Ingredient name: Crotonaldehyde

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Provisional Peer-Reviewed Toxicity Values for

Crotonaldehyde
(CASRN 123-73-9)

Superfund Health Risk Technical Support Center
National Center for Environmental Assessment
Office of Research and Development
U.S. Environmental Protection Agency
Cincinnati, OH 45268

Commonly Used Abbreviations

BMC	Benchmark Concentration
BMD	Benchmark Dose
BMCL	Benchmark Concentration Lower bound 95% confidence interval
BMDL	Benchmark Dose Lower bound 95% confidence interval
HEC	Human Equivalent Concentration
HED	Human Equivalent Dose
IRIS	Integrated Risk Information System
IUR	inhalation unit risk
LOAEL	lowest-observed-adverse-effect level
$LOAEL_{ADJ}$	LOAEL adjusted to continuous exposure duration
$LOAEL_{HEC}$	LOAEL adjusted for dosimetric differences across species to a human
NOAEL	no-observed-adverse-effect level
$NOAEL_{ADJ}$	NOAEL adjusted to continuous exposure duration
$NOAEL_{HEC}$	NOAEL adjusted for dosimetric differences across species to a human
NOEL	no-observed-effect level
OSF	oral slope factor
p-IUR	provisional inhalation unit risk
p-OSF	provisional oral slope factor
p-RfC	provisional reference concentration (inhalation)
p-RfD	provisional reference dose (oral)
POD	point of departure (oral)
RfC	reference concentration (inhalation)
RfD	reference dose
UF	uncertainty factor
UF_A	animal to human uncertainty factor
UF_C	composite uncertainty factor
UF_D	incomplete to complete database uncertainty factor
UF_H	interhuman uncertainty factor
UF_L	LOAEL to NOAEL uncertainty factor
UF_S	subchronic to chronic uncertainty factor
WOE	weight of evidence

PROVISIONAL PEER-REVIEWED TOXICITY VALUES FOR CROTONALDEHYDE (CASRN 123-73-9)

Background

On December 5, 2003, the U.S. Environmental Protection Agency's (EPA) Office of Superfund Remediation and Technology Innovation (OSRTI) revised its hierarchy of human health toxicity values for Superfund risk assessments, establishing the following three tiers as the new hierarchy:

1) EPA's Integrated Risk Information System (IRIS).
2) Provisional Peer-Reviewed Toxicity Values (PPRTVs) used in EPA's Superfund Program.
3) Other (peer-reviewed) toxicity values, including
   - Minimal Risk Levels produced by the Agency for Toxic Substances and Disease Registry (ATSDR),
   - California Environmental Protection Agency (CalEPA) values, and
   - EPA Health Effects Assessment Summary Table (HEAST) values.

A PPRTV is defined as a toxicity value derived for use in the Superfund Program when such a value is not available in EPA's IRIS. PPRTVs are developed according to a Standard Operating Procedure (SOP) and are derived after a review of the relevant scientific literature using the same methods, sources of data, and Agency guidance for value derivation generally used by the EPA IRIS Program. All provisional toxicity values receive internal review by two EPA scientists and external peer review by three independently selected scientific experts. PPRTVs differ from IRIS values in that PPRTVs do not receive the multiprogram consensus review provided for IRIS values. This is because IRIS values are generally intended to be used in all EPA programs, while PPRTVs are developed specifically for the Superfund Program.

Because new information becomes available and scientific methods improve over time, PPRTVs are reviewed on a 5-year basis and updated into the active database. Once an IRIS value for a specific chemical becomes available for Agency review, the analogous PPRTV for that same chemical is retired. It should also be noted that some PPRTV documents conclude that a PPRTV cannot be derived based on inadequate data.

Disclaimers

Users of this document should first check to see if any IRIS values exist for the chemical of concern before proceeding to use a PPRTV. If no IRIS value is available, staff in the regional Superfund and Resource Conservation and Recovery Act (RCRA) program offices are advised to carefully review the information provided in this document to ensure that the PPRTVs used are appropriate for the types of exposures and circumstances at the Superfund site or RCRA facility in question. PPRTVs are periodically updated; therefore, users should ensure that the values contained in the PPRTV are current at the time of use.

It is important to remember that a provisional value alone tells very little about the adverse effects of a chemical or the quality of evidence on which the value is based. Therefore,

users are strongly encouraged to read the entire PPRTV document and understand the strengths and limitations of the derived provisional values. PPRTVs are developed by the EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center for OSRTI. Other EPA programs or external parties who may choose of their own initiative to use these PPRTVs are advised that Superfund resources will not generally be used to respond to challenges of PPRTVs used in a context outside of the Superfund Program.

Questions Regarding PPRTVs

Questions regarding the contents of the PPRTVs and their appropriate use (e.g., on chemicals not covered, or whether chemicals have pending IRIS toxicity values) may be directed to the EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center (513-569-7300), or OSRTI.

INTRODUCTION

There is no RfD or RfC assessment for crotonaldehyde (2-butenal; see Figure 1 for chemical structure) on IRIS (U.S. EPA, 2008), the Drinking Water Health Advisories list (U.S. EPA, 2006), or the HEAST (U.S. EPA, 1997). The Chemical Assessments and Related Activities (CARA) list (U.S. EPA, 1991, 1994) includes a Health and Environmental Effects Document (HEED) for crotonaldehyde (U.S. EPA, 1989). The HEED (U.S. EPA, 1989) concluded that a chronic or subchronic RfD could not be derived from the available animal data. Crotonaldehyde has not been the subject of a toxicological profile by ATSDR (2008). A World Health Organization (WHO, 2008) Concise International Chemical Assessment Document (CICAD) declined to derive toxicity values due to the lack of suitable data. The American Conference of Governmental Industrial Hygienists (ACGIH, 2008) has adopted a threshold limit value-short-term exposure limit (TLV-STEL) (ceiling limit) of 0.3 ppm for crotonaldehyde based on the potential for irritation to the eyes and upper respiratory tract (quantitation based on analogy to formaldehyde). The time-weighted average-recommended exposure limit (TWA-REL) established by the National Institute for Occupational Safety and Health (NIOSH, 2008), and the current enforceable Occupational Safety and Health Administration (OSHA, 2008) permissible exposure limit (PEL), is 2 ppm (6 mg/m^3).

