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2.1 BACKGROUND AND ENVIRONMENTAL EXPOSURES TO PHENOL IN THE UNITED STATES

Phenol is a naturally occurring and manufactured chemical that is widely distributed in the environment. It is found in various consumer products including throat lozenges, mouthwashes, and antiseptic lotions. The most likely route of exposure to phenol is through dermal contact either in the work environment or at home using ointments and other household products containing phenol.

Phenol is a product of combustion of coal wood and municipal solid waste; therefore, residents near coal and petroleum fueled facilities as well as residents near municipal waste incinerators may have increased exposure to phenol. Phenol is also a product of auto exhaust, and therefore, areas of high traffic likely contain increased levels of phenol. Recent data on concentrations of phenol in air are lacking; it was found at a median concentration of 30 parts per trillion (ppt) in 7 samples from one U.S. urban/suburban site in 1974 and at a median concentration of 5,000 ppt in 83 samples from seven sites between 1974 and 1978. The individual medians of the seven source sites ranged from 520 to 44,000 ppt. Higher phenol concentrations may occur when there is smog or in highly contaminated air.

Phenol has been detected in surface waters, rainwater, sediments, drinking water, groundwater, industrial effluents, urban runoff, and at hazardous waste sites. Levels of up to 1 ppb have been detected in unpolluted groundwater and concentrations ranging from 0.01 to 1 ppb were detected in unpolluted rivers. Phenol has been detected in Lake Huron water at 3–24 ppb and industrial rivers in the United States at 0–5 ppb.

Little information is available concerning the presence of phenol in soils. Phenol generally does not adhere very strongly to soils and tends to filter rapidly through soil, which may account for the lack of monitoring data, since any phenol released to soils is likely to move to groundwater.

Phenol is degraded rapidly in air (half-life of approximately 15 hours), but may persist in water for a somewhat longer period. In soil, phenol will biodegrade rapidly; the half-life in soil is generally <5 days.

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Although low levels of phenol have been detected in certain foods and tap water, these levels do not constitute major sources of exposure for most people. Phenol has been reported at concentrations of 7 and 28.6 ppm in smoked summer sausage and smoked pork belly, respectively, and was identified but not quantified in mountain cheese, fried bacon, fried chicken, and black fermented tea.

Since plants can metabolize phenol readily, exposure through eating food derived from plants grown in phenol-containing soil is probably minimal. Due to rapid biodegradation in water and soil, this contamination should be limited. People with contaminated tap water can be exposed from drinking the water or eating foods prepared with it. In addition, inhalation can occur during showering, bathing, and cooking with contaminated water. People can also be exposed to phenol through dermal contact due to bathing or showering with contaminated water.

There are no known unique sources of exposure to children. No reports of phenol in breast milk or baby foods were found. Children are likely to be exposed to phenol through inhalation of contaminated air from wood, coal, and waste incineration as well as from second-hand smoke. Nonsmokers who live with smokers may be exposed to 6–14 µg/day of phenol.

2.2 SUMMARY OF HEALTH EFFECTS

Information about the health effects of phenol in humans is derived from studies of workers and members of the general population following inhalation, oral, and dermal exposure. These studies indicate that phenol is an irritating and corrosive substance, making the skin and mucosal membranes targets of toxicity, but other effects have also been reported. However, the data for humans exposed to phenol by inhalation or ingestion are inadequate to establish concentration-response relationships, which are needed to identify adverse effects levels. Fatalities due to ingestion or contact with a significant area of the skin have been reported. A minimal lethal oral dose of approximately 70 mg/kg has been estimated in adults. Other estimates indicate that an oral dose as low as 1,000 mg could be fatal in humans, but patients occasionally survived doses as high as 65,000 mg. Postmortem examination typically showed serious mucosal alterations in the gastrointestinal tract. Other than the skin and mucosal membranes, the liver and cardiovascular system might be considered targets for phenol toxicity. An epidemiological study of workers found that exposure to phenol was associated with an increased incidence of cardiovascular disease. Electrocardiographic alterations have been reported following acute oral and dermal exposure to phenol, as well as vomiting and lethargy. Studies of populations whose drinking water was contaminated with phenol found increased incidences of nausea and diarrhea, but exposure to chlorophenols may have

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also occurred. Also, liver effects, as judged by increased serum activities of alanine aminotransferase (ALT) and aspartate amino transferase (AST), were reported in a case of prolonged inhalation exposure to phenol and in workers in an oil-refining plant, but exposure to other solvents could not be ruled out in the latter case. An increased incidence of headaches was reported among people who used drinking water contaminated with phenol and probably chlorophenols also.

