2. RELEVANCE TO PUBLIC HEALTH

2.1 BACKGROUND AND ENVIRONMENTAL EXPOSURES TO ACRYLAMIDE IN THE UNITED STATES

Acrylamide is an industrial chemical used mainly in the production of polyacrylamides, which are used primarily as flocculants for clarifying drinking and treating municipal and industrial effluents. In the oil industry, acrylamide is used as a flow control agent to enhance oil production from wells. Acrylamide and polyacrylamides are also used in the production of dyes and organic chemicals, contact lenses, cosmetics and toiletries, permanent-press fabrics, textiles, pulp and paper products; also in ore processing, sugar refining, and as a chemical grouting agent and soil stabilizer for the construction of tunnels, sewers, wells, and reservoirs. In 2008, approximately 141,000 metric tons of acrylamide were produced in the United States.

Acrylamide may be released to the environment during production and use of polyacrylamides, which are used as clarifiers in water treatment. Residual monomer in the polyacrylamide is the main source of drinking water contamination by acrylamide. It can be released to soil and land from plastics and dye industries and from acrylamide-containing grouting agents used in reservoirs and wells. Acrylamide is rarely found in the atmosphere.

Acrylamide is not considered highly persistent in the environment. It is expected to be highly mobile in soil and water and is highly susceptible to biodegradation in both media. It can be removed from soils by hydrolysis, though it is typically not present in soil or the atmosphere. Acrylamide is not expected to significantly bioconcentrate.

Acrylamide is sometimes present in drinking water due to leaching of monomer during treatment processes. Acrylamide was identified in food samples cooked at high temperatures. Concentrations of acrylamide in food vary with the type of food, cooking method, temperature, water content, food thickness, and length of heating. Carbohydrate-rich foods typically contain the highest levels of acrylamide.

Exposure to acrylamide occurs mainly through ingestion, dermal contact, and inhalation. Ingestion of foods containing acrylamide appears to be one of the most common methods of exposure for the general public. Average estimated intake of acrylamide from food sources ranged from 0.3 to 0.8 μg/kg bw/day. Children may be susceptible to food-borne exposure 2–3 times that of adults on a body weight basis.
Ingestion of contaminated drinking water can result in exposure to acrylamide. Inhalation of tobacco smoke, including second-hand smoke, can result in inhalation exposure for both adults and children. In the body, acrylamide can cross the placenta and result in exposure to unborn children. Breast milk has been shown to contain acrylamide in mothers with diets high in acrylamide-containing foods. Occupational exposure to acrylamide is primarily due to dermal contact when handling bags and drums of the chemical, followed by inhalation of dust or aerosols.

2.2 SUMMARY OF HEALTH EFFECTS

Neurological Effects. Ataxia and skeletal muscle weakness are hallmark symptoms of acrylamide-induced central-peripheral neuropathy. Available information in humans includes case reports and cross-sectional studies in which reported symptoms of neuropathy were primarily associated with occupational exposure potential via both inhalation and dermal contact. Numerous animal studies confirm the neurological effects of acrylamide exposure. Available information in animals primarily involves oral exposure. Clinical signs can be elicited following single oral dosing in the range of 100–200 mg/kg; lower dose levels require repeated dosing to elicit clinical signs. Histopathological evidence of acrylamide-induced peripheral neuropathy has been observed in rats receiving oral doses as low as 1 mg/kg/day for 3 months; the observed degenerative effects in peripheral nerve fibers at such dose levels have been shown to be completely reversible within a few months following the cessation of exposure.

Reproductive Effects. Pre- and postimplantation losses and decreased numbers of live fetuses have been observed in rats and mice receiving repeated oral doses of acrylamide in the range of 3–60 mg/kg/day. Results of dominant lethality testing and crossover trials indicate that acrylamide-induced reproductive effects are male mediated. Other reported effects in acrylamide-treated laboratory animals include decreases in sperm concentration and mobility, degenerative effects in spermatids and germinal epithelium of testis and epididymis, and testicular atrophy, which were observed at repeated oral doses as low as 3–5 mg/kg/day in some studies.