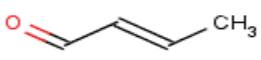

Figure 1. Chemical Structure of Crotonaldehyde

IRIS (U.S. EPA, 2008) includes a Group C, *Possible Human Carcinogen*, cancer weight-of-evidence designation for crotonaldehyde based on inadequate human evidence and limited evidence for carcinogenicity in animals (increased incidence of hepatocellular neoplasms in male rats exposed for up to 113 weeks to crotonaldehyde in drinking water by Chung et al., 1986). The source document for this assessment, which was verified 03/07/91, was the HEED (U.S. EPA, 1989). IRIS (U.S. EPA, 2008) does not include derivation of quantitative cancer risk values for crotonaldehyde due to the limited nature of the positive cancer data (positive response only in the low-dose group). The HEED (U.S. EPA, 1989) had derived an oral slope factor of 1.9 per mg/kg-day for this chemical by dropping the high-dose group, and this value is reported in the HEAST (U.S. EPA, 1997). The International Agency for Research on Cancer (IARC, 1995) assigned crotonaldehyde to Group 3, *Not Classifiable as to its Carcinogenicity to Humans*, based on inadequate data in humans and animals. ACGIH (2008) classified crotonaldehyde as a Group A3 carcinogen (*Confirmed Animal Carcinogen with Unknown Relevance to Humans*) based on the Chung et al. (1986) study.

Literature searches were conducted from the 1960s through February 2010 for studies relevant to the derivation of provisional toxicity values for crotonaldehyde. Databases searched included: MEDLINE, TOXLINE (and NTIS), CCRIS, DART, EMIC, HSDB, GENETOX, RTECS, TSCATS, CANCERLIT, Chemical Abstracts, BIOSIS, and Current Contents (last 6 months).

REVIEW OF PERTINENT DATA

Human Studies

In a study examining the role of oxidative stress in the etiology of Alzheimer's disease, Kawaguchi-Niida et al. (2006) used immunohistochemical analysis to measure levels of protein-bound crotonaldehyde in hippocampi obtained at autopsy of Alzheimer's disease patients and age-matched controls. Crotonaldehyde is formed endogenously during lipid peroxidation and reacts with proteins to form stable adducts (WHO, 2008). In Alzheimer's disease patients, statistically significant ($p < 0.01$) higher levels of protein-bound crotonaldehyde were observed when compared with age-matched controls. In addition, the protein-bound crotonaldehyde was localized in reactive astrocytes and microglia around senile plaques in Alzheimer's patients.

Animal Studies

Oral Exposure

Subchronic Studies—Groups of 30 male and 30 female Sprague-Dawley rats were treated with crotonaldehyde (purity not reported) in water by daily gavage for 91–93 days at dose levels of 0, 5, 15, or 45 mg/kg-day (TRL, 1986). Groups of 10 rats per sex from each dose group were sacrificed on Day 42 or 43, and remaining surviving rats were sacrificed on Days 91–93. Clinical signs and behavior were noted twice daily, and food consumption and body weight were recorded weekly. Ophthalmoscopic examinations were performed before treatment and at 13 weeks. Blood and urine samples were collected before exposure (from a fifth group of 10 males and 10 females, and at the interim and final sacrifices. From these samples, hematology, including hemoglobin [Hgb], hematocrit [Hct], erythrocyte count [RBC], mean cell volume [MCV], mean cell hemoglobin [MCH], mean cell hemoglobin concentration [MCHC],

total and differential white blood cell [WBC] count, and platelet count, was assessed. Serum chemistry (alkaline phosphatase [ALP], blood urea nitrogen [BUN], aspartate aminotransferase [AST], alanine aminotransferase [ALT], glucose, total protein, albumin, globulin, total bilirubin, electrolytes, inorganic phosphate, carbon dioxide, cholesterol, and creatinine), and urine chemistry (pH, specific gravity, glucose, protein, ketones, bilirubin, urobilinogen, and sediment microscopy) were also assessed from these samples. Upon sacrifice, all animals were subjected to gross necropsy, and the following organs were weighed: brain, heart, liver, spleen, kidneys, adrenals, thyroid, parathyroid, testes with epididymides, and ovaries. Histopathology (>25 tissues) was performed on all animals of the control and high-dose groups; the liver, kidneys, heart, and forestomach only were examined histologically in all low- and mid-dose rats.

There were two mortalities unrelated to treatment; one low-dose female and one high-dose female (TRL, 1986). Exposure-related clinical signs of toxicity were observed only in the high-dose group. Immediately after dosing, salivation (lasting about 15 minutes) was observed in about half of the high-dose rats; this effect was observed throughout the study. Transient hypoactivity, lasting less than an hour immediately following dosing, was observed in a maximum of 15% of high-dose rats. Labored respiration was also observed in 5–7% of high-dose rats but only during Weeks 1 and 2. Scattered incidences of other clinical signs including lacrimation, rales, prostration, tremors, retching, hypothermia, and/or ataxia occurred in the high-dose animals; no further details were provided. Statistically significant ($p \leq 0.05$) effects of treatment on food consumption, weight gain, and body weight were only apparent in high-dose males; these changes were largely confined to Weeks 1 and 2 on study. During Weeks 10–12, food consumption in high-dose males was significantly ($p \leq 0.05$) lower than controls, but there were no statistically significant effects on body weight or weight gain. By study termination, the mean body weight of high-dose males was decreased by 6% compared with controls; the difference was not statistically significant. Body weight, weight gain, and food consumption were not significantly affected by treatment in high-dose females or at lower doses. Ophthalmoscopy was not impacted by exposure to crotonaldehyde. Treatment-related serum chemistry changes were confined to males, which exhibited statistically significantly ($p \leq 0.05$) decreased ALP (22–42% lower than controls at all doses), AST (high-dose only), and ALT (high-dose only) activities at the interim (but not final) sacrifice. The biological significance of decreases in these serum biomarkers of liver toxicity is uncertain. Examination of the data revealed no consistent exposure-related differences between the exposed and control groups in hematology or urine chemistry variables, or in other serum chemistry parameters. Increases in the relative, but not absolute, weights of brain (7.5% higher than controls, $p \leq 0.05$) and kidneys (7.6% higher than controls, $p \leq 0.05$) were observed in high-dose males; these changes may have been related to the lower body weight in this exposure group. Exposure-related necropsy and histology findings were restricted to the forestomach of high-dose animals. At the interim sacrifice, 8/10 high-dose males exhibited gross lesions described as white, raised, and/or pitted areas of the forestomach; these lesions were also observed at the final sacrifice in 20/20 males and 8/19 females in the high-dose group. Histology revealed hyperkeratosis in the forestomach of all high-dose rats (except the female that died early) and parakeratosis in one high-dose animal of each sex. No changes were observed in the glandular portion of the stomach. This study identified a NOAEL of 15 mg/kg-day and a LOAEL of 45 mg/kg-day for hyperkeratosis of the forestomach and clinical signs of toxicity (hypoactivity, labored respiration) in Sprague-Dawley rats.