There is only one modern study of inhalation exposure of animals to phenol. The rest of the inhalation database for phenol is outdated and not useful for risk assessment, although it serves to identify some targets for phenol toxicity. However, no single especially sensitive target emerged from these studies. Short-term (5 minutes) exposure of mice to phenol caused respiratory irritation, as judged by the animals' reflex reduction in respiratory rate; a lowest-observed-effect level (LOEL) was not defined; but the exposure concentration that reduced the respiratory rate by 50% was 166 ppm. In rats exposed nose-only intermittently to concentrations up to 25 ppm for 2 weeks, phenol caused no gross or microscopic alterations in major tissues and organs, including the nasal cavity, but some rats showed an increased incidence in a red nasal discharge possibly due to the irritating properties of phenol. Phenol caused pneumonia, necrosis of the myocardium, centrilobular degeneration, and necrosis of the liver and renal lesions in rabbits and guinea pigs, but not in rats, exposed whole-body intermittently to 26 ppm phenol for intermediate durations. Since a lower exposure concentration was not tested, the lowest-observed-adverse-effect level (LOAEL) for the various effects may be lower. In yet another study in rats, continuous whole-body exposure to 26 ppm phenol for 15 days caused signs of neurological impairment including muscle tremors, twitching, and gait disturbances during the first 3–5 days of exposure. At termination, serum transaminases were elevated suggesting liver damage, but no histological examination was conducted. Again, 26 ppm was the only exposure concentration tested; therefore, the LOAEL for liver and neurological effects may be lower. Neurological effects, including loss of coordination and tremors, were also observed in rats exposed to 234 ppm phenol for 8 hours. In summary, inhaled phenol can affect several organs and tissues and produce neurological effects, but few generalizations can be made from the available studies due to the different exposure protocols used (i.e., nose-only vs. whole-body; intermittent vs. continuous) and incomplete reporting. Toxicokinetics information indicates that phenol is readily absorbed through the skin of humans and animals, so that whole-body exposure may result in considerably more absorbed phenol than in nose-only exposures.

Application of phenol to the skin of animals has caused edema, erythema, necrosis, and death; the cause of death was not provided in the studies available. The effects of phenol on the skin are due to its property to impair the stratum corneum and produce coagulation necrosis by denaturing and precipitating

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proteins. Lethality is influenced by the surface area exposed as well as the concentration of the applied solution. Systemic effects also have been described in animals following dermal exposure to phenol. Rabbits that received a dose of phenol of 24 mg/cm²/kg suffered cardiac arrhythmia. Tremors leading to convulsions were reported in rats following application of 107 mg/kg of phenol to an unspecified surface area.

In contrast to the limited inhalation database, there is an extensive database of oral studies in animals; yet, it is not easy to characterize the toxicity of orally administered phenol. A key factor contributing to the inability to do so is that phenol administered by oral gavage is much more toxic than when it is administered in the drinking water, a phenomenon that is related to the toxicokinetics of phenol. Studies have shown that the toxicity of phenol is correlated with peak blood concentration rather than with total dose, such as the area under the blood concentration curve (AUC). Thus, end points that appear sensitive to phenol administered by oral gavage are not affected by the same total daily dose given via the drinking water.

