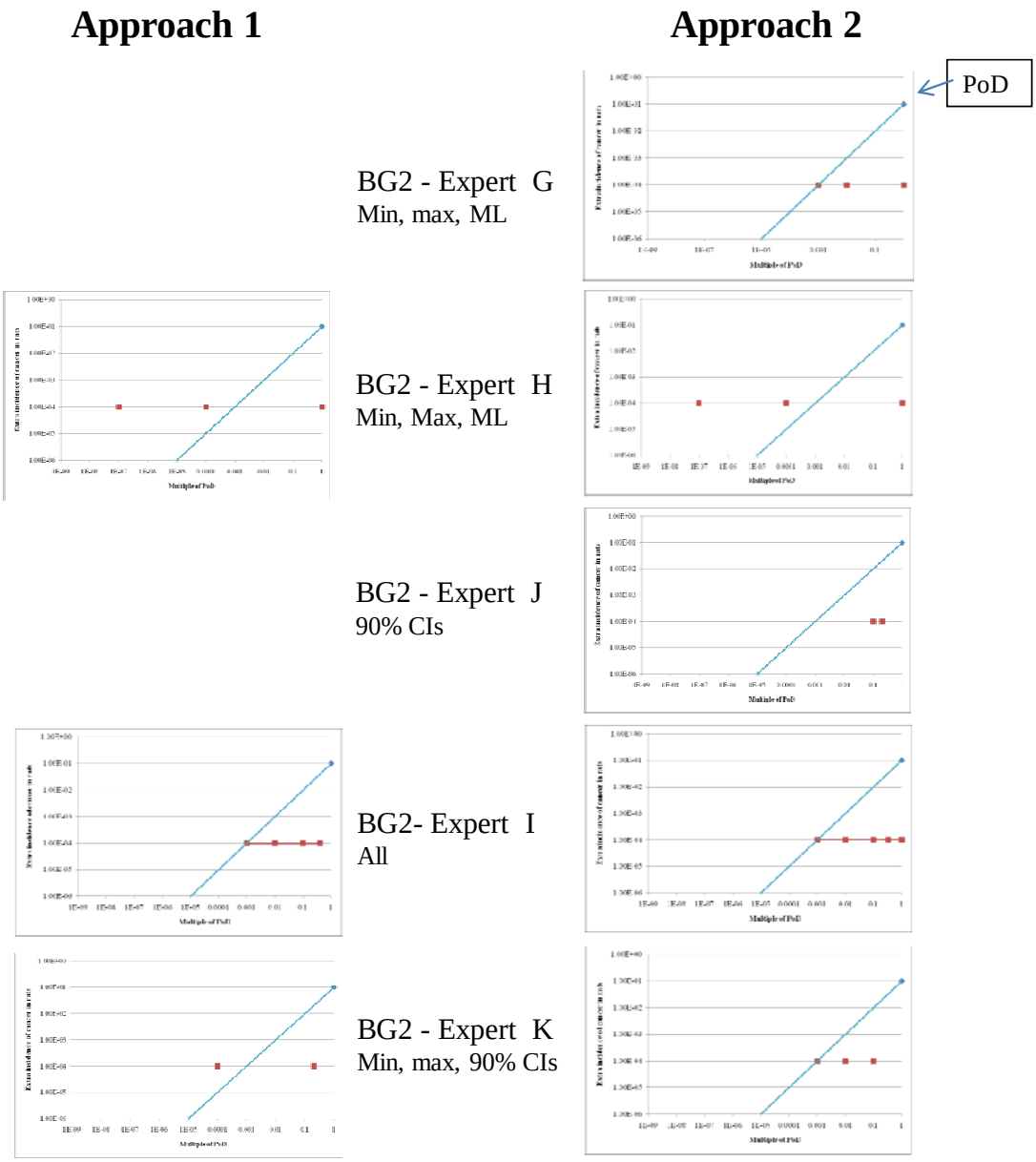

Approach 1
Approach 2
PoD
BG2 - Expert G
Min, max, ML
BG2 - Expert H
Min, Max, ML
BG2 - Expert J
90% CIs
BG2- Expert I
All
BG2 - Expert K
Min, max, 90% CIs

Figure 3B

Figure 3: Quantitative estimates of exposure that could lead to a specified number of additional cancer cases in human (approach 1) or inbred rat (approach 2) general population that were elicited from the experts in breakout group 1 (A) and in breakout group 2 (B)

Experts in breakout group 2 felt that they could not deliver any quantitative estimates of the exposure that could lead to an extra 1 in 100,000 or 1 in 1,000,000 cancer cases because it would be extremely difficult to distinguish this from the background rates of cancer incidence.

