6.3: Dose-Response Assessment

Dose-Response Assessment

The dose-response assessment step of the risk assessment process quantitates the hazards that were identified in the previous step. It determines the relationship between dose and incidence of effects in humans. There are normally two major extrapolations required:

1. From high experimental doses to low environmental doses.
2. From animal doses to human doses.

The procedures used to extrapolate from high to low doses are different for assessing carcinogenic effects and noncarcinogenic effects:

- **Carcinogenic effects** in general are not considered to have a threshold and mathematical models are generally used to provide estimates of carcinogenic risk at very low dose levels.
- **Noncarcinogenic effects** *(for example neurotoxicity)* are considered to have dose thresholds below which the effect does not occur. The lowest dose with an effect in animal or human studies is divided by safety factors to provide a margin of safety.


Risk assessment and risk management process, with the dose-response assessment phase highlighted

Figure 6.3.1. Dose-response assessment is a step in the risk assessment process
(Image Source: ORAU, ©)

Carcinogen (Cancer) Risk Assessment

Cancer risk assessment involves two steps:

1. **Perform qualitative evaluation of all epidemiology studies, animal bioassay data, and biological activity *(for example, mutagenicity)*.**The substance is classified as to its carcinogenic risk to humans based on the weight of evidence. If the evidence is sufficient, the substance may be classified as a definite, probable or possible human carcinogen.
2. **Quantitate the risk for those substances classified as definite or probable human carcinogens.** Mathematical models are used to extrapolate from the high experimental doses to the lower environmental doses.

The two primary cancer classification schemes are those of the **Environmental Protection Agency** (EPA) and the **International Agency for Research on Cancer** (IARC). The EPA and IARC classification systems are quite similar.

1. Qualitative Evaluation of Cancer Risk

The EPA's cancer assessment procedures have been used by several Federal and State agencies. The Agency for Toxic Substances and Disease Registry (ATSDR) relies on EPA's carcinogen assessments. A substance is assigned to one of five descriptors shown below in Table 6.3.1.

Descriptor	Definition
Carcinogenic to Humans	Strong evidence of human carcinogenicity
Likely to Be Carcinogenic to Humans	Evidence is adequate to demonstrate carcinogenic potential to humans but does not reach the weight of evidence for the "Carcinogenic to Humans" descriptor.
Suggestive Evidence of Carcinogenic Potential	The weight of evidence is suggestive of carcinogenicity; a concern for potential carcinogenic effects in humans is raised, but the data are judged insufficient for a stronger conclusion.
Inadequate Information to Assess Carcinogenic Potential	Available data are judged inadequate for applying one of the other descriptors. Additional studies generally would be expected to provide further insights.
Not Likely to Be Carcinogenic to Humans	Available data are considered robust for deciding that there is no basis for a substance to be considered a human carcinogen.

Table 6.3.1. Hazard Descriptors from the EPA's Guidelines for Carcinogen Risk Assessment (March 2005)

Cancer Data for Humans

The basis for **sufficient human evidence** is an epidemiology study that clearly demonstrates a causal relationship between exposure to the substance and cancer in humans.

The data are determined to be **limited evidence in humans** if there are alternative explanations for the observed effect.

The data are considered to be **inadequate evidence in humans** if no satisfactory epidemiology studies exist.

Cancer Data for Animals

An increase in cancer in more than one species or strain of laboratory animals or in more than one experiment is considered **sufficient evidence in animals**. Data from a single experiment can also be considered sufficient animal evidence if there is a high incidence or unusual type of tumor induced. Normally, however, a carcinogenic response in only one species, strain, or study is considered as only **limited evidence in animals**.

2. Quantitative Evaluation of Cancer Risk

When an agent is classified as a Human or Probable Human Carcinogen, it is then subjected to a **quantitative risk assessment**. For those designated as a Possible Human Carcinogen, the risk assessor can determine on a case-by-case basis whether a quantitative risk assessment is warranted.

The key risk assessment parameter derived from the EPA carcinogen risk assessment is the **cancer slope factor**. This is a toxicity value that quantitatively defines the relationship between dose and response. The cancer slope factor is a plausible upper-bound estimate of the probability that an individual will develop cancer if exposed to a chemical for a lifetime of 70 years. The cancer slope factor is expressed as mg/kg/day.

Linearized Multistage Model (LMS)

Mathematical models are used to extrapolate from animal bioassay or epidemiology data to predict low-dose risk. Most assume linearity with a zero threshold dose.