Results from a 28-day drinking water study in mice provided the lowest effect levels in the oral database for phenol and suggested that hematological, neurochemical, and immunological end points may be particularly sensitive to phenol toxicity. However, since the effects reported in that study occurred at dose levels much lower than in any other study available, these findings should be interpreted with caution until supporting results are available. In this study, phenol induced a significant decrease in red blood cell counts in mice at ≥ 1.8 mg/kg/day. While this finding could have been due to macrocytosis, the study did not provide enough information to evaluate this possibility. Only two additional studies provided information on hematological parameters after phenol exposure. In pregnant mice, a single gavage dose of 265 mg/kg of phenol (only dose tested) on gestation day (GD) 13 induced a reduction in the ratio of poly/normochromatic erythrocytes in the bone marrow, whereas phenol administered to rats in doses of up to 320 mg/kg/day in the drinking water for 10 weeks did not significantly affect a comprehensive number of hematological parameters monitored. Other long-term drinking water studies in rats and mice did not evaluate hematological parameters.

The neurochemical effects caused by phenol also occurred at ≥ 1.8 mg/kg/day and consisted of alterations in the levels of neurotransmitters in various brain areas. This is difficult to interpret in light of the absence of clinical signs and lack of supporting evidence from other studies. Phenol, however, did induce neurological effects in other studies. Short-term oral gavage administration of doses ≥ 120 mg/kg/day of phenol caused muscle twitching and tremors in rats and mice, but no effects were observed at

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<40 mg/kg/day. Decreased motor activity was reported in female rats dosed with 360 mg/kg/day of phenol in the drinking water for 13 weeks, but no effects were seen at 107 mg/kg/day. No neurochemical evaluations were conducted in these studies. Other long-term drinking water studies in rats and mice that used much higher doses of phenol did not observe significant changes in gross or microscopic appearance of the brain of the animals, but no neurological tests or neurochemical evaluations were conducted in these studies.

Phenol caused a significant decrease in antibody response to immunization with sheep red blood cells (SRBC) in mice at ≥ 6.2 mg/kg/day in a drinking water study. Lymphoproliferative responses to T and B cell mitogens were also significantly suppressed at 33.6 mg/kg/day. A study that also conducted the plaque-forming cell assay to SRBC (but not the antibody titer) and evaluated lymphoreticular organs of rats exposed to up to 321 mg/kg/day of phenol in the drinking water for 10 weeks found no significant alterations. In another study, a single dose of 224 mg/kg of phenol administered to rats by oral gavage caused necrosis or atrophy of the spleen or thymus, but no other immunological end point was evaluated. Long-term drinking water studies in rats and mice did not report any significant gross and histological alteration in lymphoreticular organs and tissues at phenol doses >1,000 mg/kg/day.

Other effects of phenol observed in oral studies include renal tubular necrosis in rats treated with a single gavage dose of 224 mg/kg or with 40 mg/kg/day for 14 days. However, long-term drinking water studies in rats and mice that received much higher doses of phenol do not suggest that the kidney is a particularly sensitive target for phenol. Phenol also induced decreases in body weight in rats and mice in 13-week and 2-year drinking water studies that were associated with significant reductions in water consumption due probably to poor palatability. Phenol reduced body weight gain in pregnant mice treated by oral gavage with 280 mg/kg/day, a dose level that also caused frank neurotoxicity. Doses of 120 mg/kg/day of phenol administered to pregnant rats during GDs 6–15 using a divided dosing protocol to minimize the adverse effects of a bolus dose caused a significant reduction in weight gain in the dams; the no-observed-adverse-effect level (NOAEL) was 60 mg/kg/day. The latter findings suggest that weight gain during pregnancy is a sensitive end point for phenol toxicity and the dose of 120 mg/kg/day was the lowest LOAEL in acute-duration oral studies in which no overt signs of toxicity were observed following administration of phenol.

