

Output for Risk assessment/characterisation purposes – specific guidance for the individual endpoints – to be inserted under:

1.5.2 Concluding on suitability for Chemical Safety Assessment in the detailed end-point guidance on carcinogenicity (RIP 3.3-2)

CARCINOGENICITY

BACKGROUND

Carcinogenicity - threshold and non-threshold effects

Considerations of the kind of dose-response relationship that is at hand for a particular agent and of possible mechanisms of action are important components of a risk assessment for carcinogenicity. Substances which are carcinogens have conventionally been divided into two categories according to the presumed mode of action: genotoxic and non-genotoxic.

Non-genotoxic carcinogens are believed to exert their carcinogenic effects through mechanisms other than genotoxicity. There are many different modes of action thought to be involved in non-genotoxic carcinogenicity. However, it is normally assumed that the modes of action of non-genotoxic carcinogens can be associated with threshold doses, and it may also be possible to define no-effect levels for the underlying toxic effects of concern.

In contrast, it is usually considered that an effect-threshold cannot be identified for most genotoxic carcinogens, i.e. it is not possible to define a “no-effect level”. However, it is also recognized that for certain (a few??) genotoxic carcinogens a threshold may be shown to exist for the underlying genotoxic effect implying a threshold also for any resulting cancer effect.

The default assumption for carcinogenic chemicals with genotoxic activity is that they have a linear non-thresholded dose-response relationship. However, both direct and indirect mechanisms of genotoxicity can be non-linear (i.e. supra-linear or sub-linear) and occasionally even truly thresholded. Thus, sometimes the default assumption of linear dose-response for genotoxicity and thus also for carcinogenicity may not be warranted.

Examples of mechanisms of genotoxicity that may be demonstrated to lead to non-linear or thresholded dose-response relationships for genotoxicity include inhibition of DNA synthesis, alterations in DNA repair, overloading of defence mechanisms, interaction with microtubule assembly leading to aneuploidy, topoisomerase inhibition, high cytotoxicity, metabolic overload, low dose/high dose metabolic shift and some physiological perturbations.

Different modes of carcinogenic action warrant dissimilar risk assessment

approaches

Both genotoxicity/mutagenicity and carcinogenicity induced by a chemical agent may be characterised as a threshold or a non-threshold adverse effect. This distinction implies paramount differences in the way risk characterisation must be conducted (Table X).

Table X

	Genotoxicity	Carcinogenicity
Threshold dose-response	<u>Yes</u> Genotoxic agent * NOAEL(s) for genotoxicity may sometimes be identified ¹	<u>Threshold carcinogen</u> * NOAEL may be identified * DNEL(s) may be determined
	<u>No</u> Non-genotoxic agent	* DNEL<-> exposure ratio approach feasible in risk characterisation
Non-threshold (linear) dose-response	<u>Yes</u> Genotoxic agent * NOAEL for genotoxicity theoretically non-existent	<u>Non-threshold carcinogen²</u> * NOAEL theoretically non-existent * No DNEL can be determined * Qualitative risk assessment applies

¹ This kind of “threshold mutagenicity” requires that special mechanistic knowledge is available showing that the underlying toxic effect elicits a definable threshold – i.e. a NOAEL can be identified.

² In cases where it can be shown that a threshold is likely to exist for the underlying genotoxic effect, a genotoxic carcinogen may be rendered a thresholded, non-linear dose-response. In such cases, it may be feasible to apply the DNEL<-> exposure ratio approach in risk characterisation.

The UN institution IPCS (International Program on Chemical Safety) has developed a conceptual framework to provide a structured and transparent approach for the assessment of the overall weight of evidence for a postulated mode of action for a carcinogen and also for individual tumour types observed (Sonich-Mullin et al., 2001). This was based partly on the so called Hill criteria for causality from epidemiological data (Hill, 1965) as modified by Faustman (1997) for developmental toxicity. The framework promotes confidence in the conclusions reached by the use of a defined procedure, which mandates clear and consistent documentation of the reasoning used and inconsistencies and uncertainties in the available data.