The blue line indicates the linear extrapolation from point of departure (PoD). Squares indicate experts' estimates of exposure that could lead to 1 in 10,000 additional cancer cases per lifetime; Triangles indicate experts' estimates of exposure that could lead to 1 in 100,000 additional cancer cases per lifetime. "X" indicates experts' estimates of exposure that could lead to 1 in 1,000,000 additional cancer cases per lifetime. Irrespective of the breakout group experts varied in the information they provided. In particular:

- Expert A and expert B for approach 2 provided all required estimates: min, max, 90% quantiles, most likely
- Expert B for approach 1 and expertC provided estimates for: min, max, 90% quantiles
- Experts D and E provided estimates for: min, max
- Expert F provided estimates for: most likely
- Experts G and H provided estimates for: min, max, most likely
- Expert J provided estimates for: 90% quantiles
- Expert I provided estimates for: min, max, 90% quantiles, most likely
- Expert K provided estimates for: min, max, most likely

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Table 1: List of experts who participated in the workshop held at Fera (22nd – 23rd May 2011); the experts were separated into two breakout groups[6] before going through a brief elicitation training exercises and eliciting individual quantitative estimates. Expert participants in breakout groups were chosen in advance by the project team with the view to ensuring similar representation from different risk assessment *status quo* practices in each group.

Expert	**Country**	**Breakout group**	
		Group 1	**Group 2**
Prof. Alan Boobis	UK	+	
Dr Philip Carthew	UK		+
Dr Rory Conolly	USA		+
Prof. Corrado Galli	IT	+	
Dr Helmut Griem	GER		+
Dr Werner Lutz	SZ		+
Barry Maycock	UK	+	
Dr Franz Oesch	SZ	+	
Dr Lorenz Rhomberg	USA	+	
Dr Lesley Rushton	UK		+
Dr Rita Schoney	USA	+	

[6] Breakout group 1 was facilitated by Dr Villie Flari, and Dr Peter Craig acted as a rapporteur. Breakout group 2 was facilitated by Dr Andy Hart and Dr John Paul Gosling acted as a rapporteur.

Table 2: Synopsis of experts' willingness to provide quantitative inputs during the first phase of our study (i.e. remotely). Min: minimum; CIs 90%: lower and upper 90% credible intervals; max: maximum.

Questions			**Experts**								
			1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
1	Given the point of departure for human dose-response (a rat BMDL10: 10mg/kg) for a hypothetical genotoxic carcinogen and taking into account your knowledge on genotoxic carcinogens, could you give us your judgments on the exposure that could potentially lead to 10^{-6} (one in a million) extra cases of cancer per lifetime for the general population in your country?										
1a		General Population	Most likely	All	Most likely	All	All	Most likely, min, max			Most likely, 90% quantiles
1b	Would any of your judgments above be any different for any particular subpopulation in your country?	Subpopulation A		All		All	All	Most likely, min, max			Most likely, 90% quantiles
1c		Subpopulation B		All			All				Most likely, 90% quantiles
2	Given your knowledge on genotoxic carcinogens, could you give us information on the number of cancer cases per lifetime that to your point of view may be attributed to genotoxic carcinogens for the general population in your country?										
2a		General Population		All	Most likely	All	All	Most likely, min, max			Most likely, 90% quantiles
2b	Could these numbers be different perhaps according to different types of cancer?	Cancer type A		All		All	All	Most likely, min, max			
2c		Cancer type B		All							
2d	Would any of your judgments above be any different for any particular subpopulation in your country?	Subpopulation A		All			Max	Most likely, min, max			
2e		Subpopulation B		All							
3	Given the point of departure for human dose-response (a a rat BMDL10 : 15 mg/kg) for a hypothetical genotoxic carcinogen and taking into account your knowledge on genotoxic carcinogens, could you give us information on the number of cancer cases per lifetime that to your point of view may be attributed to a hypothetical genotoxic carcinogens for: a) the general population in your country assumed to be exposed at 0.001 mg/kg/day, or b) the population that works in the industry assumed to be exposed at 0.1 mg/kg/day.										
3a		General Population		All		All	All	Most likely, min, max			Most likely, 90% quantiles
3b		Occupational exposure		All		All					Most likely, 90% quantiles
3c	Could these numbers be different perhaps according to different types of cancer?	General Population		All							
3d	Would any of your judgments above be any different for any particular subpopulation in your country?	Subpopulation A		All		All		Most likely, min, max			
3e		Subpopulation B		All							

Table 3: During the first phase of this elicitation exercise (i.e. online structured questionnaire) experts were invited to cite the uncertainties they took into account when delivering their judgment (mainly an input or a lack of input, e.g. Expert h). The number of uncertainties that each expert cited is shown in brackets by the code letter of each expert. Experts were also invited to rank the uncertainties they cited according to their significance in the low dose extrapolation step; the ranks are shown in the table, 1 being the most significant. When an uncertainty is marked with an "X" it implies that the uncertainty was mentioned by the expert but it was not ranked.

