USE OF BENCHMARK DOSES FOR REPROTOXIC CHEMICALS

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OBJECTIVES

An interdisciplinary working group was created by the French Agency for Environmental and Occupational Health Safety (AFSSET) to define a French method for the derivation of human health toxicity values (HTV) for reprotoxic chemicals. Within this framework, we conducted a pilot study to evaluate the interest of using the benchmark dose (BMD) approach and its one-sided 95% lower confidence limit (BMDL) as the critical dose, instead of the usual NOAEL/LOAEL. We tested the construction of BMD/BMDL in the case of 3 reprotoxic chemicals for which the toxicological database isn't quantitatively and qualitatively equivalent: di-n-Butyl phthalate (DnBP), linuron and 2-ethoxyethanol (EGEE).

METHODS

The toxicological profile and the choice of the critical effect and study for each substance was identified (Table 1). The data were adjusted for the derivation of BMDLs, using BMDS Software from the US Environmental Protection Agency (1.3.2. version). Several mathematical models were tested each time. The data were adjusted with the maximum likelihood approach.

Table 1: studies used to test the BMD/BMDL approach in the context of HTV derivation

	DnBP	Linuron	EGEE
Study	Lee *et al.* 2004 (*"Diverse developmental toxicity of DnBP in both sexes of rat offspring after maternal exposure during the period from late gestation through lactation"*)	McIntyre 2000 (*"Effects of in utero exposure to linuron on androgen-dependant reproductive development in the male Crl:CD(SD)BR rat"*)	Doe *et al.* 1984 (*"Ethylene glycol monoethyl ether and ethylene glycol monoethyl ether acetate teratology studies"*)
Critical effect	Reduction of spermatocyte development	Testis hypoplasia	Fetal defects
Species/ Exposure	Female rat - diet GD15 – PND21	Female rat – gavage GD12 – GD21	Female rabbit – inhalation GD6 – GD19
Protocol	4 dose groups + control 8 males (dams) examined per group	3 dose groups + control 44-69 males (dams) examined per group	3 dose groups + control 96-136 dams examined per group

RESULTS

The approach taking into account the issue of "litter effect" couldn't be used because the data wasn't available in the papers analysed.

DnBP: In males, a reduction of spermatocyte development was observed from 20 ppm with dose-dependant increased incidence and/or severity. A $BMDL_{10}$ was estimated using the gamma and weibull model at $4{,}4.10^{-5}$ mg/kg/j. It is several orders of magnitude less than BMD_{10} (Table 2). This difference reveals the large uncertainty which is due to the low number of dams examined (8 per group) and to the level of response observed for the first dose tested (50%). The study failed to identify a NOAEL. It questions the use of BMD versus LOAEL and it reveals a low confidence level in the derivation of a HTV with these data.

Table 2: comparison of BMD, BMDL and LOAEL for DnBP

NOAEL	$BMDL_{10}$	BMD_{10}	LOAEL
-	$4{,}4.10^{-5}$ mg/kg/d	0,08 mg/kg/d	2 mg/kg/d

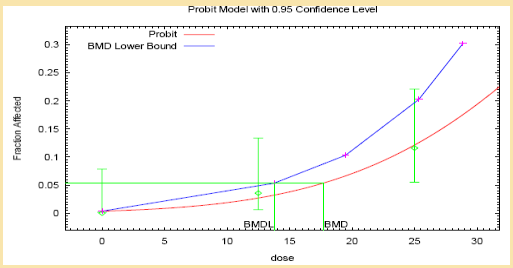
Linuron: No NOAEL was identified in the study (LOAEL=12,5 mg/kg/j, first dose tested). An experimental point (corresponding to 50 mg/kg/j) for linuron had to be deleted because no model adjusted suitably with the data (figure 1). The $BMDL_{05}$ calculated with the probit model is slightly higher than the LOAEL and the BMD_{05} (Table 3). It reveals the low uncertainty and the high confidence to derive a HTV. However, we note that the interpretation of these critical doses in health risk assessment differs: the $BMDL_{05}$ is a value for which we are sure (at 95%) it produces 5% of effect or less, the LOAEL is a value for which we are almost sure it produces an effect, without precise quantification.

Table 3: comparison of BMD, BMDL and LOAEL for linuron

NOAEL	LOAEL	$BMDL_{05}$	BMD_{05}
-	12,5 mg/kg/d	14 mg/kg/d	18 mg/kg/d

EGEE: The BMD/BMDL for fetal defects was estimated using the logit and probit models. 10% extra risk was selected as the benchmark response.

Figure 1: estimation of the BMD and BMDL for linuron



A BMD_{10} of 20,5 mg/m³ and a $BMDL_{10}$ of 0,37 mg/m³ were estimated. The LOAEL and the NOAEL were respectively 644 and 184 mg/m³. The data show a level of response of 50% for the control group for fetal defects and a level of 70% for the LOAEL. So the BMD_{10} (extra risk) associated with a level of 55% of response, is largely less than the LOAEL. It can be explain by the high response rate in the control group (50%) and the variability of the response observed in the first dose groups. It questions the use of this experimental study for the derivation of the HTV.

Table 4: comparison of BMD, BMDL, NOAEL and LOAEL for EGEE

$BMDL_{10}$	BMD_{10}	NOAEL	LOAEL
0,37 mg/m³	20,5 mg/m³	184 mg/m³	644 mg/m³

DISCUSSION - CONCLUSION

The main issue reveals frequent uncertainties in the use of BMDL for the derivation of HTVs. It raises queries as to:

- data availability (see "litter effect")
- data quality (deletion of experimental points, variability in the response),
- uncertainty (BMDL often several orders of magnitude less than BMD),
- BMDL interpretation (the level of response is not explicitly "zero").

These examples cannot illustrate all the cases and answer all the questions. It emphasizes the need for continuing such work and to have a critical feedback in the derivation of HTVs. This work highlighted the need for having exploitable data. The REACH regulation will be able to provide such studies.

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