Phenol has induced developmental effects in rodents, but, with one exception, it appears that this occurs at dose levels that also affect the mothers. Doses of 120 mg/kg/day of phenol on GD 6–15 produced a 7% decrease in average fetal body weight in the absence of maternal effects in a study. In the study that used

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the divided dosing protocol mentioned above, there were no developmental effects at 120 mg/kg/day, but decreased ossification sites were seen at the highest dose level, 360 mg/kg/day. In a two-generation reproductive study in which the parental generation received doses of up to 301–321 mg/kg/day of phenol via the drinking water, decreased pup weight and percent live pups on postnatal day 4 was reported at a dose level that also significantly decreased maternal water consumption, including during gestation and lactation. In pregnant mice, doses of 280 mg/kg/day on GDs 6–15 produced a significant decrease in fetal weight and also caused tremors and ataxia in the dams.

A study of phenol-exposed wood industry workers reported a small, nonsignificant excess of respiratory cancers and a study of phenol production workers reported a small, non-significant excess of Hodgkin's disease and of lung, esophageal, rectal, and kidney cancers. However, the interpretation of these findings is complicated due to lack of dose-response and potential for confounding. Phenol has been tested for carcinogenicity in long-term drinking water bioassays in rats and mice. Statistically significant increased incidences of pheochromocytomas of the adrenal gland and leukemia or lymphomas were observed in male rats exposed to the low dose of phenol, but not to the high dose of phenol. No significant effects were seen in female rats or in mice. Phenol has consistently been found to be a promoter in initiation-promotion studies in mouse skin. Based on inadequate evidence in humans and in animals, EPA assigned phenol to Group D, not classifiable as to human carcinogenicity. Under updated guidelines, the data regarding carcinogenicity of phenol are: "inadequate for an assessment of human carcinogenic potential."

2.3 MINIMAL RISK LEVELS (MRLs)

Estimates of exposure levels posing minimal risk to humans (MRLs) have been made for phenol. 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 1994a), 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

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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

- An MRL of 0.02 ppm has been derived for acute-duration inhalation exposure (14 days or less) to phenol.

The acute-duration inhalation database for phenol is very limited. It includes a few animal studies of limited scope (Aranyi et al. 1986; De Ceaurriz et al. 1981; Flickinger 1976) and a well-conducted study that used modern methodology to evaluate a number of relevant end points (Hoffman et al. 2001). No relevant human studies were located. In the animal studies, a target for phenol toxicity was not clearly defined; however, for an irritant substance such as phenol, it is reasonable to assume that portals of entry, such as the respiratory tract, could be potential targets. Of the studies mentioned above, only Hoffman et al. (2001) conducted a careful evaluation of the respiratory tract. Hoffman et al. (2001) exposed rats to various exposure levels for 2 weeks and evaluated a number of end points including histopathology, hematology, and clinical chemistry and reported no adverse effects. De Ceaurriz et al. (1981) exposed mice to various concentrations of phenol in air for 5 minutes and determined an RD₅₀ (concentration that reduced the respiratory rate by 50%, a protective reflex response in rodents) of 166 ppm. Aranyi et al. (1986) also exposed mice to 5 ppm phenol 3 hours/day for 5 days and reported no significant changes in susceptibility to airborne bacterial agents relative to mice exposed to filtered air. Flickinger (1976) observed loss of coordination and tremors in rats exposed to 234 ppm phenol for 8 hours; a 1-hour exposure was without effect. No other exposure concentration was tested and no control group was used. Fourteen days later, the rats were sacrificed and subjected to gross necropsy. Flickinger (1976) indicated that no gross lesions were observed, but the scope of the examination was not specified. Of all the studies available, the one conducted by Hoffman et al. (2001) is the most complete, better-reported, and used modern methodology and was used as principal study for MRL derivation.