Cancer. Available cohort mortality studies of occupationally-exposed workers have not found associations between acrylamide and mortality from cancer. Available human data on cancer risk from acrylamide in food include case-control studies and reports from ongoing prospective cohort studies. Most of these studies found no significant associations between dietary acrylamide and the risk of cancers; however, risks for selected cancers were slightly elevated in a few instances.
Results of two similarly-designed lifetime studies in orally-exposed rats provide clear evidence of acrylamide-induced carcinogenicity. Reported effects common to both studies include significantly increased incidences of thyroid (follicular cell) tumors, mesotheliomas of the tunica vaginalis testis, and mammary gland tumors. Findings of significantly increased incidences of adrenal pheochromocytomas in male rats and tumors of the oral cavity, central nervous system (glial origin), pituitary, and clitoris or uterus in female rats in the earlier bioassay were not replicated in the second bioassay.

More recent 2-year cancer bioassays in rats and mice exposed to acrylamide via the drinking water confirm the earlier findings of acrylamide carcinogenicity. In these bioassays, treatment-related significantly increased incidences of selected tumors were observed in the epididymis, heart, pancreatic islets, and thyroid gland of male rats; clitoral gland, mammary gland, oral mucosa or tongue, skin, and thyroid gland of female rats; Harderian gland, lung, and forestomach of male mice; and Harderian gland, lung, mammary gland, ovary, and skin of female mice.

Acrylamide was reported to initiate skin tumors in female mice administered repeated oral doses of acrylamide followed by dermal applications of 12-O-tetradecanoylphorbol-13-acetate (TPA, a tumor promoter) to the shaved back.

EPA has characterized acrylamide as “likely to be carcinogenic to humans” based on findings that chronic oral exposure to acrylamide in the drinking water of laboratory animals induced significantly increased incidences of selected tumors in multiple bioassays; that oral, intraperitoneal, or dermal exposure to acrylamide initiated skin tumors in two strains of mice; that intraperitoneal injections of acrylamide induced lung tumors in one strain of mice; and that there is ample evidence for the ability of acrylamide to induce genotoxic effects in mammalian cells. The International Agency for Research on Cancer assigned acrylamide to Group 2A (probably carcinogenic to humans) based on similar assessment. The National Toxicology Program has determined that acrylamide is reasonably anticipated to be a human carcinogen based on sufficient evidence of carcinogenicity from studies in experimental animals.

**Body Weight Effects.** Depressed body weight, including actual body weight loss, and depressed body weight gain were consistently reported in laboratory animals following oral exposure to acrylamide. For example, adverse effects on body weight have been observed following single or repeated dosing at relatively high dose levels for short durations (≤5 days). In one study of rats administered oral doses as low as 15 mg/kg/day for 5 consecutive days, body weight gain was depressed by approximately 40%; higher doses (30–60 mg/kg/day) caused actual weight loss. An 8-week dosing period to dogs at
7 mg/kg/day resulted in weight loss. Weight loss was reported in cats receiving acrylamide orally for 16 weeks at 15 mg/kg/day.

**Developmental Effects.** Most animal studies found no signs of acrylamide-related overt developmental effects in the offspring of animals whose mothers had received nontoxic doses of acrylamide during the development of their fetuses and pups. However, there is some evidence that relatively low oral doses (in the range of 4–25 mg/kg/day) during pre- and postnatal periods of development may result in signs of delayed motor development, repressed learning ability and motivation, decreased brain levels of selected chemicals, and impaired neurogenesis.

### 2.3 MINIMAL RISK LEVELS (MRLs)

Estimates of exposure levels posing minimal risk to humans (MRLs) have been made for acrylamide. An MRL is defined as an estimate of daily human exposure to a substance that is likely to be without an appreciable risk of adverse effects (noncarcinogenic) over a specified duration of exposure. MRLs are derived when reliable and sufficient data exist to identify the target organ(s) of effect or the most sensitive health effect(s) for a specific duration within a given route of exposure. MRLs are based on noncancerous health effects only and do not consider carcinogenic effects. MRLs can be derived for acute, intermediate, and chronic duration exposures for inhalation and oral routes. Appropriate methodology does not exist to develop MRLs for dermal exposure.

