Evaluation of Draft Chloroform HID

Susan A. Rice, Ph.D., D.A.B.T.

DART Committee Meeting
November 4, 2004

Criteria in Evaluation of Causal Association

- Strength of the association
- Dose-response relationship
- Specificity of the association
- Appropriate temporal association
- Consistency across multiple studies
- Biologic plausibility
- Coherence of the evidence

Epidemiologic Studies
No Evidence of DART

- The strength and direction of associations of chloroform and DART outcomes are not consistent across studies
- A dose-response for chloroform is not present or is very weak
- Misclassification of subjects in exposure categories is a major weakness

Exposure Misclassification

- Chloroform concentrations in home drinking water were not directly measured in the majority of studies
- Exposure categories
  - Water source (ground & water)
  - Concentration at the distribution source
  - At best, a metric of concentration × reported home tap water consumption
- Contributions from other sources not evaluated

Additional Considerations

- Misclassification of gestational age significantly affects weight-related outcomes
- Confounding factors were not consistently controlled across studies
- Total trihalomethanes (TTHM) are an inappropriate surrogate for chloroform
- No causal link (U.S. EPA 2001)

Animal Studies

- The majority of animal studies showed no effects of chloroform on development or reproduction
- Dose-response not present when statistically significant results reported
- Lack of consistency among outcomes between studies
- Maternal toxicity can explain effects

Animal & *In Vitro* Studies

- Pilot & range-finding studies are inappropriate of scientific evaluation
- Abstracts are inappropriate for scientific evaluation
- *In vitro* studies are irrelevant
  - Chloroform concentrations would be lethal to humans

Effects on Sperm
Chang et al. (2001)

- Laboratory worker
- Estimated chloroform exposure
  - As high as 450 ppm
  - As long as 2 hr/day, 5.5 days/wk, 8 months
- Other solvent exposure
  - Isooctane, tetrahydrofuran, & others
- Reduced sperm motility
- Morphology not evaluated

Effects on Sperm - Land, 1981

- (C57B1/C3H)F1 mouse
- 400 & 800 ppm
- 4 hr/day, 5 days in early spermatogenesis
- 10% mortality in each group
- Abnormal spermatozoa
  - Control - 1.42 ± 0.08% (SE)
  - 400 ppm - 2.74 ± 0.31% (SE)
  - 800 ppm - 3.48 ± 0.66 (SE)
- Relevance to fertility?

Male Reproduction - Animals

- *Chapin et al. (1997) and NTP (1988)*
  - CD-1 mice, NTP continuous breeding protocol
  - 6.6, 15.9, and 41.2 mg/kg
    - 7 days prior to mating
    - During a 98-day co-habitation period
  - Normal
    - Sperm motility,
    - Sperm density
    - % abnormal sperm

Male Reproduction - Animals

- *U.S. EPA (1980)*
  - Osborne-Mendel rats
  - 90-day subacute toxicity study
  - 20, 38, 57, 81, and 160 mg/kg-day orally
  - No effects (as part of a complete necropsy)
    - Testes,
    - Prostate
    - Seminal vesicles

Male Reproduction - Animals

- *Heywood et al. (1979)*
  - Beagle dogs
  - 15 or 30 mg/kg-day orally
  - 6 days/week for 7.5 years
  - 20-24 week recovery period
  - No effect on weight
    - Testes
    - Prostate

Human - Normal Sperm

- Findings predictive of fertility
  - Sperm count (concentration): >48 million/ml
  - Initial sperm motility: >63%
  - Normal sperm morphology: >12%
- Findings suggestive of infertility
  - Sperm count (concentration): <13.5 million/ml
  - Initial sperm motility: <32%
  - Normal sperm morphology: <9%

    - Reference: Guzik (2001), NEJM 345:1388-93

Conclusions

- No evidence for
  - Developmental effects
  - Male or female reproductive effects
- Epi studies – no causal association
- Case report – uncontrolled
- Animal studies
  - DART only observed at chloroform exposures that produce significant maternal or paternal toxicity

Conclusion

- There is insufficient evidence to classify chloroform as a developmental or reproductive toxicant
- Proposition 65 standard for listing states that any agent to be listed must be
  - *"clearly shown through scientifically valid testing according to generally accepted principles to cause . . . reproductive toxicity"*