Aspect of risk assessment	**Uncertainties**	**Experts' ranks**								
		1 (0)	**2 (6)**	**3 (1)**	**4 (8)**	**5 (8)**	**6 (4)**	**7 (0)**	**8 (2)**	**9 (14)**
Chemical to chemical differences	Chemical to chemical differences in life-stage-specific sensitivities for carginogenesis									X
	Incomplete database (projections from chemical to chemical)									X
Dose-response model	Shape of dose-response curve below POD						3			
	Pharmacodynamics and Pharmacokinetics - high to low dose projections of total metabolism rate in humans									X
Human exposure	Concurrent exposures to other carcinogens					8				
	Exposure variability – steady state, episodic, short term					7				
	Human exposures are unintended, variable, and only estimated (not known exactly)									X
	MOA may not be the same at different levels of exposure. These are highly uncertain assumptions. It is more likely that all these factors would differ for different exposure levels				X					
	Pharmacokinetic processes may not be the same different exposure levels. These are highly uncertain assumptions. It is more likely that all these factors would differ for different exposure levels.				X					
	Size of exposed populations						4			
Interspecies extrapolation	Appropriate target tissue correspondence between humans and test animals?		1							
	Interspecies projection - chronic toxicity and/or acute toxicity and /or enzyme activities									X
	Interspecies differences in intake/absorption processes									X
	Interspecies differences in potentially interacting processes leading to specific types of cancers - that is the numbers and nature of "stages" in molecular pathological pathways to cancer in different organs of different species, and differences in background rates of transitions among these stages									X
	Mapping test animal life stages on human ages/exposure periods									X
	Pharmacodynamic differences at the target site would lead to different mutation rates between humans and test animals?		5							
	Pharmacokinetics differences in humans and test animals				2					
	Quantitative test animal-human differences in primary response effects. Are the particular suDNA adducts cleared more/less efficiently in humans? Do they have the same potential to induce critical mutations?				3					

Intra species variability	Age related differences in stem cell proliferation, DNA repair, and differentiation processes that affect sensitivity to the production of pro-carcinogenic transitions									X
	Age-Related differences in susceptibility to carcinogenesis									X
	Make-up of the exposed population Demographic make-up of population				X	4				
	Statistical uncertainty in the central estimates of the life-stage-specific sensitivity factors estimated in Hattis et al. (2004)									X
	Subjects of human epidemiological studies are subject to a variety of selection biases									X
	What are the correct values of mean and relative standard deviation for the susceptibility distribution in the population of interest?								1	
	What is the extent of human variability that would lead to different AUC at the target site, different mutation rates at the target site, and different "yields" of tumours per mutated cell (e.g., immune surveillance)?		3							
	Variabiltity in susceptibility among members of population General human inter individual					6				X
Mode of Action	Dose-response functions for steps in mode of action					5				
	No knowledge of the mode-of-action / mechanism Mode of action in humans Mode of action in animals				X		1 2			
	Steps, key events in a mode of action					2				
	Understanding of whether a chemical is mutagenic, directly DNA reactive, anuegenic etc. is critical					1				
Other	Are there other factors contributing to the response in the rat at the point of departure that would lead to a different slope at lower doses than at the POD (i.e., analogy to the dose-rate effectiveness factor in radiation risk assessment)?		6							
Pharmacokinetics	Pharmacokinetc variability. This generally encompasses several steps.					3				
Stochastic nature of cancer process	Data contained in extrapolations almost always fall into regular statistical distributions suggesting considerable regularity in biological processes									X
	Do different organs or types of tumour show specific distributions?								2	
	Etiologic mechanisms of the background cancer at the target tissue in the human population?		2							
	Primary response dose-transitions, such as induction or saturation of DNA repair processes in humans: at what dose-ranges do these occur, what is the extent of their impact. These are the potential sources of high-low dose nonlinearity!				1					
	Rate of mutation is elicited by the genotoxic mechanism of the active moiety?		4							
Toxicodynamics	Uncertainty about possible deviations from10 linearity in toxicodynamics			X						

Table 4: Experts who filled the online questionnaire were invited to cite their scientific arguments behind their judgments – these are listed below. The categorisation of experts' views as either an assumption or a scientific argument was carried out by the project team.