Although methods have been established to derive these levels (Barnes and Dourson 1988; EPA 1990), uncertainties are associated with these techniques. Furthermore, ATSDR acknowledges additional uncertainties inherent in the application of the procedures to derive less than lifetime MRLs. As an example, acute inhalation MRLs may not be protective for health effects that are delayed in development or are acquired following repeated acute insults, such as hypersensitivity reactions, asthma, or chronic bronchitis. As these kinds of health effects data become available and methods to assess levels of significant human exposure improve, these MRLs will be revised.

**Inhalation MRLs**

No inhalation MRLs were derived due to the lack of appropriate data on the effects of inhalation exposure to acrylamide.
Numerous accounts of associations between exposure to acrylamide and neurological impairment are available (Auld and Bedwell 1967; Bachmann et al. 1992; Calleman et al. 1994; Davenport et al. 1976; Donovan and Pearson 1987; Dumitru 1989; Fullerton 1969; Garland and Patterson 1967; Gjerlof et al. 2001; Hagmar et al. 2001; He et al. 1989; Igisu and Matsuoka 2002; Igisu et al. 1975; Kesson et al. 1977; Mapp et al. 1977; Mulloy 1996; Myers and Macun 1991; Takahashi et al. 1971). Most associations were made from occupational exposure scenarios that likely included inhalation and dermal (and possibly oral) exposure. Acrylamide levels in the workplace air were measured in some instances; however, reliable exposure-response data are not available.

Acrylamide-induced health effects in laboratory animals following inhalation exposure to acrylamide were assessed in two studies that did not include multiple exposure levels. Mortalities and central nervous system effects were observed in dogs exposed to acrylamide dust at a concentration of 15.6 mg/m$^3$ for 6 hours/day for up to 16 days (American Cyanamid Company 1953a). There were no signs of adverse effects in similarly-exposed guinea pigs. No adverse effects were seen in cats exposed to what was described as a “saturated” vapor for 6 hours/day, 5 days/week for 3 months at a mean analytical concentration of 1.65 ppm (4.8 mg/m$^3$) (Dow Chemical Company 1957).

**Oral MRLs**

- An MRL of 0.01 mg/kg/day has been derived for acute-duration oral exposure (14 days or less) to acrylamide.

Available human data are limited to case reports that include findings of persistent peripheral neuropathy in a subject who intentionally ingested 18 g of acrylamide crystals (Donovan and Pearson 1987) and signs of central and peripheral neurological deficits in family members exposed (likely via oral and dermal routes) to acrylamide in well water at a concentration of 400 ppm (Igisu and Matsuoka 2002; Igisu et al. 1975). Epidemiologic studies designed to evaluate noncancer health effects in groups of orally-exposed subjects have not been conducted.

Single oral doses elicit clinical signs of acrylamide-induced neurological effects in laboratory animals at lethal or near-lethal doses (American Cyanamid Company 1953c; Fullerton and Barnes 1966; McCollister et al. 1964; Tilson and Cabe 1979). Repeated oral dosing elicits clinical signs at lower daily dose levels. For example, convulsions and ataxia were noted as early as day 14 in rats administered acrylamide by daily gavage at 25 mg/kg/day for 21 days (Dixit et al. 1981). Daily exposure of male mice to acrylamide in the drinking water for 12 days at an estimated dose of 25.8 mg/kg/day caused decreased rotarod
performance and increased hindlimb splay as early as days 6 and 8, respectively (Gilbert and Maurissen 1982). NTP (2011b) noted hind-leg paralysis in all male and female F344/N rats receiving acrylamide for 14 days from the drinking water at 70–77 mg/kg/day or from the food at 52–63 mg/kg/day. Among similarly-treated male and female B6C3F1 mice, hind-leg paralysis was observed in one of four males and one of four females receiving acrylamide from the drinking water at approximately 150 mg/kg/day; there were no indications of neurological effects in the male or female mice receiving acrylamide from the food at doses as high as 66–76 mg/kg/day (NTP 2011b).