In another subchronic gavage study, groups of 10 male and 10 female F344 rats were administered crotonaldehyde (purity not reported) in corn oil by gavage at doses of 0 (vehicle control), 2.5, 5, 10, 20, or 40 mg/kg-day, 5 days/week for up to 13 weeks (Hazleton Laboratories, 1986a). The animals were subjected to twice daily mortality/morbidity checks; food consumption and body weight were recorded weekly. Blood samples were collected on Days 4 and 16, and at the beginning of Week 13 for hematology (RBC, Hgb, Hct, MCV, MCH, MCHC, total and differential WBC, reticulocyte count, platelet count, and RBC, WBC, and platelet morphology) and serum chemistry (sorbitol dehydrogenase [SDH], gamma glutamyltransferase [GGT], ALT, ALP, BUN, creatinine) analyses. The report suggested that urine samples were also collected but did not discuss analysis of the samples. Assessment of sperm morphology and vaginal cytology was performed at the end of the study on animals in the 0-, 2.5-, 5.0-, and 10-mg/kg-day groups. At sacrifice, all animals were subjected to gross necropsy, and the brain, heart, liver, right kidney, lung, and thymus were weighed. A complete histopathological examination was performed on all gross lesions and tissue masses, all control rats, all rats in the highest dose group with at least 60% survivors at time of sacrifice, and all rats in the higher dose groups where death occurred prior to study termination. Target organs (nasal cavities and forestomach) from all rats in all groups were also examined microscopically. The National Toxicology Program convened a Pathology Working Group (NTP-PWG, 1987) to review selected slides from this study and a companion study of B6C3F1 mice (Hazleton Laboratories, 1986b).

Early deaths occurred at the following incidences in the control through high-dose groups: 0/10, 0/10, 0/10, 3/10, 3/10, and 5/10 for males, and 1/10, 0/10, 1/10, 1/10, 7/10, and 5/10 for females (Hazleton Laboratories, 1986a). The NTP-PWG (1987) concluded that nearly all of the early deaths were associated with gavage trauma and/or oil in the lungs, and that the early deaths should not be used as criteria for selecting doses for a chronic study. High-dose males showed decreases in body-weight gain at Weeks 11 and 13; mean terminal body weight was about 10% lower than the control mean ($p < 0.05$). Review of the data showed that terminal body weights in other exposed male groups and female groups were not different from controls. Scattered statistically significant differences in organ weights and in hematological and clinical chemistry variables occurred in some exposed groups, but none were considered biologically significant by the NTP-PWG (1987). At gross necropsy, exposure-related lesions were only observed in the forestomach (thickened forestomach and/or forestomach nodules) of rats of both sexes in the two highest dose groups (20 and 40 mg/kg-day). Microscopic examination showed epithelial hyperplasia in the forestomach beginning at the 10-mg/kg-day dose level. Incidences of epithelial hyperplasia in the 10, 20, and 40 mg/kg-day groups were 3/10, 3/10, and 8/10 in males and 1/10, 4/10, and 8/10 in females. None of the rats in the control or lower dose groups showed epithelial hyperplasia of the forestomach. The NTP-PWG (1987) concluded that no-effect levels for forestomach lesions were 5 and 10 mg/kg-day for males and females, respectively, noting that the lesion was equivocal in the single affected female in the 10-mg/kg-day group. The corresponding LOAELs for forestomach lesions are 10 and 20 mg/kg-day for males and females, respectively. The only other exposure-related microscopic change reported by Hazleton Laboratories (1986a) was described as nasal inflammation. Incidences at control through 40 mg/kg-day were 0/10, 0/10, 0/10, 3/10, 3/10, and 7/10 in males; and 1/10, 0/10, 1/10, 1/10, 4/10, and 6/10 in females. The NTP-PWG (1987) concluded that the nasal lesions were serous exudation and not acute inflammation as reported by Hazleton

Laboratories (1986a), and that the effect was likely a localized effect from exhaled crotonaldehyde rather than an effect from blood-circulated crotonaldehyde. For the purpose of this review, a LOAEL of 10 mg/kg-day is identified based on forestomach lesions in males; the NOAEL is 5 mg/kg-day.

Hazleton Laboratories (1986b) also conducted a study of crotonaldehyde exposure in mice. Groups of 10 male and 10 female B6C3F1 mice were administered crotonaldehyde in corn oil by gavage at doses of 0 (vehicle control), 2.5, 5, 10, 20, or 40 mg/kg-day, 5 days/week for up to 13 weeks. The original report for this study was not available, and the NTP-PWG (1987) review of the data did not report results. According to NTP-PWG (1987), the study followed the same protocol as for the companion study in F344 rats (Hazleton Laboratories, 1986a), excluding the clinical laboratory studies. Histopathological evaluation and selection of tissues examined microscopically from the control and treated-mouse groups followed the criteria used in the F344 rat study (Hazleton Laboratories, 1986a). The forestomach was designated as the target organ due to lesions observed at initial histopathological examination. According to study protocol, tissue sections of the forestomach from all mice in the control and treated groups were examined microscopically. The NTP-PWG (1987) reviewed selected slides from this study.