In the principal study (Hoffman et al. 2001), groups of Fischer 344 rats (20/sex/group) were exposed nose-only to phenol vapors in concentrations of 0, 0.5, 5, or 25 ppm 6 hours/day, 5 days/week for 2 weeks. Rats were observed daily for morbidity and mortality and for adverse neurobehavioral signs. At termination, 10 rats/sex/group were used for clinical chemistry and hematology evaluations and for histopathological examinations of nasopharyngeal tissues, larynx, trachea, lungs with mainstem bronchi,

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kidney, liver, and spleen. The remaining 10 rats per group were kept for a 2-week recovery period, after which time they were sacrificed. There were no chemical-related deaths. The only clinical signs were observations of a red nasal discharge during the 2 weeks of exposure. However, the investigators stated that there was no clear pattern and that the signs had almost disappeared after the 2-week recovery period. It should be noted that an unpublished version of the study (CMA 1998) showed that the incidence of the red nasal discharge was concentration-related in males during the second week of exposure. Pairwise comparison with controls shows a statistically significant increased incidence at 5 and 25 ppm in males. However, there is evidence that a tear-like nasal discharge in rats can be a generalized response to stress from a variety of causes. Porphyrins in the discharge lead to a red color. A red nasal discharge also was noticed in rats in a two-generation drinking water study in all groups, including controls, although the incidence was higher in phenol-treated rats (Ryan et al. 2001). The fact that the incidences were concentration-related in the Hoffman et al. (2001) study suggests that the effect is likely related to phenol, possibly to the irritating properties of phenol. Yet, in the absence of nasal histopathology, the nasal discharge is not considered an adverse effect. There were no significant alterations in body weight gain during the study. The only significant hematology change was an increase in prothrombin time in 0.5 ppm females at the recovery sacrifice. The only significant clinical chemistry change was an increase in serum albumin in 25 ppm females at the recovery sacrifice. Significant changes in organ weights were limited to a decrease in relative liver and spleen weight in 5 ppm females at terminal sacrifice and a decrease in spleen to brain weight in the same group of females. Gross and microscopic examinations of the tissues were unremarkable. The study NOAEL is 25 ppm; no LOAEL was defined.

Several possibilities exist regarding derivation of an acute-duration inhalation MRL for phenol. One approach could be not to derive an acute-duration inhalation MRL based on the limited database available and on the fact that the highest exposure concentration in the Hoffman et al. (2001) study is a free-standing NOAEL and, therefore, the true NOAEL may be much higher. Although the use of such a NOAEL for MRL derivation may yield an overly conservative MRL, deriving an MRL is preferred over not doing so since MRLs serve as screening levels for health assessors to identify contaminants and health effects of concern at hazardous waste sites. Secondly, if the Hoffman et al. (2001) study is chosen as the basis for an acute-duration MRL, the issue is to decide what the critical end point should be because the highest exposure concentration was a NOAEL. The choices are the respiratory tract or a systemic extrarespiratory end point (i.e., hepatic, renal). Given that phenol is a recognized respiratory irritant, the respiratory tract is chosen as critical target, and, within the respiratory tract, the nasal region is selected, as it is likely to be the first region of contact and the most sensitive. Finally, the NOAEL/LOAEL approach is used for MRL derivation rather than the benchmark dose (BMD) analysis

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because studies in which responses are at the same level as background or at or near the maximal response level are inadequate for benchmark dose analysis (EPA 2000c). No physiologically based pharmacokinetic (PBPK) models that have been validated in animals and humans are available for phenol.

The acute-duration inhalation MRL was calculated using EPA's methodology (EPA 1994a) for a category 1 gas, as detailed in Appendix A. The study NOAEL of 25 ppm was adjusted for intermittent exposure yielding a $NOAEL_{[ADJ]}$ of 4.5 ppm. The basic equation is:

$$NOAEL_{[HEC]} = NOAEL_{[ADJ]} \times RGDR_{ET} \text{ where}$$

$$NOAEL_{[ADJ]} = 25 \text{ ppm} \times 6/24 \text{ hours} \times 5/7 \text{ days} = 4.5 \text{ ppm and}$$

$RGDR_{ET}$ = ratio of the regional gas dose in rats to that of humans for the extrathoracic region

$$RGDR_{ET} = (VE/SA_{ET})_A / (VE/SA_{ET})_H \text{ where}$$

VE = minute volume (0.137 L/minute for rats, 13.8 L/minute for humans [EPA 1994a]) and

SA_{ET} = surface area of the extrathoracic region (15 cm² for rats and 200 cm² for humans [EPA 1994a])

$$NOAEL_{[HEC]} = 4.5 \text{ ppm} \times (0.137 \text{ L/minute}/15 \text{ cm}^2) / (13.8 \text{ L/minute}/200 \text{ cm}^2) = 0.6 \text{ ppm}$$

Applying an uncertainty factor of 30 (3 for extrapolation from animals to humans with dosimetric adjustment and 10 for human variability) to the $NOAEL_{[HEC]}$ yields an acute-duration inhalation MRL of 0.02 ppm for phenol.

