SAB Review: IRIS Toxicological Review of Acrylamide

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Presentation Overview

- EPA has updated the previous version of the IRIS Toxicological Review of Acrylamide (1988) based on more recent data, and current guidance and improved methods for deriving toxicity values.
- The current draft of this IRIS Tox Review (12/28/2008) represents the work of many scientist and has undergone numerous internal Agency and Interagency peer review, including reviews by scientist at the USDA, the President's Office of Management and Budget (OMB), and the FDA.
- The charge questions for the Science Advisory Board (SAB) address the main scientific issues identified by the Agency and Interagency reviewers.
- This presentation will provide:
  - A brief overview of acrylamide's potential adverse health effects.
  - The proposed revised reference values compared with the previous values.
  - The issues for SAB consideration.

Background
IRIS Tox. Review of Acrylamide

- EPA's Mission – To protect human health and the environment.
  - Integrated Risk Information System (IRIS) - an electronic database containing information on human health effects that may result from exposure to various substances in the environment.
  - IRIS is prepared and maintained by the EPA's National Center for Environmental Assessment (NCEA) within the Office of Research and Development (ORD).
- The IRIS Tox Review of Acrylamide describes the potential for adverse health effects in humans from exposure to acrylamide, and quantitatively characterizes the dose-response for:
  - Noncancer effects to derive an oral reference dose (RfD) and an inhalation reference concentration (RfC).
  - Cancer effects to derive an oral slope factor and an inhalation unit risk.

Background
Toxicity Values

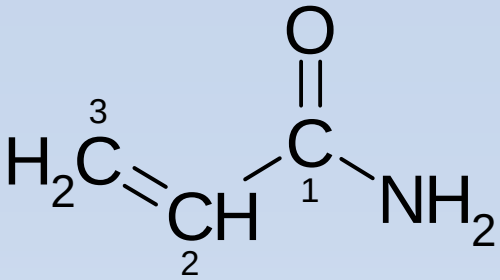
- **RfD** - an estimate (with uncertainty spanning perhaps an order of magnitude) of a daily exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious effects during a lifetime. [mg of substance / kg body weight-day] .

- **RfC** - analogous to the oral RfD but for an estimated continuous inhalation exposure [mg of substance / m^3 air] .

- **Oral Slope Factor** - an upper bound, approximating a 95% confidence limit, on the increased cancer risk from a lifetime exposure to an agent by ingestion [units of proportion of a population (e.g., 1 in a 1,000,000) affected per mg of substance / kg body weight-day].

Background
Toxicity Values (continued)

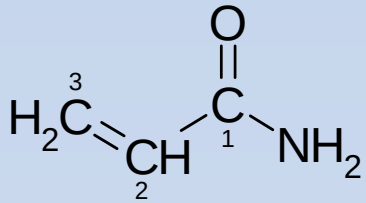
- **Unit Risk** - an upper-bound excess lifetime cancer risk estimated to result from continuous exposure to an agent at a concentration of 1 µg/L in water or 1 µg/m^3 in air.
  - For a substance in drinking water - if unit risk = 2 x 10-6 per µg/L, 2 excess cancer cases (upper bound estimate) are expected to develop per 1,000,000 people if exposed daily for a lifetime to 1 µg of the substance in 1 liter of drinking water.
  - For a substance in air - if unit risk = 2 x 10-6 per µg/m^3, 2 excess cancer cases (upper bound estimate) are expected to develop per 1,000,000 people if exposed daily for a lifetime to 1 µg of the substance in 1 cubic meter of air.



Acrylamide

CAS # 79-06-1

- Acrylamide (AA) has the chemical formula C3H5NO (structural formula CH2=CH-CONH2) and a molecular weight of 71.08.
- It is an odorless, white, crystalline solid.
- AA is a highly water-soluble α,β-unsaturated amide that reacts with nucleophilic sites in macromolecules in Michael-type additions (Calleman, 1996; Segerbäck et al., 1995).
- Monomeric AA readily participates in radical-initiated polymerization reactions, whose products form the basis of most of its industrial applications (Calleman, 1996).



Acrylamide

Characteristics (continued)

- **Molecular weight:** 71.08 (Verschueren, 2001)
- **Chemical Formula:** C3H5NO (Verschueren, 2001)
- **Boiling point:** 192.6°C (Verschueren, 2001)
- **Melting point:** 84.5°C (Verschueren, 2001)
- **Vapor pressure:** 0.007 mm Hg at 25°C (HSDB, 2005)
- **Density:** 1.12 g/mL at 30°C (Budavari, 2001)
- **Vapor density:** 2.46 (air = 1) (Verschueren, 2001)
- **Water solubility:** 2.155 g/mL at 30°C (Verschueren, 2001)
- **Partition coefficient (Kow):** log Kow = –0.67 (octanol/water) (Hansch et al., 1995)
- **Partition coefficient (Koc):** log Koc = 1 (organic carbon/water) (HSDB, 2005)
- **pH:** 5.0–6.5 (50% aqueous solution) (HSDB, 2005)
- **Bioconcentration factor:** 1 for fingerling trout (Petersen et al., 1985)
- **Stability:** Stable at room temperature but may polymerize violently on melting (HSDB, 2005)
- **Conversion factor:** 1 mg/m^3 = 0.34 ppm, 1 ppm = 2.95 mg/m^3