Assumption underlying expert judgment	**Scientific argument underlying expert judgment**	**Experts**								
		A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**I**
Basic assumption is of equal risk given equal AUC of the active moiety at the target site, and assumes that BMDL is already corrected for any differences of this sort. In the absence of any other information (see below), this is a reasonable assumption.	This is supported empirically by the very old established correlation between slope factors derived from cancer bioassays and those derived from epidemiology. A good specific example of this is Vinyl Chloride (genotoxic metabolite).		X							
It is also reasonable to assume linear extrapolation...	... given data from radiation, which epidemiologically is linear as far as one can measure. Moreover, among chemicals, genotoxic carcinogens are the closest analogy to radiation.		X							
	In addition, tobacco smoke provides another example where the dose-response appears linear as far as one can measure.		X							
	Also, substantial evidence both theoretic (multi-stage model) and empirical (e.g., radiation, classical mutagens) that mutations linearly increase risk of cancer.		X							
	Finally, there is no evidence of threshold from epidemiologic data on secondary cancers resulting from chemotherapy.		X							
Note however, in most cases, the low dose risk at environmental exposures is not a scientific question, since it almost invariably cannot be verified or falsified experimentally.			X							
The agent being dosed does not require metabolic activation, or its activation rate is dose-rate-limited.					X					
Uncertainty in clearance is ~ 4x.	Humans clear/eliminate agents up to about 4 times slower than rats.				X					
Uncertainty in response sensitivity of humans vs. rats ~ 10x.					X					
Much of the uncertainty is in animal-human differences, independent of dose.					X					
Assumed that also a carcinogen in other animal species, both male and female rats.					X					
Uncertainty from high-to-low dose extrapolation increases *somewhat* the further down the extrapolation, but difference between 100x and 100,000x in relative uncertainty is minimal.					X					
	Risk estimated is an average population risk, so inter-individual variability is not a factor.				X					
Considering the large number of modulating factors for carcinogenesis, the cumulative log-normal curve is a reasonable assumption for the distribution of	Individual susceptibility for cancer induction is controlled by numerous factors (*although the extent that they all operate within a population to determine susceptibility for a given cancer is unknown*) Factors that affect the steps between the intake of a genotoxic carcinogen and a								X	

susceptibilities.	mutant frequency have been addressed in the perspective. These include metabolic activation and detoxication of the carcinogen, rates of DNA repair, as well as rates of cell proliferation and death. Additional factors must now be added for the steps between mutation and cancer. Most important is the inheritance of constitutively activated oncogenes or inactive tumour suppressor genes. This can result in a reduction of the number of steps required for malignant transformation of a cell. For carcinogen dose-response relationships, it implies that individuals that are genetically predisposed require a lower dose to manifest the tumour within a defined period of exposure and observation. Differences for the number of mutations required may also explain differences in the latency period between exposure and tumour manifestation, such as between hematopoietic tumours and epithelial cancer.									
	What type of distribution is now expected for the end point "cancer", taking into account the large number of factors that sum up to define the individual susceptibility? The central limit theorem of statistics states that the sum of N independent variables becomes normally distributed with increasing N. Upon a multiplicative combination of susceptibility factors, a lognormal distribution would follow. Using this function, we can now estimate the drop in cancer risk with decreasing dose. Dose steps are expressed as multiples of the standard deviation (SD) of the normal distribution; the point of departure is the dose that halves the probability of staying tumour-free throughout the period of observation (TD50). Following the cumulative normal curve and stepping down 1, 2, 3, 4, or 5 SDs below the TD50 () mean), the risks are 0.16, 0.023, 1.3x10E-3, 3.2x10E-5, and 2.9x10E-7; that is, the drop in risk is not proportional to the drop in dose. Expressed in relative terms, the risk reduction factor increases at low doses. For instance, while the reduction factor between 1 and 2 SD below the mean is 7 (0.16/0.023), the decrease between 4 and 5 SD is associated with a risk reduction by a factor of more than 100. With decreasing dose, the risk rapidly drops to zero. Note that because of the logarithmic dose scale, the dose steps represent factors. On the basis of this type of dose-incidence relationship, the SD estimated from a log-normal distribution expressed as log (dose) becomes the dominant quantitative factor for risk extrapolation. The wider the distributions of the contributing susceptibility factors are, the flatter the dose-response curve is. This also means that data based on the heterogeneous human population will show larger coefficients of variation than data from animal bioassays.								X	
	Make an analogy with a directly observable variability where the specific chemical case to be evaluated can be seen as a member of a class of similar putatively analogous cases. The rationale for doing so is that processes related to chemical toxicity are common to many different chemicals and occur through a (limited) number of biological mechanisms.									X
Straight linear projection on a mg/kg basis										X
Linear, no-threshold low dose-response extrapolation assumed						X	X			
Children at 10X increased risk							X			

Table 5: List of advantages (highlighted in grey) and disadvantages of the identified alternative approaches that experts could follow when applying the low dose extrapolation (LDE) step; this information was disseminated by the experts during the first plenary session of the experts' workshop.