For the purpose of comparison, basing the MRL on a systemic end point rather than the respiratory tract would have yielded a slightly higher acute-duration inhalation MRL. Using EPA's methodology for a category 3 gas, the $NOAEL_{[HEC]}$ would be calculated by multiplying the $NOAEL_{[ADJ]}$ of 4.5 ppm by 1 (since no data were located on the blood/air partition coefficient for phenol in rats or in humans a default value of 1 is used for this term). Applying an uncertainty factor of 100 (10 for animal to human extrapolation and 10 for human variability) to the $NOAEL_{[HEC]}$ would result in an acute-inhalation MRL of 0.04 ppm for phenol.

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An intermediate-duration inhalation MRL was not derived for phenol due to lack of adequate data. No relevant human data were located and the available animal studies had numerous limitations including poor control of exposure levels, unclear scope of the evaluations, and limited reporting. The intermediate-duration database consists of only three studies (Dalín and Kristoffersson 1974; Deichmann et al. 1944; U.S. Air Force 1961). Dalín and Kristoffersson (1974) exposed a small number of rats to 0 or 26 ppm phenol continuously for 15 days and reported mild motor disorders (impaired balance, abnormal gait, muscle twitching) during the first few days of exposure. At termination, the activities of serum transaminases were significantly increased indicating liver damage, but no histopathology examination was conducted. Because the exposure chamber was not of modern design, there is some uncertainty as to the actual exposure levels. Deichmann et al. (1944) exposed guinea pigs, rats, and rabbits intermittently for 6–12 weeks to a concentration of phenol in air that apparently could not be controlled with any precision, but could have ranged from 26 to 52 ppm. No controls were used and no actual data were presented; the paper contains only a narrative of the results. Exposure to phenol caused serious histological alterations in the lungs, heart, liver, and kidneys in rabbits and guinea pigs, but no significant changes were reported in rats. U.S. Air Force (1961) exposed monkeys, rats, and mice continuously to 0 or 5 ppm phenol for 90 days. No information was provided regarding the frequency of monitoring the test atmosphere, but the concentration of phenol was reported to remain in the range of 4.5–5.5 ppm after the first few days of the experiment. Although the report indicates that there were no significant histological alterations in organs and tissues, incomplete reporting of the results suggests that there may have been some lung, liver, and kidney pathology. In addition, no data were presented to support the assertion that there were no effects on hematology (three species), blood chemistry (monkeys only), urinalysis (three species), and kidney function tests (monkeys and rats).

A chronic-duration inhalation MRL for phenol was not derived due to lack of data for this duration. Occupational studies in humans are limited by lack of exposure data and simultaneous exposure to multiple chemicals. No chronic inhalation study in animals was located.

Oral MRLs

No reliable human data were located for derivation of oral MRLs for phenol. As mentioned in Section 2.2, effects of phenol administered to animals by oral gavage are different than those observed in drinking water studies. Administration of phenol by oral gavage, as was done in almost all acute-duration oral studies, results in adverse effect levels that are much lower than those identified in drinking water