All mice survived treatment to terminal sacrifice, and there were no statistically significant differences between treated and control groups for body-weight gain (Hazleton Laboratories, 1986b; NTP-PWG, 1987). Although statistically significant changes in absolute organ weights and organ/body-weight ratios were seen between treated and control groups, these differences were not considered toxicologically significant. Treatment related lesions were not observed at gross necropsy. Microscopic lesions were observed in the forestomach mucosa of males and females from the high-dose group (40 mg/kg) only. These lesions consisted of focal-to-diffuse thickening of the epithelium, with an irregular basal layer of the epithelial lining of the forestomach (hyperplasia). The NTP-PWG (1987) report did not give incidences of these effects, and the original report was not available. In addition to the epithelial hyperplasia, two males in the high-dose (40 mg/kg-day) group had moderate chronic inflammatory lesions into the forestomach submucosa. Inflammation of the forestomach was not seen in the controls, high-dose female mice, or any of the lower dose groups. No significant pathological findings were reported for lower dose groups. The NTP-PWG (1987) identified a NOAEL of 20 mg/kg-day and a LOAEL of 40 mg/kg-day for epithelial hyperplasia of the forestomach in both sexes of B6C3F1 mice exposed to crotonaldehyde; these effect levels were adopted for this review.

Chronic Studies—In the only chronic study available, male F344 rats (23–27 per group) were exposed to crotonaldehyde (>99% pure) in drinking water for 113 weeks at concentrations of 0, 0.6, or 6.0 mM (0, 42, or 421 mg/L) beginning at 6 weeks of age (Chung et al., 1986). Total doses were reported to be 0, 9.5, or 70 mmol/rat based on average water consumption data for an exposure period of 113 weeks. Corresponding daily doses of 0, 2, and 17 mg/kg-day were calculated using approximate average body weights of 425 g for the control and 0.6-mM groups, and 375 g for the 6.0-mM group (from Figure 2 in Chung et al., 1986). Drinking water consumption was measured twice weekly, and body weight was measured weekly for 40 weeks and then biweekly. Upon sacrifice at the end of exposure, gross necropsy was performed on all animals, and histopathology was assessed on gross lesions and major organs (not specified).

Survival percentages were greater than 95% for exposed and control groups through 70 weeks of exposure but began to decline thereafter (Chung et al., 1986). At 110 weeks, respective survival percentages for the control, low-, and high-dose groups were 70% (16/23), 63% (17/27), and 56% (13/23). Differences from controls were not statistically significant. Body weight was decreased in the high-dose group beginning at the eighth week of exposure and continuing throughout the study (approximately 10% lower than controls at study termination based on visual inspection of data presented graphically). Water consumption was reduced in high-dose animals (15 mL/day) compared with low-dose animals (20 mL/day), but water consumption in controls was not reported.

Liver tumors, described as neoplastic nodules or hepatocellular carcinomas, were found at statistically significant ($p < 0.001$) elevated incidences in the low-dose, but not high-dose, group compared with controls (Chung et al., 1986). Incidences of neoplastic nodules in the control through high-dose groups were 0/23, 9/27, and 1/23. Two rats in the low-dose group showed hepatocellular carcinomas in addition to the neoplastic nodules. Incidences of altered liver foci, considered to be a preneoplastic lesion, were elevated in both exposed groups compared with controls (1/23, 23/27, and 13/23 in the control, low-, and high-dose groups, respectively; $p < 0.001$). The number of altered liver foci per square centimeter was also increased at both doses (0.1 ± 0.4, 12.4 ± 7.4, and 3.5 ± 3.8 in control through high-dose groups; $p < 0.001$). Moderate-to-severe liver damage (fatty metamorphosis, focal liver necrosis, fibrosis, cholestasis, and mononuclear cell infiltration) occurred in the 10/23 high-dose rats that did not have preneoplastic or neoplastic lesions in the liver. No mention was made of degenerative liver lesions in the control or low-dose groups. Inspection of the tumor data showed that incidences of neoplastic lesions in tissues other than the liver were not affected by treatment. However, two rats at the low-dose level had bladder tumors as well. No mention was made of nonneoplastic lesions in tissues other than the liver. A NOAEL and LOAEL cannot be identified from this study due to the limited assessment and reporting of noncancer endpoints and confounding due to the elevated incidence of liver tumors at the low-dose.

Reproductive/Developmental Studies—Hazleton Laboratories (1987) also conducted a reproduction study in rats. Groups of 20 male and 20 female F344 rats were administered crotonaldehyde in corn oil by daily gavage at doses of 0 (vehicle control), 2.5, 5, or 10 mg/kg-day. Males were dosed for 61 days prior to mating and during the 7-day cohabitation period and then sacrificed. Females were dosed for 30 days prior to mating, during mating, and through gestation to Postpartum Day 5, when they and their surviving pups were sacrificed. Females that did not get pregnant were sacrificed 30 days after the cohabitation period. Mortality checks were performed twice daily. Body weights were recorded weekly, and pregnant females were weighed on Gestation Days 0, 7, 14, and 20; at parturition; and again at termination. All females were subjected to a vaginal cytology evaluation prior to mating and again just prior to termination for nonpregnant females. At study termination, all males were subjected to a sperm morphology evaluation, and weights were taken of the right testes and epididymis. Blood was collected from all animals at termination for possible hormone evaluations. Gross examination was conducted on all animals at necropsy, and histopathology was performed on the reproductive tissues (testes, epididymis, vagina, uterus, cervix, oviducts, and ovaries). Pups were weighed, counted, sexed, and examined at birth and again 5 days later.

All animals except for one mid-dose male survived to study termination; the cause of death was reported not to be treatment-related (Hazleton Laboratories, 1987). Clinical signs were few and not related to treatment. The data showed no significant changes in body weights, testes weights, or epididymis weights among any treated male rats. The data also showed no significant differences from controls in body weights among pregnant females through gestation to Lactation Day 5. Among female rats that did not get pregnant, there appeared to be a decrease in body weight in the treated groups relative to controls during the postmating period. However, the control group for this analysis included only two animals, and the treated groups also included small numbers (n = 3–5), which limits interpretation of these results. The researchers suggested that this effect was probably not related to treatment. No significant histological lesions were observed in males or females. The data for reproductive and litter parameters showed no effect of treatment. Results of the sperm morphology and vaginal cytology studies were not presented. The high dose of 10 mg/kg-day was a NOAEL for parental and reproductive toxicity in this study.

