

B. Toxicological Profile

1. *Acute toxicity.* Dichlormid has low acute toxicity as indicated by a range of studies including: A rat acute oral study with a lethal dose (LD)₅₀ of 2,816 milligrams/kilogram (mg/kg) for males and 2,146 mg/kg for females, respectively; a rat acute dermal study with an LD₅₀ of >2,040 mg/kg, and a rabbit acute dermal study with an LD₅₀ of >5,000 mg/kg; a rat inhalation study with an LD₅₀ of >5.5 milligrams/liter (mg/L); a primary eye irritation study in the rabbit showing mild ocular irritation; a primary dermal irritation study in the rabbit showing severe skin irritation; and a skin sensitization study which showed that dichlormid was a mild skin sensitizer in the guinea pig.

2. *Genotoxicity.* Dichlormid was not mutagenic in a range of *in vitro* assays, including the Salmonella/microsome (Ames) assay, the human lymphocyte cytogenetic assay (both assays with and without metabolic activation), and an unscheduled DNA synthesis (DNA repair) assay in hepatocytes. In the L5178Y mouse lymphoma assay, small increases in mutant frequency were observed only at cytotoxic concentrations, and were not considered to be significant. *In vivo*, dichlormid was negative in the mouse micronucleus test and in the rat unscheduled DNA synthesis assay, when tested at the maximum tolerated dose.

3. *Reproductive and developmental toxicity.* In a developmental toxicity study, rats were dosed orally by gavage with 0, 10, 40, or 160 mg/kg/day. The no observed adverse effect level (NOAEL) for maternal toxicity was 10 mg/kg/day based on a reduction in body weight gain and food consumption at 40 and 160 mg/kg/day. The developmental NOAEL was determined to be 40 mg/kg/day based on marginal fetotoxic effects, including extra 14th ribs probably due to maternal stress, slight sternbra misalignment and some centra unossified, at 160 mg/kg/day.

In a developmental toxicity study, rabbits were dosed orally by gavage with 0, 5, 30, or 180 mg/kg/day. The lowest observed effect level (LOAEL) for both maternal and fetotoxicity was 180 mg/kg/day characterized by reduced body weight gain and food consumption, and a small increase in post implantation loss, an increased number of early resorptions, a decreased number of fetuses per litter and evidence of fetotoxicity (partial ossification and misshapen/fused sternbrae). The NOAEL for both maternal and developmental toxicity was 30 mg/kg/day.

In a two-generation reproduction study in rats fed diets of 0, 15, 75, and 500 ppm of dichlormid, dietary administration of 500 ppm dichlormid (48.5 mg/kg/day) for two successive generations resulted in decreased body weights and increased liver weights in parents and pups of both generations. There were no effects on reproductive performance or reproductive organs at dose levels up to and including 500 ppm dichlormid. There were no toxicologically significant effects in parents or offspring at a dose level of 75 ppm dichlormid (>7.4 mg/kg/day).

4. *Subchronic toxicity.* In a subchronic toxicity study, groups of 12 male and 12 female Wistar-derived alpk:ApfSD rats were fed diets containing 0, 20, 200, or 2,000 ppm dichlormid for 90 days. Significant reductions in body weight gain and food consumption were seen in male and female rats receiving 2,000 ppm dichlormid, and to a lesser degree, in females at 200 ppm. The liver was identified as the principal target organ (enlargement increased APDM activity in females, centrilobular hypertrophy, increased bile duct pigmentation) in the 2,000 ppm group. The NOAEL was 20 ppm (equivalent to approximately 1.8 mg/kg/day), and the LOAEL was 200 ppm based on reduced body weight gain and food consumption, and a marginal increase in APDM activity in females and liver enlargement in males.

In a 90-day dog feeding study, previously submitted and reviewed by EPA, animals were dosed (4 dogs/sex/dose) at 0, 1, 5, 25, and 50 mg/kg/day. The NOAEL was 5 mg/kg/day, and the LOAEL 25 mg/kg/day based on reduced body weight gain, increased liver weight and degenerative changes involuntary muscle with an associated increase in plasma creatine kinase and alkaline phosphatase activity between 6 and 10 weeks.

In a 14-week rat inhalation study, groups of 18 male and 18 female Sprague-Dawley CD rats were subjected to a whole body exposure of 0, 2.0, 19.9, or 192.5 mg/m³ for 6 hours per day, 5 days per week. The NOAEL was 2.0 mg/m³ based on histopathologic tissue alterations to the nasal olfactory epithelium at 19.9 and 192.5 mg/m³, suggesting that dichlormid was a mild irritant to the nasal cavity. An increase in relative liver, kidney and lung weights at 19.9 and 192.5 mg/m³ was not supported by gross or histopathological observations.

5. *Chronic toxicity.* Rats (64/sex/group) were fed diets containing 0, 20, 100, or 500 ppm dichlormid (0, 1.3, 6.5, 32.8 mg/kg/day for males and 0, 1.5, 7.5, 37.1 mg/kg/day for females) for up to 2

years. At 500 ppm in both males and females, there were treatment-related effects on growth and food consumption, minor reductions in plasma triglycerides, and in males, increased liver weights accompanied by hepatocyte vacuolation and pigmentation effects. In females, there was a slight overall increase in malignant tumors, primarily uterine adenocarcinomas, at 500 ppm, but this specific increase was within the spontaneous incidence observed in historical data. It was concluded that there was no evidence of oncogenicity associated with dichlormid treatment. The NOAEL for chronic toxicity was 100 ppm (6.5 and 7.5 mg/kg/day for males and females, respectively).

In an 18-month oncogenicity study, mice (55/sex/group) were fed dichlormid at doses of 0, 10, 50, or 500 ppm (0, 1.4, 7.0, 70.7 mg/kg for males and 0, 1.84, 9.2, 92.4 mg/kg for females). At 500 ppm, there was a slight increase in mortality for females from week 64 onward, and body weights and food utilization were reduced in males, and to a lesser extent, in females. Also, mice fed 500 ppm dichlormid showed non-neoplastic changes which were minor and consisted of changes in severity or incidence of common spontaneous findings. Based on these effects, the chronic NOAEL was 50 ppm (7.0 and 9.2 mg/kg/day for males and females, respectively). There was a marginal increase in Harderian gland adenomas in males at 500 ppm, but this was considered to reflect the variable spontaneous tumor rate seen in this strain and sex of mouse. It was concluded there was no evidence of oncogenicity associated with dichlormid treatment.

Based on available chronic toxicity data, the reference dose (RfD) for dichlormid is 0.07 mg/kg/day. This RfD is based on the 2-year feeding study in rats with a NOAEL of 7 mg/kg/day. An uncertainty factor of 100 was used to account for interspecies extrapolation and intraspecies variability. The 2-year rat study is consistent with, but supersedes the 90-day rat study. The 2-year rat NOAEL of 7 mg/kg/day lies between 1.8 and 18 mg/kg/day derived from the NOAEL and LOAEL figures of 20 and 200 ppm, respectively, for the most recent 90-day rat study. Thus, the overall NOAEL in the rat for both chronic and subchronic exposure should be regarded as 7 mg/kg/day. Based on the proposed Guidelines for Carcinogenic Risk Assessment (July 1999), dichlormid is not likely to be a human carcinogen, and a margin of exposure (MOE) approach should be used for human risk assessment.