Area of interest	**Approach 1: LDE after extrapolating animal data to humans**	**Approach 2: LDE before extrapolating animal data to humans**	**Both approaches**
Data needed	More scientific data driven approach (e.g. Pharmacokinetics)	In view of lack of data, approach 2 is preferred	The final choice of approach would depend on availability of data; choice would need to be made on a case by case basis
	Better able to incorporate future information on individual susceptibility		
Low dose extrapolation step	Low dose extrapolation in humans more satisfying; involving many factors which are bound to be different in rats	How to go from "non-effect dose" (i.e. below point of departure – low dose) in rats to humans?	
	Makes inter-species extrapolation easier; kinetics' models are in the range of Point of departure doses.	Point of departure has a different meaning in rats from humans	
	Represents differences in the susceptibility of individuals in the population of interest	Not easy to take into account role of individual variation in susceptibility	
		Inter-individual variability in rats: removed by inbreeding?	
	Less use of animal Mode of Action model in the low dose extrapolation step	Allows direct use of animal Mode of Action information	
Perception - communication	Familiarity in place (particularly in USA)		
	Comfort to extrapolate the Point of departure from rats to humans		
	Rationale easier to explain to audience/s		
	Shifts focus to humans early in the process		
Uncertainties	Better approach to explain "*what I do not know*"		

Supplementary material

Appendix I: Elicitation document on "*Low dose-response relationships for genotoxic carcinogens – Contributing factors and uncertainties*" that was sent to the experts who participated in the online structured survey.

ELICITATION DOCUMENT

Low dose-response relationships for genotoxic carcinogens – Contributing factors and uncertainties

Brief overview of the problem

A rather large (theoretically infinite) number of low dose-response models for genotoxic carcinogens exist. Different countries and organisations support different approaches. For the moment, regulatory agencies in the USA, and possibly The Netherlands, favour the linear model. The UK and others do not like to make any extrapolation as they say it is too uncertain, and also they do not like to make estimates of cancer incidence that may alarm people when they are so uncertain. The EFSA has a similar opinion (EFSA, 2005) and that was the motivation behind developing the margins of exposure (MoE) approach – to have a measure of cancer risk other than estimated incidence. Different suggested models may be based on different physiological aspects regarding possible effects of genotoxic carcinogens; nevertheless, so far, they all lack a detailed, transparent, rigorous scientific rationale to justify their employment in risk assessment of genotoxic carcinogens. Also, some experts advocate the choice of a dose-response model on a "case by case" basis.

Questions to be answered in the expert elicitation exercise:

- What are the known and unknown underlying the different low dose-response models for genotoxic carcinogens?
- What is the rigorous scientific rationale to support the choice among the different dose-response models for genotoxic carcinogens in view of incomplete knowledge?

How do we aim to use the results?

The current document is an initial survey to elicit individual opinions from experts who have agreed to participate. We plan to analyse qualitative data obtained through elicited individual opinions and discuss further collected opinions in a structured expert workshop that is to be held in May 2011. Possible outcomes of this exercise and the workshop are: a) a decision tree to assign dose-response models for different sets of chemicals; b) a comprehensive list of uncertainties that burden such a decision tree.

Dissemination of results

- Experts' workshop report
- Abstract at the 47th Congress of European Society of the Toxicology, August 2011
- Invited presentation (*via* teleconference) at the Dose-Response Group of the Society of Risk Analysis, USA
- Publication in a peer-reviewed journal

Anonymity – confidentiality of participants

Please note, that your individual responses to this survey will remain anonymous and confidential.

This survey is divided into four parts:

1. In Part I, you are invited to report your professional affiliation, and to provide a declaration of interest.

2. In Part II, you are invited to provide information about your expertise.

3. In Part III, you are asked to indicate your **estimates** and **uncertainty** about the carcinogenicity of chemicals at low exposure doses.

4. In Part IV, you are asked to explain: a) the scientific rationale that supports your elicited judgements, and b) the scientific uncertainties that may burden those judgements.

Please remember that the deadline for submitting your completed survey is the **28th February 2011**.
If you have any further questions, please let us know by e-mail (villie.flari@fera.gsi.gov.uk) and we will try to clarify these as soon as possible.

Part I – Personal attributes and declaration of interest

Personal attributes

- Professional affiliation (please highlight one)
    - Academia, Government, Industry, Non Governmental Organisation, Research Center / Institute, Other
    - If "other", please give details:

Declaration of interests

- Please provide information on any potential interests that you may have (for example, membership of a committee providing advice on genotoxic carcinogens).

Part II – Information on expertise

- What is your expertise in the field of genotoxic carcinogens? Please, insert your text in the following table.

- What evidence (*i.e.* relevant information) are you aware of that would be useful when making judgments on low dose extrapolation for genotoxic carcinogens? Please, insert your text in the following table.

- What do you consider to be the key references (e.g. reports, guidance documents, articles, publications, etc.) when considering low dose extrapolation for genotoxic carcinogens? Please, insert your text in the following table.