Acrylamide Uses & Environmental Fate

- Mostly used in synthesis of polyacrylamides for use as water-soluble thickeners, in waste water treatment (flocculent), gel electrophoresis (SDS-PAGE), papermaking, ore processing, manufacture of permanent press fabrics; some use in manufacture of dyes or other monomers.
- Release of acrylamide to the environment may occur during production and use, or in the production of polyacrylamide.
- Acrylamide is expected to be highly mobile in water and soils but is not expected to accumulate in the environment due to fairly rapid physical and biological degradation.
- Volatilization of acrylamide from dry or moist soil surfaces is not expected to be an important fate process.
- Vapor-phase acrylamide will be degraded in the atmosphere by reaction with photochemically-produced hydroxyl radicals; the half-life for this reaction in air is estimated to be 1.4 days.

Human Exposure to Acrylamide

- Human exposure to acrylamide had been thought to occur primarily in the workplace from dermal contact and inhalation of dust and vapor, with the general public being potentially exposed to low levels of acrylamide only through contaminated drinking water.
- Public exposure to acrylamide in air has not been an issue to date.
- In early 2002, however, Swedish scientists reported high concentrations of acrylamide in certain fried, baked, and deep-fried foods (Swedish National Food Agency, 2002).
- The Swedish results were reproducible and there was a dramatic increase in interest in non-industrial sources of acrylamide exposure to the general public.
- Subsequent research demonstrated that acrylamide forms de novo during processing of some foods, especially during high temperature cooking of carbohydrate-rich foods that contain asparagine [via a Maillard reaction, a non-enzymatic browning reaction] (Tareke et al., 2000, 2002).

EPA Activity Related to Acrylamide

- EPA regulatory activity - When polyacrylamide is used as a flocculent to remove solids in the purification of drinking water, some residual acrylamide monomer may be present as a contaminant. EPA requires drinking water authorities to certify that the level of acrylamide monomer in the polymer does not exceed 0.05%, and that the application rate for the polymer does not exceed 1 mg/L.
- In 1991, EPA/OPPT proposed a rule to prohibit use of acrylamide and N-methylolacrylamide (NMA) in grouts to protect grouters from neurotoxic and carcinogenic risks from significant dermal and inhalation exposure. The rule was withdrawn in 1992 with the advent of affordable personal protective equipment that adequately protect workers from exposure.
- No other on-going regulatory activities within EPA for acrylamide. EPA is, however, evaluating the potential for ground/drinking water contamination from waste site dumping of industrial coagulated solids from polyacrylamide treated water.