Normally, a lack of positive responses in appropriate genotoxicity/mutagenicity assays would imply a threshold dose-response for a carcinogenic substance. The hazard assessment may then be concluded based on an appropriate NOAEL. DNEL(s) may be derived after correction of the starting point and application of appropriate assessments factors in a fashion that is similar to the procedures used for e.g. the end-point Repeated Dose Toxicity (c.f. Section XX above).

For carcinogenic substances with genotoxic activity, where the default assumption of linear, non-thresholded dose-response relationships applies, a true NOAEL do not exist and consequently no DNEL can be calculated. For such cases, the general provisions for chemical safety assessment in REACH (Annex I, point 6.5) foresee a “qualitative” approach to be applied. Thus, a qualitative assessment of the likelihood that effects are avoided when implementing the exposure scenario shall be carried out.

The concept of “adequate control” (i.e. exposure < DNEL) do thus not seem to apply for non-threshold carcinogens. Meanwhile, the general provisions to ensure that such substances do not adversely affect human health (REACH Article 1.3) would implicitly call for a low risk [i.e. a {*qualitatively?*} high likelihood that risks are avoided should be shown for each exposure scenario].

The possibility of deriving “DMELs” or “DAELs” (Derived Minimum Effect Level // Derived Acceptable Effect Level) for some non-threshold agents such as genotoxic carcinogens has been discussed. Such values might be used in similar ways as the DNELs – i.e. to show that “adequate control” is at hand. The “qualitative assessment” mentioned above could thus be substituted (or possibly substantiated) with a more straight forward procedure. Adequate control (or high likelihood that risks are avoided) would have been arrived at if exposures can be shown to be < DMEL.

However, the application of the DMEL-approach for risk characterisation would require a revision of parts of REACH Annex I. In particular points 6.3-5 in Annex I would have to be amended. A review of Annex I shall be carried out by the CION within 12 months after the entry into force of REACH according to REACH Article 137.4. Appropriate amendments to Annex I are to be adopted via regulatory comitology.

It should be noted that such an amendment to the REACH provisions might have consequences not only for risk assessment of non-threshold agents, but may also have bearing on the granting of authorisations for e.g. CMR-substances (Article 59.2-4). A non-threshold carcinogen could presently not gain an authorisation via Article 59.2 since “adequate control” cannot be shown in the absence of a DNEL.

The remaining possibility is authorisation via Article 59.4, but then a more thorough assessment of risks, socio-economic benefits and alternatives (substitution!) including their risks would be required. It should be noted that these issues are highly controversial and EP, the Council and CION seem presently to have divergent opinions.

For the purpose of the discussions in the DNEL Drafting group it is anticipated in this paper that the quantitative DMEL-approach for risk characterisation of non-threshold carcinogens will be allowed for in REACH.

1 DERIVATION OF TYPICAL DOSE DESCRIPTORS – [STEP 1 GENERAL DNEL CHAPTER]

1.1 Typical dose descriptors

The task – according to Dinant’s paper [Proposed structure DNEL part endpoint chapters.doc](#) :

- *Derivation of typical dose descriptors [e.g. NOAELs, BMD, LD50, T25, TD50....] from all available studies*

The data available for hazard assessment of carcinogenicity may be life-time rodent bioassay results or human data from epidemiological studies.

1.1.1 Threshold carcinogens

The task:

- *Describe the most frequently used dose descriptor(s) for risk assessment purposes for this end-point.*
- *If it is sometimes (or always) not possible to identify a dose descriptor for this endpoint: describe other ways than the dose-response for this end-point is normally addressed in a risk assessment*

1.1.1.1 Animal data

Risk characterisation of non-genotoxic and some genotoxic threshold carcinogens generally relies on the identification of a dose or a concentration below which cancer effects are unlikely to occur in relevant experimental animal studies. Besides the NOAEL for the tumour end-point(s) in appropriate long-term cancer bioassays, the NOAEL for an identified underlying toxic effect, derived from the bioassay and/or other experimental/mechanistical studies, may serve as a direct measure for the threshold (NOAEL) dose.