Inhalation Exposure

WHO (2008) briefly summarized a subchronic inhalation study of crotonaldehyde published in Russian (Voronin et al., 1982). Efforts to obtain the original study were not successful. WHO (2008) reported that rats and mice were exposed continuously to crotonaldehyde for 3 months (exposure concentrations not reported). According to WHO (2008), exposure to 1.2 mg/m^3 crotonaldehyde was associated with alterations of motor activity and blood levels of hemoglobin. It was not clear from the summary whether both species were affected, or the direction or magnitude of the effects; no other information was available.

In a study published only as an abstract, Hilkevich (2002) tested the acute and chronic effects of inhaled crotonaldehyde in rats. According to the abstract, concentrations of 1, 5, 87, and 130 mg/m^3 were tested in the acute experiments, while concentrations of 0.14, 0.56, and 5.18 mg/m^3 were tested for chronic toxicity; exposure durations were not reported. The authors reported that acute exposure to concentrations of 87–130 mg/m^3 resulted in "oppression" of the blood circulation and toxicity to WBCs but no changes in peripheral nerve, muscle, or liver function. Chronic exposure to concentrations of 0.56 and 5.18 mg/m^3 reportedly resulted in stimulation of the central nervous system, "oppression" of respiratory receptor function, pulmonary hypoventilation, and cytotoxicity but no effects on liver function (no other details provided). The authors stated that there were no effects at 0.14 mg/m^3. No other information was available in the abstract.

Other Studies

Acute or Short-term Studies

Borriston Research Laboratories (1980) evaluated the toxicity of crotonaldehyde (purity not reported) in Sprague-Dawley rats (groups of 5/sex/dose) exposed via the diet to target doses of 0, 22, 44, 88, or 175 mg/kg-day for 14 days. Daily observations and measurements of food consumption were performed, and body weight was recorded weekly. At sacrifice at the end of exposure, all animals received gross necropsy, and liver and kidney weights were measured. Based on food consumption and body-weight data, the authors calculated doses of 19, 36, 73, and 139 mg/kg-day in males, and 17, 36, 68, and 136 mg/kg-day in females. Inspection of the data indicated that there were no treatment-related effects on clinical signs, body weight, food

consumption, organ weights, or gross necropsy findings. No organs were examined microscopically.

DERIVATION OF PROVISIONAL SUBCHRONIC AND CHRONIC ORAL RfD VALUES FOR CROTONALDEHYDE

Oral studies of crotonaldehyde include two subchronic gavage studies (TRL, 1986; Hazleton Laboratories, 1986a; NTP-PWG, 1987) and a reproduction study (Hazleton Laboratories, 1987) in rats, one subchronic gavage study in mice (Hazleton Laboratories, 1986b; NTP-PWG, 1987), and a chronic drinking water study conducted primarily as a cancer bioassay in rats (Chung et al., 1986). Table 1 summarizes these data. The three 13-week subchronic gavage studies all showed effects in the forestomach of both rats and mice. The chronic drinking water study did not report forestomach lesions but observed altered foci, tumors, and degenerative changes in the liver. No significant effects on reproduction were observed in rats treated by gavage with up to 10 mg/kg-day of crotonaldehyde.

Subchronic p-RfD

Hazleton Laboratories, 1986a and NTP-PWG, 1987 observed forestomach lesions at doses ≥10 mg/kg-day in rats and at 40 mg/kg-day in mice. The absence of forestomach lesions in the chronic drinking water study suggests that the observed lesions may be due in part to bolus dosing in the subchronic studies. However, crotonaldehyde is one of the class of α,β-unsaturated aldehydes that has been noted for its ability to bind covalently to protein and DNA. It is possible that this binding may be a mechanism of forestomach lesion development and would be relevant in humans. In the absence of more definitive information, the forestomach lesions are considered a valid endpoint for human health risk assessment.

The lowest LOAEL and NOAEL were observed in the Hazleton Laboratories (1986a; NTP-PWG, 1987) study of rats; this study was chosen for dose-response modeling (see Table 2). Data for forestomach lesion incidence in both male and female rats were modeled. Appendix A contains details of the modeling and plots of the best fitting models. The BMD_{10} for male rats was 7.9 mg/kg-day, and the $BMDL_{10}$ was 4.8 mg/kg-day. The BMD_{10} for female rats was 10.0 mg/kg-day, and the $BMDL_{10}$ was 5.5 mg/kg-day. The lower $BMDL_{10}$ of 4.8 mg/kg-day from the male rats was selected as the point-of-departure (POD) for derivation of the subchronic p-RfD. The $BMDL_{10}$ (4.8 mg/kg-day) based on gavage dosing 5 days/week was converted to an equivalent continuous (7 days/week) dose of 3.4 mg/kg-day.

Table 1. Summary of Oral Noncancer Dose-response Information

Species and Study Type (*n*/sex/group)	Exposure	NOAEL (mg/kg-day)	LOAEL (mg/kg-day)	Duration-adjusted[a] NOAEL (mg/kg-day)	Duration-adjusted[a] LOAEL (mg/kg-day)	Responses at the LOAEL	Comments	Reference
Sprague-Dawley Rat Subchronic Gavage 30/sex/dose	0, 5, 15, and 45 mg/kg-day daily for up to 93 days	15	45	15	45	Hyperkeratosis of the forestomach and clinical signs		TRL, 1986
F344 Rat Subchronic Gavage 10/sex/dose	0, 2.5, 5, 10, 20, and 40 mg/kg-day, 5 days/week for 13 weeks	5	10	3.6	7.1	Forestomach lesions		Hazleton Laboratories, 1986a; NTP-PWG, 1987
B6C3F1 Mouse Subchronic Gavage 10/sex/dose	0, 2.5, 5, 10, 20, and 40 mg/kg-day, 5 days/week for 13 weeks	20	40	14	29	Forestomach lesions		Hazleton Laboratories, 1986b; NTP-PWG, 1987
F344 Rat Chronic Drinking water 23–27 M/dose	Approximately 0, 2, and 17 mg/kg-day daily in drinking water for 113 weeks	NA	NA	NA	NA	NA	Altered hepatic foci and liver tumors at low dose; also degenerative liver lesions at high dose	Chung et al., 1986
F344 Rat Reproduction Gavage 20/sex/dose	0, 2.5, 5, and 10 mg/kg-day daily prior to and during mating and through gestation and lactation	10	NA	10	NA	NA		Hazleton Laboratories, 1987