Acrylamide Metabolism

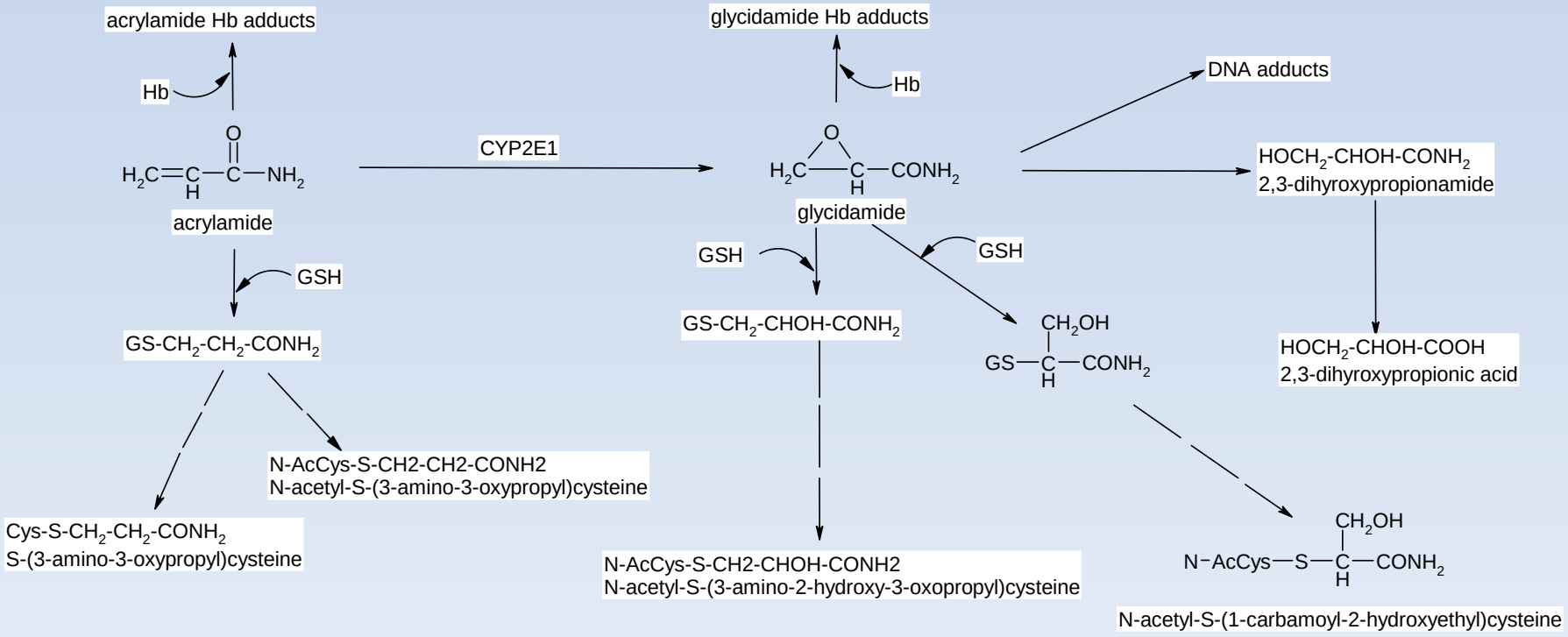


Figure 3-1. Metabolic scheme for acrylamide (AA) and its metabolite glycidamide (GA).
Note: Processes involving several steps are represented with broken arrows. Abbreviations: Hb, hemoglobin; GSH, reduced glutathione; N-AcCys, N-acetylcysteine. Sources: Adapted from Sumner et al. (1999); Calleman (1996); IARC (1994a).

From page 25 of the Draft IRIS Assessment for Acrylamide (12-28-08)
Available at: http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=187729

Hemoglobin Adduct Formation

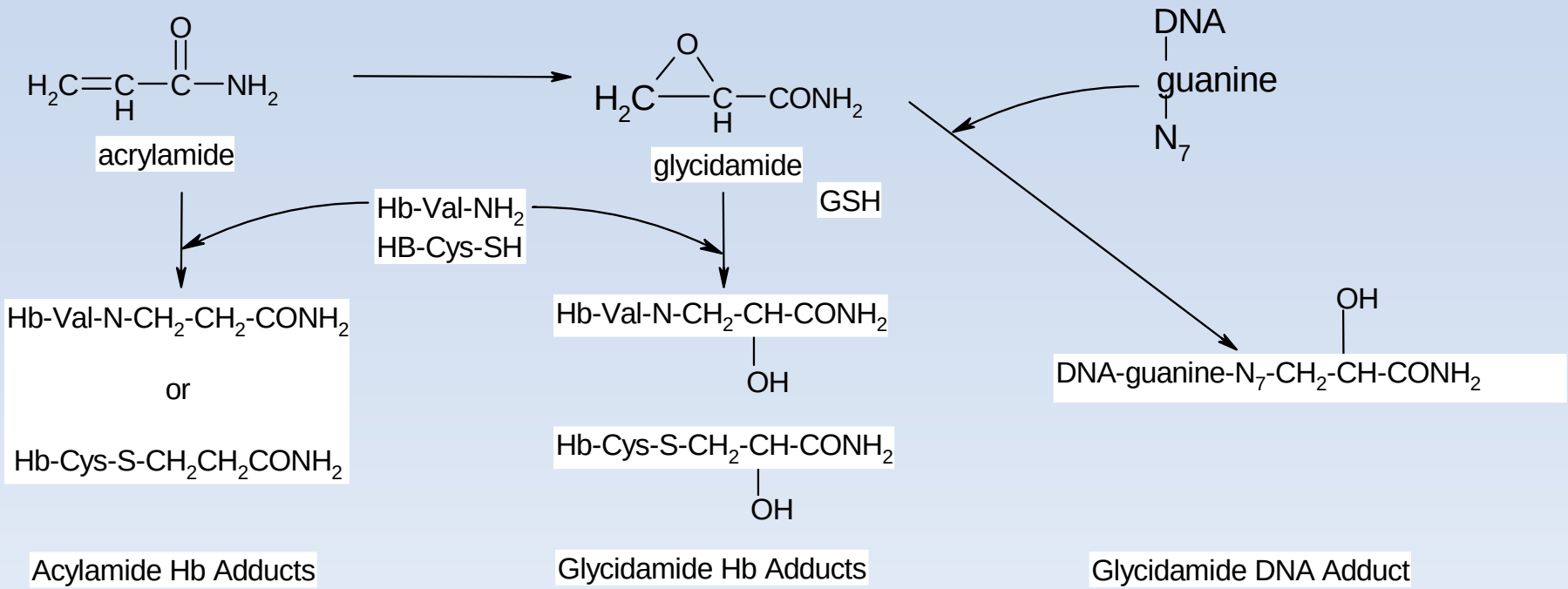


Figure 3-2. Hemoglobin and DNA adducts of acrylamide and glycidamide.
Sources: Dearfield et al. (1995); Bergmark et al. (1993, 1991).