Part III – Quantitative information

Question C.1

Given the point of departure for human dose-response (a rat BMDL10[7]: 10mg/kg) for a hypothetical genotoxic carcinogen and taking into account your knowledge on genotoxic carcinogens, could you give us your judgments on the exposure that could potentially lead to 10^{-6} (one in a million) extra cases of cancer per lifetime for the general population in your country?

We would like you to make your judgments in terms of how many orders of magnitude the dose is from BMDL10. Therefore, a judgment of -1 is equivalent to saying it is one order of magnitude less than the corrected BMDL10, and a judgment of 5 is equivalent to saying it is five orders of magnitude higher than the corrected BMDL10. Indicate your estimates and your uncertainty in the table below (units: none as we are referring to a difference in the log [BMDL10]):

Most likely	**90% interval**[8]		**Minimum**	**Maximum**
	Low	High		

Would your judgment be any different for any particular subpopulation in your country? If yes, please insert your judgments in the table below.

Subpopulation A:	**Most likely**	**90% interval**		**Minimum**	**Maximum**
		Low	High		
Subpopulation B:	**Most likely**	**90% interval**		**Minimum**	**Maximum**
		Low	High		

Please, enter any notes you may have for the above judgments below:

[7] The BMDL10 is the value that corresponds to the lower limit of a one-sided 95% confidence interval for a benchmark dose that shows a 10% increase.

[8] Indicate the interval in which you believe that the true estimate of number of cases of cancer per lifetime lies in 90 % of times.

Question C.2

Given your knowledge on genotoxic carcinogens, could you give us information on the number of cancer cases per lifetime that to your point of view may be attributed to genotoxic carcinogens for the general population in your country? Indicate your estimates and your uncertainty in the table below (*units:* number of extra cases of cancer per lifetime).

Most likely	**90% interval**		**Minimum**	**Maximum**
	Low	High		

Could these numbers be different perhaps according to different types of cancer?

	Most likely	**90% interval**		**Minimum**	**Maximum**
		Low	High		
Type of cancer A:					
Type of cancer B:					

Would any of your judgments above be any different for any particular subpopulation in your country? If yes, please insert your judgments in the table below.

Subpopulation A:	**Most likely**	**90% interval**		**Minimum**	**Maximum**
		Low	High		
Subpopulation B:	**Most likely**	**90% interval**		**Minimum**	**Maximum**
		Low	High		

Please, enter any notes you may have for the above judgments below:

Question C.3

Given the point of departure for human dose-response (a a rat BMDL10[9]: 15 mg/kg) for a hypothetical genotoxic carcinogen and taking into account your knowledge on genotoxic carcinogens, could you give us information on the number of cancer cases per lifetime that to your point of view may be attributed to a hypothetical genotoxic carcinogens for: a) the general population in your country assumed to be exposed at 0.001 mg/kg/day, or b) the population that works in the industry assumed to be exposed at 0.1 mg/kg/day. Indicate your estimates and your uncertainty in the table below (*units:* number of extra cases of cancer per lifetime).

	Most likely	**90% interval**		**Minimum**	**Maximum**
		Low	High		
Exposure for general population					

[9] The BMDL10 is the value that corresponds to the lower limit of a one-sided 95% confidence interval for a benchmark dose that shows a 10% increase.

Occupational exposure					

Could these numbers be different perhaps according to different types of cancer?

	Most likely	**90% interval**		**Minimum**	**Maximum**
		Low	High		
Type of cancer A:					
Type of cancer B:					

Would any of your judgments above be any different for any particular subpopulation in your country? If yes, please insert your judgments in the Table below.

Subpopulation A:	**Most likely**	**90% interval**		**Minimum**	**Maximum**
		Low	High		
Subpopulation B:	**Most likely**	**90% interval**		**Minimum**	**Maximum**
		Low	High		

Please, enter any notes you may have for the above judgments below:

<u>Part IV – reasoning behind the judgements</u>

Question D.1: Please lay out the scientific rationale that you would use to support your judgements, and list any further information you may have needed to know in order to refine your estimates.

Scientific arguments behind your judgments
Further information that would be helpful in refining your judgments

Question D.2: Please lay out the scientific uncertainties that burden your estimates/judgments and rank the uncertainties in terms of their significance. For the ranking, use 1 to indicate the most significant uncertainty, then 2 for the next most significant and so on.

Source of uncertainty in making judgments about low dose extrapolations for genotoxic carcinogens	**Rank**

Appendix II: Document used for elicitation of judgments about low dose extrapolation (Route 1) during the Experts' Workshop

Please fill in all of the boxes you can and make subsequent revisions with a different colour pen.

Expert's name:

Given a point of departure for human dose-response, could you give us your judgements on the exposure that could lead to 10^{-6} (one in a million) cases of cancer per lifetime for the general human population?

Minimum		Maximum
90% interval		
Low		*High*
Most likely		

Given a point of departure for human dose-response, could you give us your judgements on the exposure that could lead to 10^{-4} (one in ten-thousand) cases of cancer per lifetime for the general human population?