[a]Adjusted for continuous exposure

Table 2. Dose-response Data for Forestomach Hyperplasia in F344 Rats[a]		
	Incidence of Forestomach Hyperplasia	
Dose (mg/kg-day)	Males	Females
0	0/10	0/10
2.5	0/10	0/10
5	0/10	0/10
10	3/10	1/10
20	3/10	4/10
40	8/10	8/10

[a]Hazleton Laboratories, 1986a; NTP-PWG, 1987

A **subchronic p-RfD** was derived by dividing the duration-adjusted $BMDL_{10}$ of 3.4 mg/kg-day by a UF of 300, as shown below:

Subchronic p-RfD = $BMDL_{10} \div UF$
= 3.4 mg/kg-day ÷ 300
= **0.01 mg/kg-day or 1×10^{-2} mg/kg-day**

The composite UF of 300 was composed of the following UFs:

- UF_H: A factor of 10 is applied for extrapolation to a potentially susceptible human subpopulation because data for evaluating susceptible human response are insufficient.
- UF_A: A factor of 10 is applied for animal-to-human extrapolation because data for evaluating relative interspecies sensitivity are insufficient.
- UF_D: The database for oral exposure to crotonaldehyde consists of subchronic toxicity studies in two species and a single-generation reproduction study in rats. A factor of 3 ($10^{0.5}$) is applied for database inadequacies because data for evaluating developmental toxicity are inadequate.

Confidence in the principal study (Hazleton Laboratories, 1986a; NTP-PWG, 1987) is high because the study tested 10 rats per sex at six dose levels (including controls), a broad array of endpoints were evaluated, and a review of the pathology results was conducted and confirmed by NTP-PWG. Confidence in the database is medium. Additional subchronic gavage studies in rats and mice provided support for the findings of the principal study, and a single generation reproduction study found no effects. However, it is unclear from the existing database the extent to which the findings of the gavage studies reflect the bolus-dosing protocol. In addition, teratogenicity has not been evaluated. Reflecting high confidence in the principal study and medium confidence in the database, confidence in the subchronic p-RfD is medium.

Chronic p-RfD

The chronic drinking water study in rats observed altered foci, tumors, and degenerative changes in the liver (Chung et al., 1986). However, this study cannot be used as the basis for a chronic p-RfD due to the limited assessment and reporting of noncancer endpoints and confounding due to the elevated incidence of liver tumors at the low-dose.

In the absence of adequate chronic data, the POD used to derive the subchronic p-RfD (adjusted $BMDL_{10}$ of 3.4 mg/kg-day for forestomach lesions in male rats) was also used to derive the chronic p-RfD. The **chronic p-RfD** for crotonaldehyde was derived as follows:

Chronic p-RfD = $BMDL_{10} \div UF$
= 3.4 mg/kg-day ÷ 3000
= **0.001 mg/kg-day or 1×10^{-3}**

The composite UF of 3000 was composed of the following UFs:

- UF_H: A factor of 10 is applied for extrapolation to a potentially susceptible human subpopulation because data for evaluating susceptible human response are insufficient.
- UF_A: A factor of 10 is applied for animal-to-human extrapolation because data for evaluating relative interspecies sensitivity are insufficient.
- UF_D: The database for oral exposure to crotonaldehyde consists of subchronic toxicity studies in two species and a single-generation reproduction study in rats. A factor of 3 ($10^{0.5}$) is applied for database inadequacies because data for evaluating developmental toxicity are inadequate.
- UF_S: A factor of 10 is applied for using data from a subchronic study to assess potential effects from chronic exposure because data for evaluating response after chronic exposure are inadequate.

As discussed for the subchronic p-RfD, confidence in the principal study is high. However, confidence in the database is reduced to low for the chronic p-RfD due to the absence of an adequate chronic study and uncertainty whether the value based on forestomach lesions would be protective for degenerative liver lesions, which were seen in the chronic study but not in the subchronic studies. Overall, confidence in the chronic p-RfD is low.

FEASIBILITY OF DERIVING PROVISIONAL SUBCHRONIC AND CHRONIC INHALATION RfC VALUES FOR CROTONALDEHYDE

The database for inhalation toxicity of crotonaldehyde is limited to a subchronic study (Voronin et al., 1982) reported only in summary form by WHO (2008), and a chronic study reported only as an abstract (Hilkevich, 2002). These studies are inadequate for derivation of inhalation toxicity values for crotonaldehyde.

PROVISIONAL CARCINOGENICITY ASSESSMENT FOR CROTONALDEHYDE

A provisional carcinogenicity assessment was not prepared for crotonaldehyde because IRIS (U.S. EPA, 2008) includes a cancer assessment for this compound.

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APPENDIX A. DETAILS OF BENCHMARK DOSE MODELING FOR SUBCHRONIC p-RfD

Model Fitting Procedure for Quantal Noncancer Data

The model fitting procedure for dichotomous noncancer data is as follows. All available dichotomous models in the EPA BMDS (version 2.1) are fit to the incidence data using the extra risk option. The multistage model is run for all polynomial degrees up to n-1 (where n is the number of dose groups including control). Adequate model fit is judged by three criteria: goodness-of-fit p-value ($p > 0.1$), visual inspection of the dose-response curve, and scaled residual at the data point (except the control) closest to the predefined benchmark response. Among all the models providing adequate fit to the data, the lowest BMDL is selected as the point of departure when the difference between the BMDLs estimated from these models is more than 3-fold (unless it appears to be an outlier); otherwise, the BMDL from the model with the lowest Akaike Information Criterion (AIC) is chosen. In accordance with EPA (2000) guidance, benchmark doses (BMDs) and lower bounds on the BMD (BMDLs) associated with an extra risk of 10% are calculated for all models.