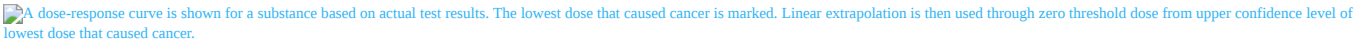

Figure 6.3.2. *The Linearized Multistage Model is used to extrapolate cancer risk from a dose-response curve using the cancer slope factor*
(Image Source: NLM)

EPA uses the **Linearized Multistage Model (LMS)** illustrated in Figure 2 to conduct its cancer risk assessments. It yields a cancer slope factor, known as the **q1*** (pronounced "Q1-star"), which can be used to predict cancer risk at a specific dose. It assumes linear extrapolation with a zero dose threshold from the upper confidence level of the lowest dose that produced cancer in an animal test or in a human epidemiology study.

Other Models

Other models that have been used for cancer assessments include:

- **One-hit model**, which assumes there is a single stage for cancer and that one molecular event induces a cell transformation. This is a very conservative model.
- **Multi-hit model**, which assumes several interactions are needed before a cell can be transformed. This is one of the least conservative models.
- **Probit model**, which assumes log normal distribution (Probit) for tolerances of exposed population. This model is sometimes used, but generally considered inappropriate for assessing cancer risk.
- **Physiologically Based Pharmacokinetic (PBPK) Models**, which incorporate pharmacokinetic and mechanistic data into the extrapolation process. This model requires extensive data and is becoming commonly used.

Application of Models to Estimate Chemical Concentrations in Drinking Water

The chemical chlordane has been found to cause a lifetime risk of one cancer death in a million persons. Different cancer risk assessment models vary in their estimates of drinking water concentrations for chlordane as illustrated in Table 6.3.2:

Model	Concentration (µg/L)
Probit	50
Multi-hit	2
Linearized multistage	0.07
One-hit	0.03

Table 6.3.2. Estimates of drinking water chlordane concentrations by various cancer assessment models

PBPK models are relatively new and are being employed when biological data are available. They quantitate the absorption of a foreign substance, its distribution, metabolism, tissue compartments, and elimination. Some compartments store the chemical (such as bone and adipose tissue) whereas others biotransform or eliminate it (such as liver or kidney). All these biological parameters are used to derive the target dose and comparable human doses.

Noncarcinogenic Risk Assessment

Historically, the **Acceptable Daily Intake (ADI)** procedure has been used to calculate permissible chronic exposure levels for humans based on noncarcinogenic effects. The ADI is the amount of a chemical to which a person can be exposed each day for a long time (usually lifetime) without suffering harmful effects. It is determined by applying safety factors (to account for the uncertainty in the data) to the highest dose in human or animal studies that has been demonstrated not to cause toxicity (NOAEL).

The EPA has slightly modified the ADI approach and calculates a **Reference Dose (RfD)** as the acceptable safety level for chronic noncarcinogenic and developmental effects. Similarly, the ATSDR calculates **Minimal Risk Levels** (MRLs) for noncancer endpoints.

The **critical toxic effect** used in the calculation of an ADI, RfD, or MRL is the serious adverse effect that occurs at the lowest exposure level. It may range from lethality to minor toxic effects. It is assumed that humans are as sensitive as the animal species unless evidence indicates otherwise.

Assessment of Chronic Exposures

In determining the ADIs, RfDs or MRLs, the **NOAEL** is divided by safety factors (uncertainty factors) in order to provide a margin of safety for allowable human exposure.

When a NOAEL is not available, a **LOAEL** can be used to calculate the RfD.

An additional safety factor is included if a LOAEL is used. A Modifying Factor of 0.1–10 allows risk assessors to use scientific judgment in upgrading or downgrading the total uncertainty factor based on the reliability and quality of the data. For example, if a particularly good study is the basis for the risk assessment, a modifying factor of <1 may be used. If a poor study is used, a factor of >1 can be incorporated to compensate for the uncertainty associated with the quality of the study.

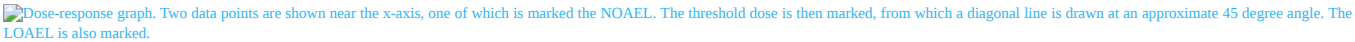

Figure 6.3.3. *Dose-response curve for noncarcinogenic effects*
(Image Source: NLM)

Figure 3 above shows a dose-response curve for noncarcinogenic effects which also identifies the NOAEL and LOAEL. Any toxic effect might be used for the NOAEL/LOAEL so long as it is the most sensitive toxic effect and considered likely to occur in humans.