If a NOAEL has not been identified, the LOAEL might be used as a starting point for risk assessment. Alternatively, a benchmark dose (BMD) can be determined if possible. The use of BMD-methodology is preferred when LOAEL to NOAEL extrapolation is performed.

1.1.1.2 Human data

In exceptional cases when reliable, positive human data is available (including adequate exposure data), such as a comprehensive epidemiological data-base, these data might be used analogously to experimental data from bioassays for carcinogenicity to derive a LOAEL.

Sometimes it is possible to use reliable negative epidemiological data to calculate the maximum cancer risk that would have remained undetected due to the (limited) statistical power of the study. It may be informative to compare such maximum cancer risks with results from calculations based on experimental (animal) data. Often such negative epidemiological data are compatible with positive experimental data due to relatively low human exposures and the low sensitivity of most epidemiological studies.

The distinction between threshold and non-threshold carcinogens must be maintained when the derivation of dose descriptors based on human data is contemplated.

1.1.2 Non-threshold carcinogens

The task:

- *Describe how the dose-response is normally reflected quantitatively (e.g. T25 or BMDL10 for non-threshold carcinogens), semi-quantitatively (e.g. high, medium, low c.f. potency group for specific conc. limits <1 ; 1-100; >100 mg/kg/d) or qualitatively*

1.1.2.1 Animal data

For non-threshold, genotoxic carcinogens no NOAELs can be determined since exposure levels above zero without a proportional carcinogenic risk are not considered to exist.

However, various measures of *carcinogenic potency* may be derived from the available data. Commonly used dose descriptors for carcinogens are described in the table below.

Denomination	Description	Common units
T25	Tumorigenic dose – 25% tumour incidence Interpolated incidence	<ul style="list-style-type: none"> • [mg/kg/day] • [mg/m³] • [ppm]
TD50	Tumorigenic dose – 50% Calculated dose resulting in 50 % of exposed animals remaining tumour free after 2 years	<ul style="list-style-type: none"> • [mg/kg/day] • [mg/m³] • [ppm]
BMD10(05)	Benchmark dose – 10(5) % Calculated dose using all data points in a multi-dose bioassay	<ul style="list-style-type: none"> • [mg/kg/day] • [mg/m³] • [ppm]
BMDL10(05)	Benchmark dose lower 10(5) % confidence interval value Calculated dose using all data points in a multi-dose bioassay accounting for experimental error	<ul style="list-style-type: none"> • [mg/kg/day] • [mg/m³] • [ppm]

These measures of carcinogenic potency are most of all useful as dose descriptors for systemic tumours occurring after exposure via any route. For locally active carcinogens the calculation and application of these dose descriptors is problematic, with a possible exception for the inhalation route.

For non-threshold end-points such as genotoxic carcinogenicity, the likelihood that effects are avoided when implementing the exposure scenario shall be qualitatively assessed. Although not yet catered for in the REACH legislation, quantitative approaches may also be useful and might be applied after a revision of REACH - e.g. Annex I.

In the qualitative approach there is no estimation of specific levels of risk in a given exposure scenario and emphasis is placed on assessing the adequacy of control of exposure in the human population of interest (e.g. workers, consumers, or humans exposed indirectly via the environment). The qualitative approach directly proceeds to the level of consequences without special consideration of the risk characterisation and evaluation part of the risk assessment process. This approach is similar to the ALARA-principle (*as-low-as-reasonably-achievable*) originally used in the area of radiation protection.