Minimum		Maximum
90% interval		
Low		*High*
Most likely		

Given a point of departure for human dose-response, could you give us your judgements on the exposure that could lead to 10^{-5} (one in a hundred-thousand) cases of cancer per lifetime for the general human population?

Minimum		Maximum
90% interval		
Low		*High*
Most likely		

Appendix III: Experts considered different lines of evidence and/or sources of their uncertainty before providing any quantitative estimates. Breakout group 1 assessed approach 2 first, whereas breakout group 2 assessed approach 1 first. The information elicited during the 1st breakout session is highlighted in grey. Text in italic indicates clarifications introduced by the project team. The classification of evidence lines in common themes (either within the breakout group and/or between the two breakout groups) was decided by the project team.

	Breakout group 1	**Breakout group 2**
Approach 1: LDE after extrapolating animal data to humans	Need to consider susceptibility factors when (*doing risk assessment*) in humans. Some variability in enzyme activity is 2-fold at max; strong disagreement among the rest of the experts. But (*there is a need to*) distinguish enzyme activity from (*the process of*) clearance which is less variable. Reminder doing genotoxic carcinogens. Mutagenicity is the important aspect for low dose linearity: (*decision making tree* - step 1) does that ever drive things? (*decision making tree* - step 2). If yes, what is the susceptibility variation? Expert still sceptical. Other expert clarified that the important susceptibility factors (if any) are those which affect mutation. What exactly is covered by susceptibility: genetics? Age, diet? Two experts do not consider diet to be a susceptibility factor, actually a modulating factor.	Other sources of inter-individual variability in susceptibility: • Variation in pharmacokinetics (less *in animals* than humans) • Immune *system* surveillance • Apoptosis • Stress-response pathways (in addition to DNA repair) • Proliferation of cells *results* supporting differences in individuals • Inter-individual differences in oncogenes and tumour suppressor genes (less in an inbred strain *than humans*)
		More environmental control in the rat experiments
		Inter-individual variability in DNA repair -experiments suggest (*uncertainty*) factor (less in rats than human, not much >1)

	Many issues are the same as (*when assessing approach 2*) before. Large random component in who gets tumours; therefore 90% not necessarily free from risk. Now (*when assessing this approach there is*) potentially more information about human variation but less information about dose-response.	
	Spanish 8-fold odds-ratio for tumours; other experts sceptical (confounding) and other literature, DNA reactivity not driving force or not many cancers caused by chemicals. Two experts concerned about multiple comparisons. One expert 8-fold is concerning but not 100-fold or 1000-fold. Also if 40% in the sensitive group then really only 4-fold compared to average. Back to 2-fold!	
		ED001 fish mega study indicates presence of threshold but *the dose* response at low exposure remains uncertain (Bailey et al., 2009).
		Results from Gary Williams *work on* aflatoxins and nitrosamines - Saw no effect levels; differences between adenomas and carcinomas (Williams et al., 1999, 2000, 2004, 2005).
		Mega mouse – bladder *is* non-linear, liver *is* not (Gaylor, 1979; Littlefield et al., 1980) BIBRA rat study – oesophagus is linear, liver is not (Peto et al, 1991)
	USA EPA did not regulate micro-organism of specific risk to HIV/AIDS subjects.	
	Who is being protected *currently*? Most sensitive? Some individuals deterministically get tumours quite young.	Epidemiological data • Fitting splines, occupational studies • Problem of high background rates

	(*Processes in place*) Not targeting really genetically unusual individuals. Have identified some sensitive sub-populations and whether should regulate for those.	• Vinyl chloride liver cancer (US EPA, 1997)
	What chemicals might this apply to? One expert thinks there are some, really a matter of principle rather than examples.	
	Issue of differences in cell state and relevance to tumours.	
		Is pharmacokinetics linear extrapolating below point of departure (PoD)? Depends whether chemical needs metabolic activation (*in which case* it will be non-linear); if it does not need activation (*the*) default = (*is*) likely (*to be*) = linear.
	Cannot separate these sources.	
	Distinctions are only semantic. Highly multi-dimensional.	
	Discussion of research about sources of cancer risk.	
		For all of the lines of evidence, not sure about the numbers that are attached for UFs. Need information on the mode of action of the chemical. Impossible to quantify the counterbalancing mechanisms of the cell.
extrapolating animal	**Historical: linear dose-response (from radioactive materials, *the conclusion is* one hit=one cancer)**	**Inter-individual variability in DNA repair -experiments suggest (*uncertainty*) factor (less in rats than human, not much >1)**
	Knowledge that at low levels very efficient repair for some chemicals (Tominaga et al., 1997)	Is pharmacokinetics linear extrapolating below PoD? Depends whether chemical needs metabolic activation (*in which case* it will be non-linear); if it