Histopathological evaluations of nervous tissues following acute-duration oral exposure to acrylamide are limited to electron microscope results from peripheral nerve preparations in subgroups of control and high-dose (20 mg/kg/day) male rats administered acrylamide in the drinking water for up to 93 days (Burek et al. 1980). No signs of acrylamide-induced degenerative effects were observed after 7 days of treatment, but degeneration in axons and myelin sheath were noted after 33 days of treatment. These interim assessments were not performed at lower dose levels and the effect of treatment following 14 days (maximum acute-duration exposure period as defined by ATSDR) was not assessed.

Acrylamide-induced effects on body weight have been reported in rats following acute-duration oral exposure to acrylamide. In one study, repeated-dose oral exposure of rats to doses as low as 15 mg/kg/day for 5 consecutive days resulted in significantly depressed body weight gain (approximately 40% less than controls); higher doses (30–60 mg/kg/day) caused actual weight loss, whereas no effect on body weight was seen at a dose level of 5 mg/kg/day (Tyl et al. 2000b). Another study reported significantly lower mean body weight (magnitude 8%) as early as treatment day 13 in male rats administered acrylamide in the drinking water at a concentration resulting in an estimated dose level of 20 mg/kg/day, but no effect on body weight at an estimated dose level of 5 mg/kg/day (Burek et al. 1980).

Daily oral administration of acrylamide to male rats at 15 mg/kg/day for as little as 5 days followed by mating with unexposed female rats resulted in reduced mating, decreased fertility, and male-mediated increased pre- and postimplantation losses; these effects were not seen at 5 mg/kg/day (Sublet et al. 1989; Tyl et al. 2000b). A similarly-designed study (Tyl et al. 2000b) confirmed the findings of acrylamide-induced increased implantation loss, although, based on pairwise comparisons with controls, statistical significance was achieved only at the highest dose level (60 mg/kg/day). Tyl et al. (2000b) also noted significantly decreased body weight gain during the 5 days of dosing at 15 mg/kg/day and actual weight loss at higher doses, findings not included in the study report of Sublet et al. (1989). Based on the reproducible results for male-mediated infertility in the studies of Sublet et al. (1989) and Tyl et al.
(2000b) and the lack of supporting information regarding the body weight effects, the male-mediated infertility was selected as the critical effect for deriving an acute-duration oral MRL for acrylamide. As discussed in detail in Appendix A, a physiologically based pharmacokinetic (PBPK) rat model for acrylamide and glycidamide (Sweeney et al. 2010) was used to estimate rat dose metrics for blood time-weighted average (TWA) acrylamide and glycidamide (a readily-formed metabolite of acrylamide in aqueous environment) doses for each of the administered acrylamide dose levels in the study of Sublet et al. (1989). All dichotomous models in the EPA Benchmark Dose Software (Version 2.1.2) were fit to incidence data for fraction of nonpregnant females (number of nonpregnant females/number of sperm-positive females) using the PBPK-modeled rat blood TWA acrylamide dose as the dose metric and using the TWA glycidamide dose as the dose metric. As described in detail in Appendix A, the best-fitting model for each dose metric provided a BMDL$_{10}$ of 0.00177669 mM for rat blood TWA acrylamide and a BMDL$_{10}$ of 0.00220167 mM for rat blood TWA glycidamide. Using these BMDLs, a human PBPK model (Sweeney et al. 2010) predicted a human equivalent dose (HED) of 0.31 mg acrylamide/kg/day based on blood TWA acrylamide and a HED of 5.25 mg acrylamide/kg/day based on blood TWA glycidamide. Both acrylamide and glycidamide are widely distributed by the blood and both are reactive. However, based on uncertainty regarding the proximal toxicant(s) responsible for acrylamide-induced reproductive toxicity in the male rat, a conservative public health approach was taken and the lowest HED of 0.31 mg acrylamide/kg/day (based on blood TWA acrylamide) was selected as the point of departure (POD) for deriving an acute-duration oral MRL for acrylamide. A total uncertainty factor of 30 (3 for interspecies extrapolation using a PBPK model and 10 for human variability) was applied to the HED of 0.31 mg/kg/day, resulting in an acute-duration oral MRL of 0.01 mg/kg/day for acrylamide.