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studies. For example, tremors were reported in rats administered a single gavage dose of 120 mg/kg (Berman et al. 1995) and in pregnant mice administered 140 mg/kg/day during gestation (NTP 1983b), but no adverse neurological signs were reported in rats administered 360 mg/kg/day in the drinking water for 13 weeks (Beyrouy 1998) or in rats or mice administered phenol in the drinking water in doses exceeding 700 mg/kg/day for up to 103 weeks (NCI 1980). This differential toxicity is related to the toxicokinetics of phenol. A study by Hiser et al. (1994) showed that the toxicity of phenol is correlated with peak blood concentration rather than with total dose, such as the AUC. Hiser et al. (1994) observed that rats given phenol by oral gavage developed a cluster of behaviors that the investigators termed “phenol twitching behavior” consisting of tremors, sudden jerks, hyper-reactivity to stimulus, and excessive blinking, none of which occurred in groups dosed via the drinking water. Hiser et al. (1994) also noticed that the twitching behavior developed almost immediately after gavage dosing, a time that also coincided with peak blood levels of phenol, and disappeared by 37 minutes after dosing. Also, for a given daily dose, peak levels of phenol in blood were much higher following gavage dosing than following continuous administration in the drinking water. Additional information that supports the idea of toxicity being associated with peak blood levels of phenol was provided by experiments done by NTP (1983a). These investigators treated pregnant rats with phenol by gavage in different volumes during GDs 6–15. In a group dosed with 125 mg/kg/day in a volume of 1 mL/kg, 7 of 10 rats died. Deaths were preceded by dose-related signs of toxicity, including tremors, convulsions, and respiratory distress, and necropsy revealed mottled liver and congested lungs. However, in a group treated with 160 mg/kg/day in a volume of 5 mL/kg only one of six rats died, as a larger dosing volume would be expected to decrease the absorption rate. Based on the information discussed in this paragraph, only drinking water studies and divided dose gavage studies will be considered appropriate for MRL derivation.

- An MRL of 0.6 mg/kg/day has been derived for acute-duration oral exposure (1–14 days) to phenol.

All of the acute-duration oral studies available administered phenol to the animals by gavage. As indicated in the preceding paragraph, dosing volume in oral gavage studies is important in the manifestation of phenol toxicity. Acute-duration studies that used a relatively low dosing volume of 1 mL/kg are those by Berman et al. (1995), Moser et al. (1995), and Narotsky and Kavlock (1995). These studies were not considered for MRL derivation even though they identified adverse effects at dose levels lower than studies that used divided gavage dosing or drinking water studies. The remaining database is essentially limited to two developmental studies, which were considered for MRL derivation. In one of these studies, rats were gavaged with phenol in doses of up to 120 mg/kg/day in a dosing volume of 5 mL/kg during GDs 6–15 (NTP 1983a). There was no maternal toxicity, but mean fetal body weight at

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this dose level was approximately 7% lower than controls. However, since historical control data showed that the concurrent control fetal weight for the CD rat was much higher (22%) than the historical control weight and a larger litter size in the high-dose group may have contributed to the smaller fetal weight in the high-dose group, the dose of 120 mg/kg/day can be considered an equivocal LOAEL for developmental effects; the NOAEL was 60 mg/kg/day. In the other developmental study, which used a divided dosing protocol and a dosing volume of 10 mL/kg, there was a dose-related decrease in maternal body weight gain during treatment days and beyond, which achieved statistical significance at 120 mg/kg/day (York 1997). Maternal body weight was also reduced, but differences with control achieved statistical significance only at 360 mg/kg/day (see details below). No fetal toxicity was seen at 120 mg/kg/day. The decrease in maternal body weight gain during gestation was the most sensitive end point and the dose level of 120 mg/kg/day is considered a LOAEL; the NOAEL is 60 mg/kg/day. Since the York (1997) study identified the most sensitive end point and utilized a dosing protocol that resembles more closely a potential environmental exposure scenario to phenol, it was selected as the principal study for the derivation of an acute-duration oral MRL for phenol.