From page 32 of the Draft IRIS Assessment for Acrylamide (12-28-08)
Available at: http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=187729

Potential for Noncancer Adverse Health Effects

(Discussed in detail in Chapters 1-4 and summarized in Chapter 6 of the Draft IRIS Assessment for Acrylamide [12-28-08])

- **Neurological impairment (including peripheral neuropathy).**
  - **Repeatedly observed in human case reports and health surveillance studies, as well as extensive laboratory animal studies.**
  - **Clearly established potential human health hazard.**
- **Impaired male reproductive performance.**
  - **Male-mediated implantation losses observed in laboratory animals orally exposed to AA at doses generally higher than the lowest doses associated with degenerative nerve changes.**
  - **To date, associations between human exposure to AA and reproductive effects have not been reported.**
- **Heritable germ cell effects in mice at relatively high doses.**
  - **Unknown dose-response relationship at low dose exposures.**

Proposed Reference Dose - RfD

Version	Principal study / critical effect	POD (mg/kg bw/day)	UFs	RfD (mg/kg bw/day)
Currently on IRIS (posted 1988)	Burek et al. (1980) Ultrastructural degeneration in the sciatic nerve of male rats (Subchronic drinking water study)	NOEL = 0.2	1000 10 – interspecies 10 – intraspecies 10 – subchr to chronic	2.0×10^{-4}
SAB / Public Review Draft (Dec. 28, 2007)	Johnson et al., 1986 Degenerative lesions in male rat peripheral nerves (Rat chronic drinking water study)	$BMDL_5$ =0.27 $HED_{PBTK\ model}$ = 0.076	30 3 – interspecies (toxicodynamic differences) 10 – intraspecies	3×10^{-3}

Key Science Issues Related to the RfD

- Choice of endpoint and relevancy to humans.
- Selection of a BMR of 5%.
- Use of a PBPK model to estimate the HED.
- Choice of the internal dose metric relative to what is known about the MOA.
- Selection of uncertainty factors.
- Quantitation of heritable germ cell effect.

Proposed Reference Concentration - RfC

Version	Principal study / critical effect	POD (mg/m^3)	UFs	RfC (mg/m^3)
Currently on IRIS	Not derived (lack of data or acceptable route-to-route methodology).			
SAB / Public Review Draft (Dec. 28, 2007)	Johnson et al., 1986 Degenerative lesions in male rat peripheral nerves (Rat chronic drinking water study)	$HEC_{PBPK\ model} = 0.25$ [PBPK model used to estimate the inhalation exposure that would be comparable to the AUC AA in blood as obtained from the oral exposure date, i.e., $BMDL_5$]	30 3 – interspecies (toxicodynamic differences) 10 – intraspecies	0.008

Key Science Issues Related to the RfC

- Choice of endpoint and relevancy to humans.
- Portal of entry effects.
- Use of a PBPK model to conduct the route-to-route extrapolation.
- Choice of the internal dose metric relative to what is known about the MOA.
- Selection of uncertainty factors.

Carcinogenic Potential

- "Likely to be carcinogenic to humans" (*Guidelines for Carcinogen Risk Assessment*, U.S. EPA, 2005) based on:
  - Increased thyroid tumors (males and females), tunica vaginalis mesotheliomas (males), and mammary gland tumors (females) in two chronic drinking water bioassays with F344 rats (Friedman et al., 1995; Johnson et al., 1986).
  - Skin tumor initiation activity in SENCAR and Swiss-ICR mice. given oral, ip, or dermal initiating doses (Bull et al., 1984a, 1984b).
  - Increased lung tumors in A/J mice, ip injection (Bull et al., 1984).
- Limited to inadequate evidence in humans.
  - No significant increased risks for cancer-related deaths in two cohort mortality studies of acrylamide workers, with the exception of an increased risk of pancreatic cancer in a subgroup of workers with the highest cumulative acrylamide exposure in one study (Three cohort mortality studies by Marsh et al., 1999; Collins et al., 1989; Sobel et al., 1986).
  - No significant associations between frequent consumption of foods with high or moderate levels of acrylamide and occurrence of large bowel, kidney, or bladder cancers (Case-control studies by Mucci et al., 2005, 2004, 2003; Pelucchi et al., 2006).
  - One prospective study (Mucci et al., 2006) or occupational exposures from inhalation and/or dermal exposure.

Evidence for a Genotoxic Mode(s) of Carcinogenic Action by Acrylamide

- AA metabolized by CYP2E1 to glycidamide (GA), a DNA-reactive epoxide.
- AA and GA are genotoxic in the Big Blue mouse following oral exposures.
  - Significant increase in lymphocyte Hprt and liver cII mutation frequencies (MFs).
  - AA and GA produced similar mutation spectra that were significantly different from controls consistent with AA exerting its genotoxicity in BB mice via metabolism to GA.
- DNA adducts of GA have been detected in mice and rats exposed to AA and GA in all relevant tissues in both males and females where tumors have been reported.
- GA is mutagenic in short-term bacterial assays.
- GA is mutagenic in male and female mouse somatic cells following oral exposure and in male mouse germ cells (heritable translocations) following intraparenteral exposure.