Model Fitting Results for Forestomach Hyperplasia in Male F344 Rats (Hazleton Laboratories, 1986a; NTP-PWG, 1987)

Applying the procedure outlined above to the data for forestomach hyperplasia in male rats, adequate model fit was achieved with all models. Table A-1 shows the modeling results. BMDLs from models providing adequate fit differed by less than 3-fold. In accordance with EPA (2000) guidance, the lowest AIC was selected from among models providing adequate fit. For this data set, the resulting benchmark dose (BMD_{10}) and associated 95% lower confidence limit ($BMDL_{10}$) were 7.88 and 4.77 mg/kg-day, respectively, based on the log probit model. Figure A-1 shows the fit of the log probit model to the data.

Table A-1. Model Predictions for the Incidence of Forestomach Hyperplasia in Male F344 Rats[a]

Model	Degrees of Freedom	χ^2	χ^2 Goodness-of-Fit *p*-Value[b]	AIC	BMD_{10} (mg/kg-day)	$BMDL_{10}$ (mg/kg-day)
Gamma[c]	4	2.89	0.58	41.55	7.94	3.69
Logistic	4	5.13	0.27	44.13	11.46	7.91
Log logistic[d]	4	3.00	0.56	41.68	7.93	3.95
Log probit[d]	**4**	**2.73**	**0.60**	**41.34**	**7.88**	**4.77**
1-degree multistage[e]	5	3.84	0.57	42.22	4.06	2.66
2-degree multistage[e]	4	3.18	0.53	41.91	7.98	3.40
3-degree multistage[e]	4	3.18	0.53	41.91	7.98	3.35
4-degree multistage[e]	4	3.18	0.53	41.91	7.98	3.31
5-degree multistage[e]	4	3.18	0.53	41.91	7.98	3.28
Probit	4	4.85	0.30	43.60	10.74	7.49
Weibull[c]	4	2.93	0.57	41.67	7.82	3.59
Quantal-linear	5	3.84	0.57	42.22	4.06	2.66

[a]Hazleton Laboratories, 1986a; NTP-PWG, 1987
[b]Values <0.10 fail to meet conventional goodness-of-fit criteria
[c]Power restricted to ≥1
[d]Slope restricted to ≥1
[e]Betas restricted to ≥0

AIC = Akaike Information Criterion; BMD_{10} = maximum likelihood estimate of the dose/concentration associated with the selected benchmark response; $BMDL_{10}$ = 95% lower confidence limit on the BMD

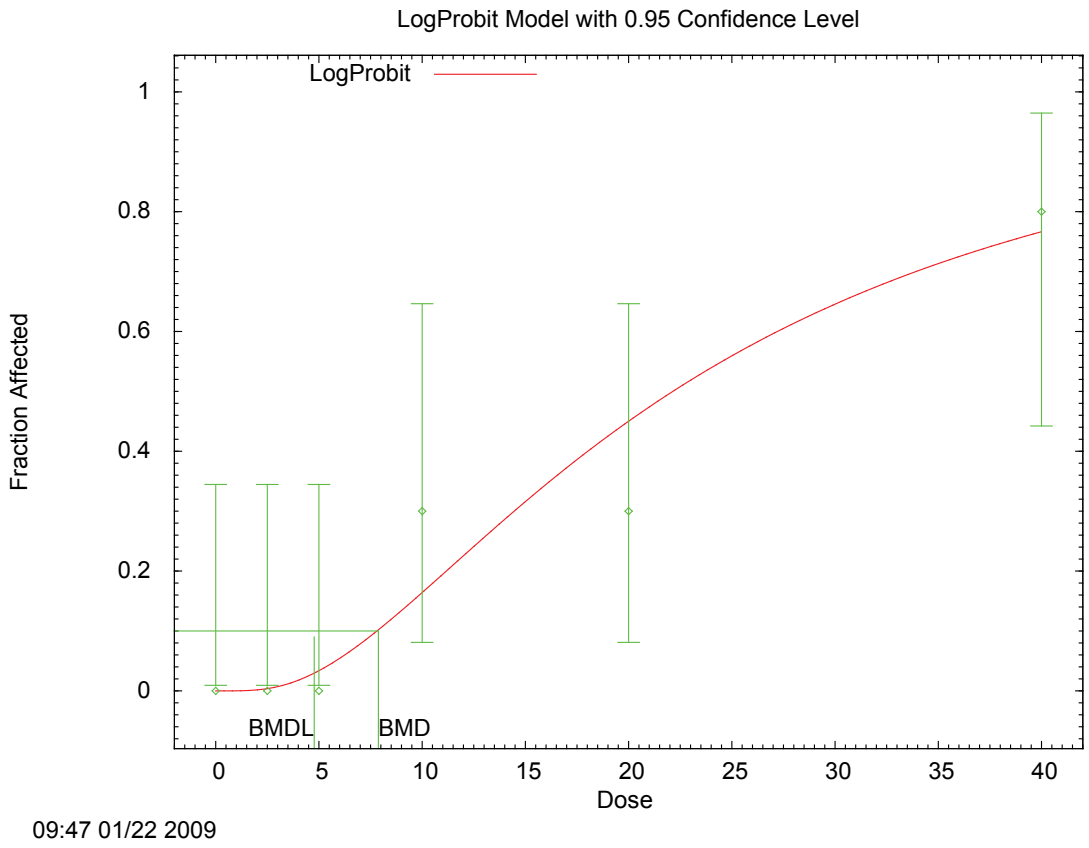

Figure A-1. Fit of Log Probit Model to Data on Forestomach Hyperplasia in Male F344 Rats (Hazleton Laboratories, 1986a; NTP-PWG, 1987)

BMDs and BMDLs indicated are associated with an extra risk of 10% and are in units of mg/kg-day.