- An MRL of 0.001 mg/kg/day has been derived for intermediate-duration oral exposure (15–364 days) to acrylamide.

No human data are available regarding health effects associated with intermediate-duration oral exposure to acrylamide.

Results of available animal studies demonstrate that neurological effects in males and females and reproductive effects in males are the most sensitive noncancer effects associated with intermediate-duration oral exposure to acrylamide (Burek et al. 1980; Chapin et al. 1995; Johnson et al. 1984, 1985, 1986; NTP 2011b; Sakamoto and Hashimoto 1986; Smith et al. 1986; Tyl et al. 2000a, 2000b; Zenick et al. 1986). The lowest dose level reported to induce male-mediated implantation losses was 2.8 mg/kg/day in male Long-Evans rats receiving acrylamide from the drinking water for 80 days; this study identified a no-observed-adverse effect level (NOAEL) of 1 mg/kg/day (Smith et al. 1986). Ultrastructural
degenerative peripheral nerve changes were observed at a dose level as low as 1 mg/kg/day in male F344 rats receiving acrylamide from the drinking water for up to 93 days; this study identified a NOAEL of 0.2 mg/kg/day (Burek et al. 1980). Available data suggest that neurological effects represent a more sensitive point of departure than reproductive effects for deriving intermediate- and chronic-duration oral MRLs for acrylamide. Therefore, degenerative nerve change was selected as the critical effect for deriving an intermediate-duration oral MRL for acrylamide. The study of Burek et al. (1980) was selected as the principal study because it identified the lowest lowest-observed-adverse-effect-level (LOAEL) for the critical effect. A NOAEL/LOAEL approach was selected because results of the ultrastructural evaluations included only 3 of 10 rats/group and were reported only as the total numbers of fields (per group) with ultrastructural changes as axolemma invaginations or Schwann cells without axons and/or with degenerating myelin. The distribution of fields exhibiting ultrastructural changes among the three rats within a particular dose group was not included in the study report. The lack of adequate quantitative data precludes the utilization of benchmark dose (BMD) analysis to derive an intermediate-duration oral MRL for acrylamide. As described in detail in Appendix A, a rat PBPK model for acrylamide and glycidamide (Sweeney et al. 2010) was used to predict the rat blood TWA acrylamide dose and TWA glycidamide dose associated with the rat NOAEL of 0.2 mg/kg/day.

Based on PBPK model-predicted rat blood TWA acrylamide dose metric at the NOAEL of 0.2 mg acrylamide/kg/day, the HED is 0.038 mg acrylamide/kg/day. Based on PBPK model-predicted rat blood TWA glycidamide dose metric at the NOAEL of 0.2 mg acrylamide/kg/day, the HED is 0.28 mg acrylamide/kg/day. Using a conservative approach, derivation of an intermediate-duration oral MRL for acrylamide was performed using the lowest HED of 0.038 mg acrylamide/kg/day. A total uncertainty factor of 30 (3 for interspecies extrapolation using a PBPK model and 10 for human variability) was applied to the HED of 0.038 mg/kg/day, resulting in an intermediate-duration oral MRL of 0.001 mg/kg/day.

- An MRL of 0.001 mg/kg/day has been derived for chronic-duration oral exposure (365 days or more) to acrylamide.