Evidence for a Genotoxic Mode(s) (continued)

- AA induces heritable translocations in male mouse germ cells following intraparenteral or dermal administration, and specific locus mutations in male germ cells following intraparenteral administration.
- Positive mouse lymphoma assay results
  - Caveat - not definitively known whether these somatic cell mutations resulted from AA-induced chromosomal alterations [chromatid and chromosome breaks and rearrangements] or GA-DNA adducts.
- Dominant lethal mutations demonstrated in rodents following subchronic oral exposure at AA dose levels in the 2.8 to 13.3 mg/kg-day range, which is near the range of chronic dose levels associated with carcinogenic effects in rats (0.5 to 3 mg/kg-day).

Hormonal Disruption as a Carcinogenic MOA for AA - Hypothesized Sequence of Events

- For the induction of tunica vaginalis and mammary gland tumors:
  - Dopamine agonist activities promote age-related hormonal changes that stimulate sustained cell proliferation in the tunica vaginalis and mammary gland, leading to progression to mesothelioma and fibroadenoma, respectively.
- For the thyroid tumors:
  - Alteration of a signal transduction pathway leads to persistent stimulation of cell proliferation in thyroid follicular cells and eventual progression to follicular cell adenomas.
- References: Shipp et al., 2006; Environ, 2002; KS Crump Group, Inc., 1999a,b.

Hormonal Disruption MOA - Evidence

- Induction of tunica vaginalis via dopamine agonist activity (at the D2 dopamine receptor):
  - Data on AA decrease in circulating levels of prolactin in male F344 rats.
  - Leydig cell tumors in rats induced via dopamine agonist activity may not be relevant to humans since human Leydig cells (as well as Leydig cells in other animal species, except male rats) do not decrease their luteinizing hormone (LH) receptors in response to decreased prolactin.
  - Tunica vaginalis mesotheliomas in F344 rats may be linked to the extent of Leydig cell neoplasia, i.e., may not be relevant to humans.
  - Additional supporting evidence would include demonstration of a lack of mesotheliomas in other animal species chronically exposed to AA; however, these data are not currently available.

Hormonal Disruption MOA Evidence (continued)

- Induction of mammary gland tumors via dopamine agonist activity (at the D2 dopamine receptor):
  - In contrast to male rats, there is little empirical evidence to support this alternative MOA in female rats.
  - Marked changes in circulating levels of prolactin have not been observed in female F344 rats exposed to AA for up to 28 days.
  - There is also no direct evidence that AA displays D1 dopamine agonist activity in female rats,
    - i.e., leading to an enhanced ovarian progesterone secretion and subsequent stimulation of cell proliferation in the stromal / fibroblast cells of the rat mammary gland.

Hormonal Disruption MOA Evidence (continued)

- Induction of thyroid tumors via dopamine agonist activity (at the D2 dopamine receptor):
  - Short-term (2–7 days) exposure of female F344 rats to AA caused follicular cell morphometric changes (decreased colloid area and increased cell height) without significantly changing circulating levels of thyroid hormones or thyroid stimulating hormone (TSH).
  - Other studies indicated that AA doses as high as 25 mg/kg-day for up to 28 days did not induce consistent, biologically significant changes in thyroid hormones or TSH levels.
  - Thus, current data do not support a MOA by which AA alters thyroid hormone homeostasis.

Other Alternate MOAs for Carcinogenicity

- Direct evidence that AA may cause follicular cell proliferation by stimulation of a cAMP cascade (without changes in TSH levels) is not currently available.
- TSH-induced mitogenic activities are mediated largely by cAMP, which in turn may activate protein kinase (PKA)-dependent and independent processes.

Cancer Assessment

Version	Cancer Characterization	Cancer Bioassay	Oral Slope factor	Inhalation Unit Risk
Currently on IRIS (posted 1988)	B2; probable human carcinogen	Johnson et al. 1986 (Chronic drinking water study)	Linearized multistage model, extra risk of combined incidence of CNS, mammary and thyroid glands, uterus, and oral cavity tumors in female rats (Johnson et al., 1986; chronic drinking water study) POD [NOEL] = 0.2 mg/kg-day Oral Slope Factor [95% UCLE] = 4.5 $(mg/kg\text{-}day)^{-1}$	Extrapolated from oral data by a method that is no longer valid (i.e., (direct conversion based on oral data; does not account for first pass metabolism) IUR = 1.3×10^{-3} $(\mu g/m^3)^{-1}$

Cancer Assessment (continued)