		does not need activation (*the*) default = (*is*) likely (*to be*) = linear.
	Data on some substances (EMS and others, e.g. Aflatoxin; Williams et al., 1999, 2000, 2004, 2005).	Other sources of between individual variability in susceptibility. • Variation in pharmacokinetics (less *in animals* than humans) • Immune *system* surveillance • Apoptosis • Stress-response pathways (in addition to DNA repair) • Proliferation of cells *results* supporting differences in individuals Inter-individual differences in oncogenes and tumour suppressor genes (less in an inbred strain *than humans*)
	One case from BMD10 to BM0.001 – trout (Bailey et al., 2009)	More environmental control in the rat experiments
	One case to BMD0.01 – mice (Gaylor, 1979)	Slope of dose-response in the experimental data
	Bacteria (Ames, 1974; Abril et al., 1994; Chen et al., 1998; Sikola et al., 2010) Models of the relationship (Peto et al, 1991; Williams et al., 1999; Williams et al., 2000, Williams et al., 2004; Williams et al., 2005; Bailey et al., 2009; Waddell, 2003a, 2003b; Littlefield et al, 1980; Lutz, 2003)	ED001 fish mega study indicates presence of threshold but response at low exposure remains uncertain (Bailey et al., 2009).
	Strain/species differences for aflatoxin dependent on liver function (Williams et al., 1999, 2000, 2004, 2005)	*Results from* Gary Williams *work on* aflatoxins and nitrosamines - Saw no effect levels; differences between adenomas and carcinomas (Williams et al., 1999, 2000, 2004, 2005)
	What is a "ridiculous" dose: • *Exposure* levels *are usually* below what is already present in body or needed for *the course of* human life. • Another expert: not that. Everything s/he knows argues against linear down as far as 1 in a million. First expert: surprised by how conservative s/he is when just being scientific.	Mega mouse – *cancer in* bladder thresholded, *but cancer in* liver not ; *therefore the* different endpoints *may imply* = different dose-responses (Gaylor, 1979; Littlefield et al., 1980)

	Feels quite different about carcinogens in general. Trout study shows non-linearity but mouse study more equivocal.	
		Mega rat – liver linear for low dose,
	Key chemical properties *that need to be taken into account when assessing low dose extrapolation* • knowledge of actual dose • other toxicity • tissue affected • detoxification at low dose • competing rates Some chemicals have been "studied to death"	Problem with high background *of cancer cases* (different across strains)
	Default of *uncertainty factor of* 100 not necessarily conservative	
	Very little evidence *exists for* (from) human population other than worker studies (especially vinyl chloride)	
	Special view of the world of one expert: • Dose-response is fundamentally a susceptibility distribution • Log-normal to reflect multiplication of susceptibility factors (central limit theorem in statistics); susceptibility factors are effectively (varying between individuals) conditional probabilities of successive biological stages towards development of actual tumours. • Slope of function is about 3, i.e. each reduction of dose by factor of 3 reduces the incidence by moving 1 standard deviation in normal distribution. • Therefore 1:100000 (linear from BMD_{10} to 1 in a million) is c.12	

	standard deviations (c. 2 standard deviations per order of magnitude) which implies negligible chance of response. • Mechanism only important if affects amount of inter-individual variability • Mantel-Bryan dropped historically "by mistake" due to stochasticity assumption, also due to lack of human data. Adduct formation probably proportional at low dose. But very different from mutation rate and tumour incidence.	
	Historically *there are* two approaches for *high to low* extrapolation: • Cancer: purely stochastic => linear dose-response. Good theory but situation more complex. • Non-cancer: susceptibility distribution approach. Inter-individual variability is the driver. • In reality both *are* operating: o *The* stochastic component real and inter-individual variability in effect. o *There is* poor knowledge of balance between them. o *It* would be good to sort out combining the two approaches • Practical/pragmatic approach *suggested*: Start with linearity and consider departures. Various models of departures: saturation of clearance, induction of repair, over-induction of repair, induction of cyto-toxicity, cell (+division) slow-down, age effects.	
	Sources of uncertainty: • only significant source of uncertainty (also variability between chemicals?) is about standard deviation of log-normal	

	• might be 3 +/- .5 but needs more consideration • World view of other experts • For relative doses down to PoD/10000, have some direct empirical evidence; key uncertainty is about extrapolation to other chemicals/species. o Below PoD/10000, have (partially) quantified mechanistic biological models of dose-response for quantitative markers but not for tumours ▪ Empirical quantitative evidence available for some such models showing sub-linear dose-response ▪ Lack mechanistic model for dependence of tumour incidence on markers ▪ Mathematically possible for sub-linear models of markers to combine to give linear or supra-linear dose-response for tumours. Supra-linear low dose-response mostly discounted (except by one expert).	