The **Uncertainty Factors** or **Safety Factors** used to derive an ADI or RfD are listed in Table 6.3.3.

Situation	Uncertainty/Safety Factor
Human variability	10x
Extrapolation from animals to humans	10x

Use of less than chronic data	10x
Use of LOAEL instead of NOAEL	10x
Modifying factor	0.1—10x

Table 6.3.3. Uncertainty/Safety factors used to derive an Acceptable Daily Intake (ADI) or Reference Dose (RfD)

The modifying factor is used only in deriving EPA Reference Doses. The number of factors included in calculating the ADI or RfD depends upon the study used to provide the appropriate NOAEL or LOAEL.

The general formula for deriving the RfD is:

The more uncertain or unreliable the data become, the higher the total uncertainty factor that is applied. An example of an RfD calculation is provided below. A subchronic animal study with a LOAEL of 50 mg/kg/day was used in the numerator. Uncertainty factors used in the denominator are 10 for human variability, 10 for an animal study, 10 for less than chronic exposure, and 10 for use of an LOAEL instead of a NOAEL.

In addition to chronic effects, RfDs can also be derived for other long-term toxic effects, including developmental toxicity.

Traditionally, the **NOAEL method** has been used to determine the **point of departure (POD)** from animal toxicology data for use in risk assessments. However, this approach has limitations such as a strict dependence on the dose selection, dose spacing, and sample size of the study from which the critical effect has been identified. Also, using the NOAEL does not take into consideration the shape of the dose-response curve and other related information.

Benchmark Dose Method

The **benchmark dose (BMD) method**, first proposed as an alternative in the 1980s, addresses many limitations of the NOAEL method. It is less dependent on dose selection and spacing and takes into account the shape of the dose-response curve (Figure 4). In addition, the estimation of a BMD 95% lower bound confidence limit (BMDL) results in a POD that appropriately accounts for study quality (i.e., sample size). With the availability of user-friendly BMD software programs, including the EPA's Benchmark Dose Software (BMDS), the BMD has become the method of choice for many health organizations worldwide.

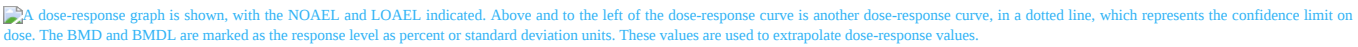

Figure 6.3.4. *Extrapolated values using the benchmark dose method reflect the shape of a dose-response curve (Image Source: EPA)*

Assessment of Noncancer Toxicity Effects

While the Agency for Toxic Substances and Disease Registry (ATSDR) does not conduct cancer risk assessments, it does derive **Minimal Risk Levels (MRLs)** for noncancer toxicity effects (such as birth defects or liver damage). The MRL is defined as an estimate of daily human exposure to a substance that is likely to be without an appreciable risk of adverse effects over a specified duration of exposure. For inhalation or oral routes, MRLs are derived for acute (14 days or less), intermediate (15–364 days), and chronic (365 days or more) durations of exposures.

The method used to derive MRLs is a modification of the EPA's RfD methodology. The primary modification is that the uncertainty factors of 10 may be lower, either 1 or 3, based on scientific judgment. These uncertainty factors are applied for human variability, interspecies variability (extrapolation from animals to humans), and use of a LOAEL instead of NOAEL. As in the case of RfDs, the product of uncertainty factors multiplied together is divided into the NOAEL or LOAEL to derive the MRL.

Assessment of Acute or Short-Term Exposures

Risk assessments are also conducted to derive permissible exposure levels for acute or short-term exposures to chemicals. Health Advisories (HAs) are determined for chemicals in drinking water. HAs are the allowable human exposures for 1 day, 10 days, longer-term, and lifetime durations. The method used to calculate HAs is similar to that for the RfDs using uncertainty factors. Data from toxicity studies with durations of length appropriate to the HA are being developed.

Assessment of Occupational Exposures

For **occupational exposures**, Permissible Exposure Levels (PELs), Threshold Limit Values (TLVs), and National Institute for Occupational Safety and Health (NIOSH) Recommended Exposure Levels (RELs) are developed. They represent dose levels that will not produce adverse health effects from repeated daily exposures in the workplace. The method used to derive is conceptually the same. Safety factors are used to derive the PELs, TLVs, and RELs.

Conversion of Animal Doses to Human Dose Equivalents

Animal doses must be converted to human dose equivalents. The **human dose equivalent** is based on the assumption that different species are equally sensitive to the effects of a substance per unit of body weight or body surface area.