Version	Cancer Characterization	Cancer Bioassay	Oral Slope factor	Inhalation Unit Risk
SAB / Public Review Draft (Dec. 28, 2007)	Likely to be carcinogenic to humans by all routes of exposure.	Friedman et al. 1995 (Chronic drinking water study)	Linear extrapolation from the $BMDL_{10}$ for combined incidence of F344 male rats with tunica vaginalis mesotheliomas or thyroid tumors (Friedman et al., 1995) Rat $BMDL_{10}$ = 0.27 mg/kg-day POD: PBPK model used to derive an HED [$BMDL_{10}$] = 0.22 mg/kg-day [Based on the internal dose metric of AUC GA] Oral Slope Factor [95% UCLE]* = 0.5 $(mg/kg\text{-}day)^{-1}$	PBPK model estimate of the inhalation exposure needed to produce a comparable AUC GA to that resulting from the oral exposure HED [$BMDL_{10}$] HEC [$BMDL_{10}$]= 0.79 mg/m^3 (Assumes a continuous 24 hour inhalation exposure for a 70 kg person who breathes 20 m3 of air per day) IUR = 1.3×10^{-4} $(\mu g/m^3)^{-1}$

* Slope factor calculated as the upper bound using a summed central estimate and a summed variance. A similar slope factor derived with the alternate method of combining incidence of animals bearing tumors.

Key Science Issues Related to the Cancer Assessment

- Mutagenic (linear) versus hormonal disruption (nonlinear) MOA for tumors in rats; the weight of evidence supports a mutagenic MOA.
- Relevance of rat testicular, mammary, and thyroid tumors to humans.
- Appropriateness of time-to-tumor analysis and method to estimate total risks for tumors from multiple sites.
- Use of a PBPK model to estimate the HED and to conduct a route-to-route extrapolation to derive the IUR.
- Choice of internal dose metric for PBPK model simulations.

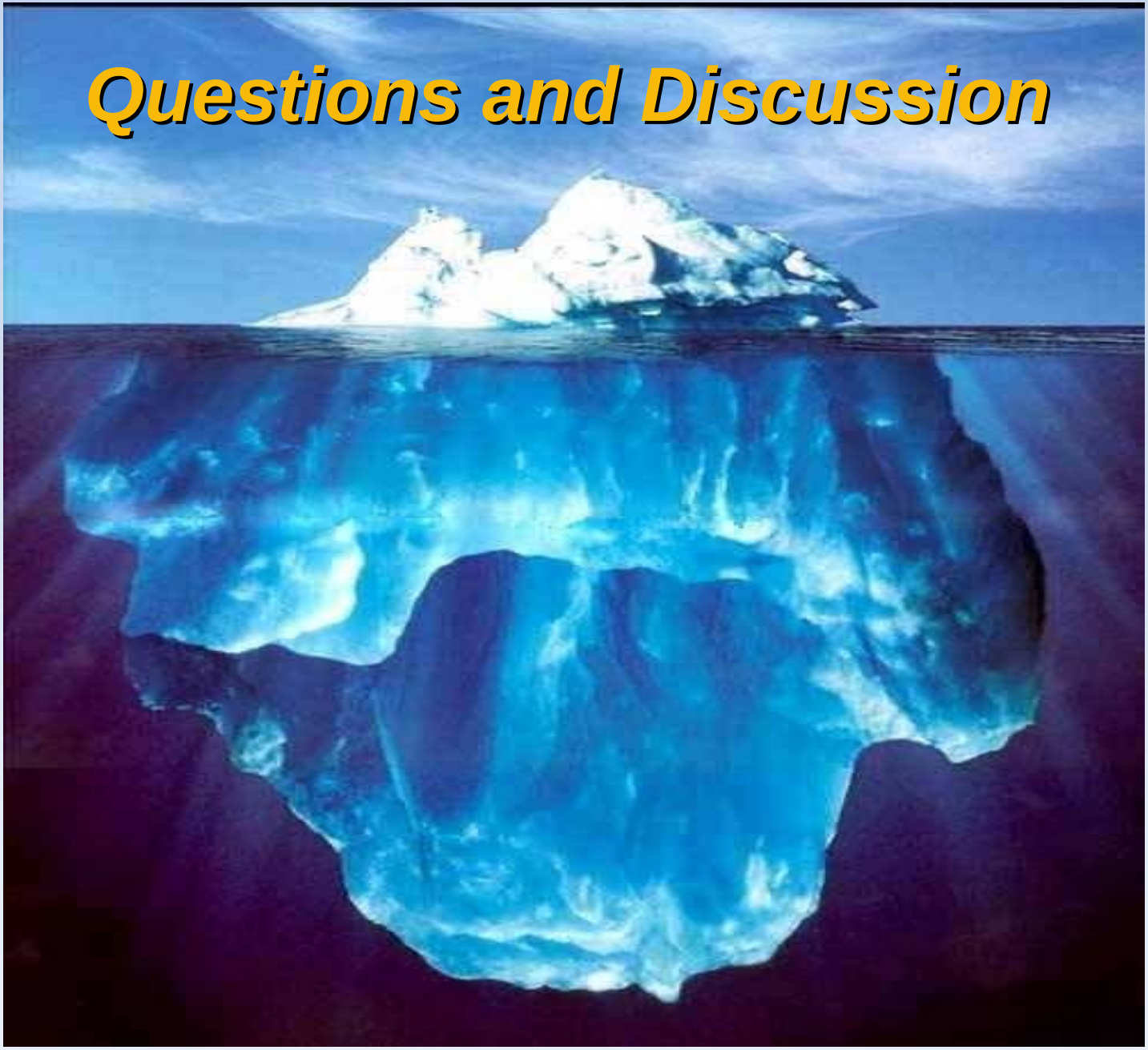
Highlights from the SAB Charge Questions

