

3. World Health Organization Review of the Reproductive and Developmental Toxicology Literature (2000)

The International Programme on Chemical Safety (IPCS) published an evaluation of Disinfectants and DBPs in its *Environmental Health Criteria* monograph series (WHO 2000). In this review of the toxicology data on reproductive and developmental effects from DBP exposure, the World Health Organization (WHO) concludes that although the data on these effects are not as robust as the cancer database, these effects are of potential health concern. The WHO concludes that reproductive effects in females have been principally embryoletality and fetal resorptions associated with the haloacetonitriles (trichloroacetonitrile, dichloroacetonitrile, bromochloroacetonitrile, and dibromoacetonitrile) and the dihaloacetates, while DCAA and DBAA have both been associated with adverse effects on male reproduction.

4. New Studies

Christian *et al.* (2001) conducted a developmental toxicity study with pregnant New Zealand White rabbits exposed to BDCM in drinking water at concentrations of 0, 15, 150, 450, and 900 ppm in drinking water on gestation days 6–29. The no observed adverse effect level (NOAEL) and lowest observed adverse effect level (LOAEL) identified for maternal toxicity in this study were 13.4 mg/kg-day (150 ppm) and 35.6 mg/kg-day (450 ppm), respectively, based on decreased body weight gain. The developmental NOAEL was 55.3 mg/kg-day (900 ppm) based on absence of statistically significant, dose-related effects at any tested concentration. Christian *et al.* (2001) also conducted a developmental study of BDCM in a second species, Sprague-Dawley rats. Rats were exposed to BDCM in the drinking water at concentrations of 0, 50, 150, 450, and 900 ppm on gestation days 6 to 21. The concentration-based maternal NOAEL and LOAEL for this study were 150 ppm and 450 ppm, respectively, based on statistically significant, persistent reductions in maternal body weight and body weight gains. Based on the mean consumed dosage of bromodichloromethane, these concentrations correspond to doses of 18.4 mg/kg-day and 45.0 mg/kg-day, respectively. The concentration-based developmental NOAEL and LOAEL were 450 ppm and 900 ppm, respectively, based on a significantly decreased number of ossification sites per fetus for the forelimb phalanges

(bones of the hand or the foot) and the hindlimb metatarsals and phalanges. These concentrations correspond to mean consumed doses of 45.0 mg/kg-day and 82.0 mg/kg-day, respectively.

Christian *et al.* (2002b) summarized the results of a two-generation reproductive toxicity study on bromodichloromethane conducted in Sprague-Dawley rats. Bromodichloromethane was continuously provided to test animals in the drinking water at concentrations of 0, 50, 150, or 450 ppm. Average daily doses estimated for the 50, 150, and 450 ppm concentrations were reportedly 4.1 to 12.6, 11.6 to 40.2, and 29.5 to 109 mg/kg-day, respectively. The parental NOAEL and LOAEL were 50 and 150 ppm, respectively, based on statistically significant reduced body weight and body weight gain; F1 and F2 generation pup body weights were reduced in the 150 and 450 ppm groups during the lactation period after the pups began to drink the water provided to the dams. Body weight and body weight gain were also reduced in the 150 and 450 ppm F1 generation males and females. A marginal effect on estrous cyclicity was observed in F1 females in the 450 ppm exposure group. Small ($\leq 6\%$), but statistically significant, delays in F1 generation sexual maturation occurred at 150 (males) and 450 ppm (males and females) as determined by timing of vaginal patency or preputial separation. The study's authors considered these effects to be a secondary response associated with reduced body weight, which appears to be dehydration brought about by taste aversion to the compound. The results of this study identify NOAEL and LOAEL values for reproductive effects of 50 ppm (4.1 to 12.6 mg/kg-day) and 150 ppm (11.6 to 40.2 mg/kg-day), respectively, based on delayed sexual maturation.

Bielmeier *et al.* (2001) conducted a series of experiments to investigate the mode of action in bromodichloromethane-induced full litter resorption (FLR). The study included a strain comparison of F344 and Sprague-Dawley (SD) rats. In the strain comparison experiment, female SD rats (13 to 14/dose group) were dosed with 0, 75, or 100 mg/kg-day by aqueous gavage in 10% Emulphor® on GD 6 to 10. F344 rats (12 to 14/dose group) were dosed with 0 or 75 mg/kg-day administered in the same vehicle. The incidence of FLR in the bromodichloromethane-treated F344 rats was 62%, while the incidence of FLR in SD rats treated with 75 or 100 mg/kg-day of bromodichloromethane was 0%. Both strains of rats showed similar signs of maternal toxicity, and

the percent body weight loss after the first day of dosing was comparable for SD rats and the F344 rats that resorbed their litters. The rats were allowed to deliver and pups were examined on postnatal days 1 and 6. Surviving litters appeared normal and no effect on postnatal survival, litter size, or pup weight was observed. The series of experiments conducted by Bielmeier *et al.* (2001) identified a LOAEL of 75 mg/kg-day (the lowest dose tested) based on FLR in F344 rats. A NOAEL was not identified. Mechanistic studies indicate that BDCM-induced pregnancy loss is likely to be luteinizing hormone (LH)-mediated (Bielmeier *et al.*, 2001). It is possible that BDCM alters LH levels by disrupting the hypothalamic-pituitary-gonadal axis or by altering the responsiveness of the corpora lutea to LH. Since these possible mechanisms are potentially relevant to pregnancy maintenance in humans, EPA believes the finding of BDCM-induced pregnancy loss in F344 rats is relevant to risk assessment, and may provide insight into the epidemiological finding of increased risk of spontaneous abortion associated with consumption of BDCM (Waller *et al.* 1998, 2001).

Christian *et al.* (2002a) recently completed a two-generation drinking water study of DBA in rats. Male and female Sprague-Dawley rats (30/sex/exposure group) were administered DBA in drinking water at concentrations of 0, 50, 250, or 650 ppm continuously from initiation of exposure of the parental (P) generation male and female rats through weaning of the F₂ offspring. Based on testicular histomorphology indicative of abnormal spermatogenesis in P and F₁ males, the parental and reproductive/developmental toxicity LOAEL and NOAEL are 250 and 50 ppm, respectively.

Previous studies by EPA have reported adverse effects of DBA, administered via oral gavage, on spermatogenesis that impacted male fertility (Linder *et al.* 1994a, 1995, 1997a) at doses-comparable to those achieved in the Christian *et al.* (2002a) study. Based on these studies collectively, it is clear that DBA is spermatotoxic. Moreover, Veeramachaneni *et al.* (2000) reported in an abstract that sperm from male rabbits exposed to DBA *in utero* from gestation days 15 and throughout life reduced the fertility of artificially inseminated females as evidenced by reduced conceptions. When published, this study may support the evidence that DBA is a male reproductive system toxicant.

In addition, research on DBA by Klinefelter *et al.* (2001) has