This approach is unpredictable and may in individual cases lead to overly protective as well as poorly protective risk management measures. Due regard is not taken to the widely varying carcinogenic potencies of chemical carcinogens. Note that the carcinogenic potency may vary by four to six orders of magnitude (x 10000-1000000) among the presently known carcinogens.

In the qualitative approach to hazard assessment and risk characterisation the measures of *carcinogenic potency* described in Table X may be used to semi-quantitatively rank the strength of the carcinogenic activity into potency groups such as high – medium – low. Cut-offs between such potency groups have been suggested at 1 mg/kg/day and 100 mg/kg/day, respectively.

If a quantitative approach is performed, it is proposed to derive a DMEL, i.e. a Derived Minimal Effect Level, while addressing methodological uncertainties. The DMEL could be chosen as the dose associated with some generally accepted lifetime cancer risks (e.g. of 10^{-4} , 10^{-5} and 10^{-6}) calculated from a predetermined, moderate cancer risk – e.g. a T25 or a BMD10 cancer potency value [derived e.g. from an rodent long-term cancer bioassay or reliable human cancer data from epidemiological studies] via a linear extrapolation procedure.

A similar quantitative approach is based on an assessment of the Margin-of-Exposure [MoE]. MoEs are arrived at as a ratios between exposures and a predetermined, moderate cancer risk – e.g. the T25 or BMD10 cancer potency value derived e.g. from a rodent long-term cancer bioassay or reliable human cancer data from epidemiological studies.

Alternatively, the T25 or BMD10 cancer potency values could be divided by a special assessment factor for *high-to-low-dose* extrapolation may be applied in addition to conventional assessment factors for e.g. inter- and intra-species variation.

The accepted risk levels (e.g. of 10^{-4} , 10^{-5} and 10^{-6}), appropriate magnitudes of MoEs as well as the magnitude of the additional *high-to-low-dose* extrapolation assessment factor would have to be harmonised and accepted at the policy level.

1.1.2.2 Human data

In exceptional cases when reliable, positive human data is available (including adequate exposure data), such as a comprehensive epidemiological data-base, these data might be used analogously to experimental data from bioassays for carcinogenicity to derive a LOAEL or a T25-value (or a potency value for another tumour incidence than 25% - e.g. T10 etc.).

Sometimes it is possible to use reliable negative epidemiological data to calculate the maximum cancer risk that would have remained undetected due to the (limited) statistical power of the study. It may be informative to compare such maximum cancer risks with results from calculations based on experimental (animal) data. Often such negative epidemiological data are compatible with positive experimental data due to relatively low human exposures and the low sensitivity of most epidemiological studies.

The distinction between threshold and non-threshold carcinogens must be maintained when the derivation of dose descriptors based on human data is contemplated.

1.2 Considerations on the use of alternative data for hazard assessment purposes

The task:

- Describe any considerations about which alternative data could be used for risk assessment purposes.
- This is a key interface to RIP 3.3 end-point groups, which in a next step should reflect and/or add to this.

This part of the guidance remains to be elaborated

1.2.1 *Threshold carcinogens*

1.2.1.1 **Animal data**

- Short and medium term bioassay data (e.g., rat liver foci model, neonatal mouse model)
- Transgenic rodent models (e.g., XPA^{-/-}, p53^{+/-}, Tg.AC);
- Genotoxicity studies *in vivo*;
- Repeated dose toxicity tests;
- Studies on the induction of sustained cell proliferation;
- Studies on immunosuppressive activity;
- Studies on toxicokinetics;
- Other studies on mechanisms/modes of action, e.g. OMICs studies (toxicogenomics, proteomics, metabonomics & metabolomics).

1.2.1.2 ***In vitro* data**

- Genotoxicity studies;
- *In vitro* cell transformation assay results
- Mechanistic studies, e.g. on:
 - Cell proliferation;
 - Altered intercellular gap junction communication;
 - Hormone- or other receptor binding
 - Other targeted mechanisms of action;
 - Immunosuppressive activity.