- Selection of Studies and Derivation of the Reference Dose (RfD).
  - Choice of endpoint, MOA; characterization of the low-dose-response relationship; discussion of heritable germ cell effects; selection of uncertainty factors; benchmark dose methods and choice of response level.
- Use of a PBPK Model in the Derivation of the RfD and the Inhalation Reference Concentration (RfC).
  - Adequacy of model and appropriateness of use; parameter values; supporting data; choice of dose metric; alternate models.

Highlights from the SAB Charge Questions (continued)

- Quantitating Heritable Germ Cell Effects.
  - Accuracy and objectiveness of the discussion; uncertainty in the quantitative characterization of risk; occurrence at low doses; additional data or analyses.
- Carcinogenicity of Acrylamide.
  - Choice of studies, clarity and scientific support for conclusions; support for the proposed MOA as well as alternate MOA(s), adequacy of the WOE discussion; use of the PBPK model and choice of dose metric; characterization of uncertainties.
- Should data be provided to support a Margin of Exposure (MOE) Analysis for various endpoints.

Questions and Discussion



Charge Questions: Selection of Studies and Endpoints for the Oral Reference Dose (RfD)

1. Please comment on the selection of neurotoxicity as the most appropriate choice for the most sensitive endpoint (in contrast to reproductive toxicity, heritable germ cell effects, or other endpoint) based upon the available animal and human data.
2. Please comment on the discussion of mode of action for acrylamide-induced neurotoxicity. Is the discussion clear, transparently and objectively described, and accurately reflective of the current scientific understanding?
3. Please comment on the qualitative discussion of acrylamide's heritable germ cell effects and whether the discussion is clear, transparently and objectively described, and reflective of the current science.

Charge Questions: Derivation of the Reference Dose (RfD)

4. Please comment on whether the selection of the Friedman et al., (1995) and Johnson et al., (1986) studies as co-principal studies has been scientifically justified. Although EPA considers Friedman et al. and Johnson et al. to be co-principal studies, the final quantitative RfD value is derived only from the Johnson study. Please comment on this aspect of EPA's approach. Please also comment on whether this choice is transparently and objectively described in the document. Please identify and provide the rationale for any other studies that should be selected as the principal study(s).

Charge Questions: RfD (continued)

5. Please comment on the benchmark dose methods and the choice of response level used in the derivation of the RfD, and whether this approach is accurately and clearly presented. Do these choices represent the most scientifically justifiable approach for modeling the slope of the dose-response for neurotoxicity? Are there other response levels or methodologies that EPA should consider? Please provide a rationale for alternative approaches that should be considered or preferred to the approach presented in the document.

Charge Questions: RfD (continued)

6. Please comment on the selection of the uncertainty factors (other than the interspecies uncertainty factor) applied to the point of departure (POD) for the derivation of the RfD. For instance, are they scientifically justified and transparently and objectively described in the document? [Note: This question does not apply to the interspecies uncertainty factor which is addressed in the questions on the use of the PBPK model (see PBPK model questions below)]

7. Please provide any other comments on the derivation of the RfD and on the discussion of uncertainties in the RfD.

Charge Questions:
Use of a PBPK Model in Derivation of the RfD and RfC

8. Please comment on whether the documentation for the recalibrated Kirman et al. (2003) PBTK model development, evaluation, and use in the assessment is sufficient to determine if the model was adequately developed and adequate for its intended use in the assessment. Please comment on the use of the PBTK model in the assessment, e.g., are the model structure and parameter estimates scientifically supportable? Is the dose metric of area-under-the-curve (AUC) for acrylamide in the blood the best choice based upon what is known about the mode of action for neurotoxicity and the available kinetic data? Please provide a rationale for alternative approaches that should be considered or preferred to the approach presented in the document.

Charge Questions: PBPK Model in RfD and RfC (continued)

9. Is the Young et al. (2007) PBTK model adequately discussed in the assessment with respect to model structure, parameter values, and data sets used to develop the model? Do you agree with the conclusion (and supporting rationale) that the recalibrated Kirman et al. (2003) model (model structure and parameter values presented in the Toxicological Review) currently represents the best model to use in the derivation of the toxicity values?

Charge Questions: PBPK Model in RfD and RfC (continued)

10. According to US EPA's RfC Methodology (1994), the use of PBTK models is assumed to account for uncertainty associated with the toxicokinetic component of the interspecies uncertainty factor across routes of administration. Does the use of the PBTK model for acrylamide objectively predict internal dose differences between the F344 rat and humans, is the use of the model scientifically justified, and does the use of the PBTK reduce the overall uncertainty in this estimate compared to the use of the default factor? Are there sufficient scientific data and support for use of this PBTK model to estimate interspecies toxicokinetic differences and to replace the default interspecies factor for toxicokinetic differences (i.e., 101/2)?