Historically, the FDA used a ratio of body weights of humans to animals to calculate the human dose equivalent. The EPA has used a ratio of surface areas of humans to animals to calculate the human dose equivalent. Some current approaches include multiplying the animal dose by the ratio of human to animal body weight raised to either the 2/3rd or 3/4th power (to convert from body weight to surface area). Toxicologists and risk assessors should check to make sure that the approach they are using is the one mandated or recommended by the regulatory agency of most relevance to their efforts.

Allowable Exposures to Contamination Sources

The last step in risk assessment is to express the risk in terms of allowable exposure to a contaminated source. Risk is expressed in terms of the concentration of the substance in the environment where human contact occurs. For example, the unit for assessing risk in air is risk per -3 g)." tabindex="0">mg/m^3 whereas the unit for assessing risk in drinking water is risk per -3 g)." tabindex="0">mg/L.

For carcinogens, the media risk estimates are calculated by dividing cancer slope factors by 70 kg (average weight of a man) and multiplying by 20 m^3/day (average inhalation rate of an adult) or 2 liters/day (average water consumption rate of an adult).

Knowledge Check

The procedures used to extrapolate from high to low doses primarily depend upon the:

- ◯ Threshold dose of the substance
- ◯ Rate of lethality in laboratory animals
- ◯ Carcinogenicity of the substance

Answer

Genotoxic carcinogenicity of the substance - **This is the correct answer.**
The procedure for extrapolation from high to low doses depend on whether or not the effects are carcinogenic. Carcinogenic effects are not considered to have a threshold dose and mathematical models are used to estimate the risk of carcinogenicity at very low doses. Noncarcinogenic effects are considered to have threshold doses and the margin of safety (MOS) is calculated.

According to EPA, a substance is classified as likely to be carcinogenic to humans when:

- ◯ There is strong evidence of human carcinogenicity
- ◯ Evidence is adequate to demonstrate potential carcinogenicity to humans, but not strongly enough to definitively classify as carcinogenic
- ◯ The weight of evidence suggests human carcinogenicity, but the data are determined not to be sufficient for a stronger conclusion
- ◯ Robust data lead to the conclusion that a substance is clearly carcinogenic to humans

Answer

Evidence is adequate to demonstrate potential carcinogenicity to humans, but not strongly enough to definitively classify as carcinogenic - **This is the correct answer.**
A substance is classified as likely to be carcinogenic to humans when evidence is adequate to demonstrate carcinogenic potential to humans but does not reach the weight of evidence for the descriptor Carcinogenic to Humans.

The primary cancer risk assessment model used by the EPA is known as the:

- ◯ Linearized Multistage Model (LMS)
- ◯ Probit Model
- ◯ Physiologically Based Pharmacokinetic Model (PB-PK)

Answer

Linearized Multistage Model (LMS) - **This is the correct answer.**
EPA uses the Linearized Multistage Model (LMS) to conduct its cancer risk assessments, producing the q1* that is used to predict cancer risk at a specific dose.

The Acceptable Daily Intake (ADI) is calculated by:

- ◯ Dividing the NOAEL by safety factors
- ◯ Dividing the NOAEL by the LOAEL
- ◯ Multiplying the RfD by a modifying factor
- ◯ Linear extrapolation from the LOAEL to the zero intercept

Answer

Dividing the NOAEL by safety factors - **This is the correct answer.**
The ADI is calculated by dividing the NOAEL by safety factors.

Animal doses must be converted to human dose equivalents for risk assessment. When doing this, toxicologists and risk assessors must:

- ◯ Multiply the animal dose by the ratio of human to animal body weight raised to the $^{2}/_{3}$ power
- ◯ Ensure they use the conversion method mandated or recommended by the regulatory agency most relevant to their efforts
- ◯ Multiply the animal dose by the ratio of human to animal body weight raised to the $^{3}/_{4}$ power

Answer

Ensure they use the conversion method mandated or recommended by the regulatory agency most relevant to their efforts - **This is the correct answer.**
Toxicologists and risk assessors should check to ensure they use the approach mandated or recommended by the regulatory agency most relevant to their efforts.

Minimal Risk Levels (MRLs) are derived:

- ◯ Similarly to deriving the RfD, but with a potentially lower uncertainty factor
- ◯ By multiplying the cancer slope factor by the lowest exposure dose
- ◯ By multiplying the LOAEL by safety factors

Answer

Similarly to deriving the RfD, but with a potentially lower uncertainty factor - **This is the correct answer.**
The MRL is calculated much like the RfD, except that the uncertainty factors of 10 may be lower (1 or 3), based on scientific judgment.