1.2.1.3 **Non-testing data**

SAR, QSAR, chemical categories, read across

1.2.2 *Non-threshold carcinogens*

1.2.2.1 **Animal data**

See above under 1.2.1 *Threshold carcinogens*.

1.2.2.2 ***In vitro* data**

See above under 1.2.1 *Threshold carcinogens*.

1.2.2.3 **Non-testing data**

See above under 1.2.1 *Threshold carcinogens*.

2 MODIFICATION OF THE STARTING POINT – [STEP 2 GENERAL DNEL CHAPTER]

2.1 Issues relevant for modification of the starting point

The task:

- *When necessary, modification of the dose descriptor to the correct starting point)*
- *NB! only fill in specific issues for this end-point, i.e. no need to repeat issues from the general DNEL chapter*
- *Describe any particular issues to take into account for this end-point in relation to modification of the starting point (only relevant if there is a quantitative dose-descriptor).*

2.1.1 Specific guidance

No specific issues identified. The general text would apply ??????

3 ASSESSMENT FACTORS – APPLICATION OF ASSESSMENT FACTORS TO THE CORRECTED STARTING POINT TO OBTAIN THE ACCEPTABLE DAILY LEVEL (ADL) – [STEP 3 GENERAL DNEL CHAPTER]

The task:

- *NB! only fill in specific issues for this end-point, i.e. no need to repeat issues from the general DNEL chapter*
- *Describe any particular issues to take into account for this end-point in relation to the use of assessment factors and particular uncertainties to be aware of for this end-point.*
- *For end-points to be assessed qualitatively (no quantitative dose descriptor): describe particular issues (e.g. in relation to uncertainties and potency) that should be reflected in a discussion on the dose-response for that end-point.*

3.1 Application of assessment factors to the dose descriptor for threshold carcinogenicity

3.1.1 General assessment factors

Traditional assessment factors are only applicable for hazard assessment of systemically and/or locally active thresholded mutagenic and/or carcinogenic effects. If a threshold dose-response can be reliably shown, the hazard assessment is concluded based on an identified NOAEL. Appropriate DNEL(s) may be derived after correction of the starting point and application of appropriate assessments factors in a fashion that is similar to the procedures used for the endpoint Repeated Dose Toxicity (c.f. Section XX above).

3.1.2 Specific assessment factors

The potentially high degree of severity of the carcinogenic effects must be recognised. This should be reflected in the choice of assessment factors for dose-response and normally an AF >> 1 would be warranted to cater for endpoint-severity.

3.2 Application of assessment factors to the dose descriptor for non-threshold carcinogenicity

Traditional assessment factors are normally only applicable for hazard assessment of systemically and/or locally active thresholded carcinogenic effects. However, if the

quantitative approach using a special assessment factor for *high-to-low-dose* extrapolation is applied (see section 1.1.2.1 above) the magnitude of such assessment factors would have to be harmonised and accepted at the policy level. The conventional assessment factors for e.g. inter- and intra-species variation would apply analogously to other end-points such as Repeated Dose Toxicity.

4 IDENTIFICATION OF THE CRITICAL MEASURE FOR DOSE-RESPONSE (DNEL(s), DMEL(s) OR “QUANTITATIVE” – [STEP 4 GENERAL DNEL CHAPTER]

Remains to be elaborated ...

Above some instances have been described where a NOAEL, LOAEL or a measure of carcinogenic potency (e.g. T25) can be calculated for carcinogenic substances. For the derivation of the corresponding DNEL(s) and DMEL(s) for systemically and/or locally active carcinogenic effects, in principle all available hazard information (in accordance with Annexes V-IX) needs to be evaluated. In order to select the leading health effect and thus the most relevant DNEL/DMEL for a given exposure pattern/route the lowest DNEL/DMEL for a given route, exposure pattern and population is to be used for risk characterisation for each exposure scenario. This value shall also be communicated down the supply chain via the SDS.