No human data are available to evaluate neurological effects following chronic-duration oral exposure to acrylamide. Three chronic toxicity and carcinogenicity bioassays are available for male and female F344 rats receiving acrylamide from the drinking water for up to 2 years (Friedman et al. 1995; Johnson et al. 1984, 1985, 1986; NTP 2011b). The study of Johnson et al. (1984, 1985, 1986) included dose levels of 0, 0.01, 0.1, 0.5, and 2.0 mg/kg/day. The study of Friedman et al. (1995) included dose levels of 0, 0.1, 0.5, or 2 mg/kg/day in males and 0, 1.0, or 3.0 mg/kg/day in females (Friedman et al. 1995). Both studies
identified a NOAEL of 0.5 mg/kg/day and a LOAEL of 2 mg/kg/day for histopathological evidence of degenerative nerve changes. The study of NTP (2011b) included dose levels of 0, 0.33, 0.66, 1.32, and 2.71 mg/kg/day in males and 0, 0.44, 0.88, 1.84, and 4.02 mg/kg/day in females; this study identified a NOAEL of 1.32 mg/kg/day and a LOAEL of 2.71 mg/kg/day for significantly increased incidences of axonal degeneration in the sciatic nerve of the male rats; the female rats exhibited a NOAEL of 1.84 mg/kg/day and a LOAEL of 4.02 mg/kg/day. All three chronic studies include sufficient incidence data for degenerative changes in peripheral nerves (detected by light microscopy) to warrant BMD analysis.

As discussed in detail in Appendix A, a PBPK rat model for acrylamide and glycidamide (Sweeney et al. 2010) was used to estimate for blood TWA acrylamide dose metric and TWA glycidamide dose metric for each of the administered acrylamide dose levels for male and female rats from each of the three chronic studies (Friedman et al. 1995; Johnson et al. 1986; NTP 2011b). All dichotomous models in the EPA Benchmark Dose Software (Version 2.1.2) were fit to the incidence data for degenerative peripheral nerve changes reported in each of the three chronic studies using PBPK-modeled rat blood TWA acrylamide as the dose metric and using PBPK-modeled rat blood TWA glycidamide as the dose metric; a benchmark response (BMR) of 10% extra risk was selected for the initial BMD modeling exercise. A BMR of 5% extra risk was also used for each model considered to provide the best fit to the male and female data from each of the three chronic studies, using TWA acrylamide as the dose metric and using TWA glycidamide as the dose metric.

A human PBPK model (Sweeney et al. 2010) was used to predict the HED corresponding to the BMDL\textsubscript{10} and BMDL\textsubscript{05} values for rat blood TWA acrylamide and TWA glycidamide from the best-fitting models for each of the three chronic studies. A BMR of 5% extra risk is justified because the chronic studies used sufficient numbers of animals (≥38 per dose group, with the exception of 20 animals in the 1.0 mg/kg/day dose group of female rats in the study of Friedman et al. 1995). The lowest PBPK model-predicted HED is 0.042 mg acrylamide/kg/day based on PBPK model-predicted blood TWA acrylamide for the male rats from the study of Friedman et al. (1995). The HED of 0.042 mg/kg/day was selected as the POD for deriving a chronic-duration oral MRL for acrylamide because it represents the most public health protective POD. A total uncertainty factor of 30 (3 for interspecies extrapolation using a PBPK model and 10 for human variability) was applied to the HED of 0.042 mg/kg/day, resulting in a chronic-duration oral MRL of 0.001 mg/kg/day.
The Fourth National Report on Human Exposure to Environmental Chemicals (CDC 2009) reported a geometric mean of 63.9 pmol acrylamide hemoglobin adduct/g hemoglobin with 50\textsuperscript{th}, and 95\textsuperscript{th} percentile values of 57.0, and 220 pmol acrylamide hemoglobin adduct/g hemoglobin, respectively for the U.S. population of males. The PBPK model-predicted acrylamide intakes associated with these acrylamide hemoglobin adduct levels are 0.003405, 0.00304, and 0.01173 mg/kg/day, respectively. A similar approach for the U.S. population of females results in PBPK model-predicted acrylamide intakes of 0.00313, 0.00285, and 0.00874 mg/kg/day, respectively. A PBPK model-predicted acrylamide hemoglobin adduct level of 787.917 pmol/g can be calculated using the HED of 42 µg/kg/day that serves as the point of departure for the chronic-duration oral MRL for acrylamide. At the chronic-duration oral MRL of 1 µg acrylamide/kg/day, the PBPK model-predicted acrylamide hemoglobin adduct level is 18.75 pmol/g hemoglobin. This value is slightly lower than the mean acrylamide hemoglobin adduct levels of 63.9 and 58.7 pmol/g hemoglobin reported for the U.S. population of males and females, respectively.