Charge Questions: PBPK Model in RfD and RfC (continued)

10 (continued). Is the remaining uncertainty factor for toxicodynamic differences scientifically justified, appropriate and correctly used?

11. Please comment on whether the PBTK model is adequate for use to conduct a route-to-route extrapolation for acrylamide to derive an RfC in the absence of adequate inhalation animal or human dose-response data to derive the RfC directly. Was the extrapolation correctly performed and sufficiently well documented?

12. Please provide any other comments on the derivation of the RfC and on the discussion of uncertainties in the RfC.

Charge Questions: Margin of Exposure (MOE) Analysis

13. Would you suggest that EPA include a Table that lists points of departure (e.g., NOAELs, BMDs, etc.) for various endpoints that could be used, in conjunction with exposure assessments, to conduct a MOE analysis?

Charge Questions: Quantitating Heritable Germ Cell Effects

14. Please comment on the discussion of methods to quantitate the dose-response for heritable germ cell effects as to whether it is appropriate, clear and objective, and reflective of the current science. Has the uncertainty in the quantitative characterization of the heritable germ cell effects been accurately and objectively described?

15. Please comment on the scientific support for the hypothesis that heritable germ cell effects are likely to occur at doses lower than those seen for neurotoxicity? What on-going or future research might help resolve this issue?

Charge Questions: Quantitating Heritable Germ Cell Effects (continued)

16. The risks of heritable germ cell effects (i.e., number of induced genetic diseases per million offspring) for some estimated exposure in workers and the population are presented in Table 5-11, and are based on the quantitative methods and parameter estimates discussed in Section 5.4 of the Toxicological Review. Please comment on whether or not the quantitation of heritable germ effects should be conducted, the level of uncertainty in the results, if Table 5-11 is useful for risk assessment purposes, and if the RfD should be included in the Table as one of the exposure levels.

Charge Questions: Quantitating Heritable Germ Cell Effects (continued)

17. Do you know of any additional data or analyses that would improve the quantitative characterization of the dose-response for acrylamide-induced heritable germ cell effects? Would these data also support the quantitative characterization of "total" male-mediated reproduction risks to offspring (i.e., lethality + heritable defect)? If data are not available, do you have any recommendations for specific needed studies?

Charge Questions: Carcinogenicity of Acrylamide

18. Have the rationale and justification for the cancer designation for acrylamide been clearly described? Is the conclusion that acrylamide is a likely human carcinogen scientifically supportable?

19. Do you agree that weight of the available evidence supports a mutagenic mode of carcinogenic action, primarily for the acrylamide epoxide metabolite, glycidamide (GA)? Has the rationale for this MOA been clearly and objectively presented, and is it reflective of the current science?

Charge Questions: Carcinogenicity (continued)

20. Are there other MOAs that should be considered? Is there significant biological support for alternative MOAs for tumor formation, or for alternative MOAs to be considered to occur in conjunction with a mutagenic MOA? Please specifically comment on the support for hormonal pathway disruption. Are data available on alternate MOAs sufficient to quantitate a dose-response relationship?

Charge Questions: Carcinogenicity (continued)

21. Two chronic drinking water exposure bioassays in Fischer 344 rats (Friedman et al., 1995; Johnson et al., 1986) were used to derive the oral slope factor, and to identify the tumors of interest for the MOA discussion. Are the choices for the studies, tumors, and methods to quantify risk transparent, objective, and reflective of the current science? Do you have any suggestions that would improve the presentation or further reduce the uncertainty in the derived values?

22. The cancer slope factor (CSF) derivation includes an adjustment for early mortality (i.e., time-to-tumor analysis). Is this adjustment scientifically supported in estimating the risk from the 2-year bioassay data for increased incidence of tumors in the rats?

Charge Questions: Carcinogenicity (continued)

23. The dose metric used in the PBTK model analysis to derive the human equivalent concentration was area under the curve (AUC) in the blood for the putative genotoxic metabolite, glycidamide. Please comment on whether AUC for glycidamide is the best choice of the dose metric in estimating the human equivalent concentration to derive the oral slope factor. If other dose metrics are preferable, please provide the scientific rationale for their selection.

Charge Questions: Carcinogenicity (continued)

24. As with the RfC, there were insufficient cancer inhalation data to derive an inhalation unit risk (IUR). The PBTK model was used in a route-to-route extrapolation of the dose-response relationship from the oral data, and to estimate the human equivalent concentration for inhalation exposure to acrylamide. Please comment on whether this extrapolation to derive the inhalation unit risk was correctly performed and sufficiently well documented.

Charge Questions: Carcinogenicity (continued)

25. The recommendation to use the age-dependent adjustment factors (ADAFs) is based on the determination of a mutagenic MOA for carcinogenicity. Is this recommendation scientifically justifiable and transparently and objectively described

26. Please provide any other comments on the CSF or IUR, and on the discussion of uncertainties in the cancer assessment.