Model Fitting Results for Forestomach Hyperplasia in Female F344 Rats (Hazleton Laboratories, 1986a; NTP-PWG, 1987)

Applying the procedure outlined above to the data for forestomach hyperplasia in female rats, adequate model fit was achieved with all models. Table A-2 shows the results. BMDLs from models providing adequate fit differed by approximately 3-fold, with the quantal-linear and 1-degree multistage model that gave the low BMDL estimate having relatively poor fit and appearing to be an outlier. The BMDL estimates from the remaining models differed by only 1.5-fold. In accordance with EPA (2000) guidance, the lowest AIC was selected from among these remaining models. The resulting BMD_{10} and $BMDL_{10}$ were 9.98 and 5.45 mg/kg-day, respectively, based on the 2-degree multistage model. Figure A-2 shows the fit of the 2-degree polynomial multistage model to the data.

Table A-2. Model Predictions for the Incidence of Forestomach Hyperplasia in Female F344 Rats[a]

Model	Degrees of Freedom	χ^2	χ^2 Goodness-of-Fit *p*-Value[b]	AIC	BMD_{10} (mg/kg-day)	$BMDL_{10}$ (mg/kg-day)
Gamma[c]	4	0.27	0.99	34.39	10.63	5.92
Logistic	4	2.26	0.69	37.00	13.06	8.96
Log logistic[d]	4	0.21	1.00	34.32	10.75	6.18
Log probit[d]	4	0.10	1.00	34.13	10.55	6.28
1-degree multistage[e]	5	4.09	0.54	38.05	4.39	2.83
2-degree multistage[e]	**5**	**0.48**	**0.99**	**32.78**	**9.98**	**5.45**
3-degree multistage[e]	5	0.48	0.99	32.78	9.98	5.35
4-degree multistage[e]	5	0.48	0.99	32.78	9.98	5.35
5-degree multistage[e]	5	0.48	0.99	32.78	9.98	5.35
Probit	4	1.79	0.77	36.31	12.51	8.57
Weibull[c]	4	0.51	0.97	34.76	10.38	5.56
Quantal-linear	5	4.09	0.54	38.05	4.39	2.83

[a]Hazleton Laboratories, 1986a; NTP-PWG, 1987
[b]Values <0.10 fail to meet conventional goodness-of-fit criteria
[c]Power restricted to ≥1
[d]Slope restricted to ≥1
[e]Betas restricted to ≥0

AIC = Akaike Information Criterion; BMD_{10} = maximum likelihood estimate of the dose/concentration associated with the selected benchmark response; $BMDL_{10}$ = 95% lower confidence limit on the BMD

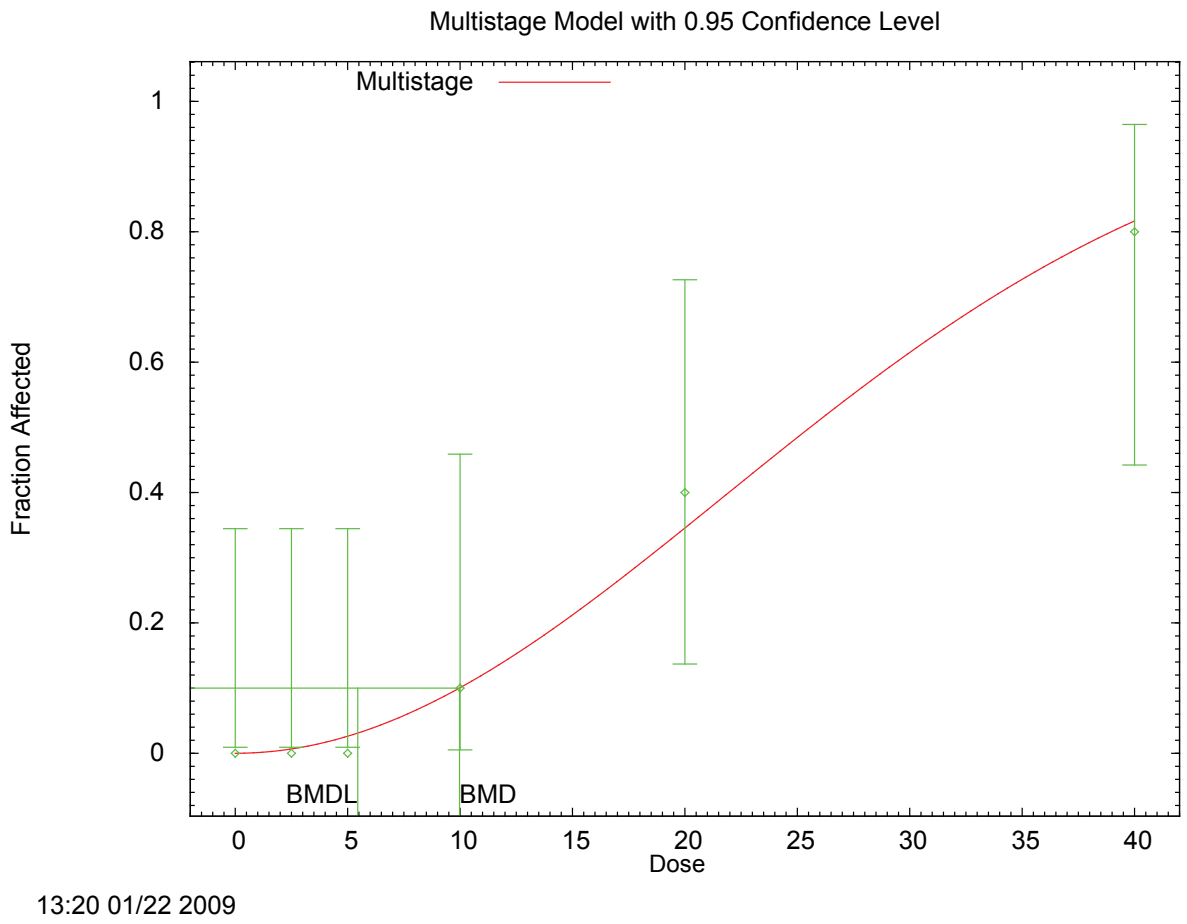

Figure A-2. Fit of Multistage (2-Degree Polynomial) Model to Data on Forestomach Hyperplasia in Female F344 Rats (Hazleton Laboratories, 1986a; NTP-PWG, 1987)

BMDs and BMDLs indicated are associated with an extra risk of 10% and are in units of mg/kg day.