In the York (1997) study, groups of pregnant Sprague-Dawley rats (25/dose group) were dosed 3 times daily with 0, 20, 40, or 120 mg phenol/kg (total daily doses of 0, 60, 120, or 360 mg/kg) by gavage in water on GDs 6–15; the dosing volume was 10 mL/kg. Maternal end points evaluated included clinical signs, body weight, and food consumption. Dams were also observed for abortions and premature deliveries. Dams were sacrificed on GD 20 and a gross necropsy was conducted. The uterus was examined for pregnancy, number and distribution of implantations, live and dead fetuses, and early and late resorptions. Fetuses were weighed and examined for sex and gross external alterations. Half of the fetuses were examined for soft tissue alterations and the remaining fetuses were examined for skeletal alterations. One dam in the 360 mg/kg/day group died on GD 11 and the death was attributed to phenol treatment. Clinical signs considered treatment-related included excess salivation and tachypnea in rats exposed to 360 mg/kg/day. Gross necropsy of the dams did not reveal any treatment-related alterations. In the 120 mg/kg/day group, maternal body weight gain was significantly reduced for GDs 6–16 (11%) and for GDs 12–16 (19%), whereas in the 360 mg/kg/day group, body weight gain was reduced 38% for GDs 6–16. Maternal final body weight in the 360 mg/kg/day group was reduced, but <10% relative to controls. Food consumption was reduced in the 360 mg/kg/day group by 16% for GDs 6–20 and by 15% for GDs 0–20; in the 120 mg/kg/day group, food consumption for GDs 6–16 was reduced 11%. Fetal body weight at the 360 mg/kg/day level was reduced 5–7% relative to controls. There was a significant decrease in ossification sites on the hindlimb metatarsals in the 360 mg/kg/day group, which was considered of minimal biological significance. At the 120 and 360 mg/kg/day dose levels, there were

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increases in litters with fetuses with "any alteration" and with "any variation", but neither reached statistical significance and there were no clear dose-response relationships. There were no significant effects on corpora lutea, implantations, litter sizes, live fetuses, early and late resorptions, or percent resorbed conceptuses. Based on decreased fetal body weight and delayed ossification, the dose of 360 mg/kg/day is a LOAEL for developmental effects; the NOAEL is 120 mg/kg/day. Based on decreased weight gain during gestation, the dose of 120 mg/kg/day is a LOAEL for decreased maternal body weight gain; the NOAEL is 60 mg/kg/day. The MRL of 0.6 mg/kg/day was derived by applying an uncertainty factor of 100 (10 for animal to human extrapolation and 10 for human variability) to the maternal NOAEL of 60 mg/kg/day.

Data from York (1997) also were analyzed using the BMD approach for MRL derivation. BMD models in the EPA Benchmark Dose Software (BMDS version 1.3.2) (linear, polynomial, power, and Hill models) were fit to the maternal body weight gain data to determine potential points of departure for the MRL. The linear model with homogeneous variance, which is the simplest model and the model that provided the best fit for the data, was selected. In the absence of a clear criteria as to what level of change in weight gain during pregnancy should be considered adverse, the benchmark response (BMR) was defined as a change in mean body weight gain equal to one standard deviation from the control mean (EPA 2000c). The corresponding BMD was 152 mg/kg/day; the corresponding benchmark dose limit (BMDL) was 125 mg/kg/day. Applying an uncertainty factor of 100 (10 for animal to human extrapolation and 10 for human variability) to the BMDL results in an acute-duration oral MRL of 1 mg/kg/day. The MRL of 0.6 mg/kg/day derived using the NOAEL/LOAEL approach is preferred because it is more health protective than the MRL derived using the BMD methodology.

An intermediate-duration oral MRL for phenol was not derived. Several studies are available that provide information on the effects of phenol following intermediate-duration exposure and all of them used drinking water to administer the test material. With the exception of one study (Hsieh et al. 1992, see below), doses tested in intermediate-duration oral studies were higher than doses tested in acute-duration oral studies. A 13-week drinking water study in rats and mice evaluated clinical signs and gross and microscopic appearance of a number of organs and tissues and found little evidence of toxicity (NCI 1980). Reduction in body weight gain was observed in both rats and mice at the highest dose levels tested (1,556 mg/kg/day in rats, 2,468 mg/kg/day in mice), which was most likely due to significant decreases in water consumption. Also available is a two-generation reproduction study that found no evidence of reproductive effects in male and female rats (301 and 321 mg/kg/day, respectively), but reported decreased pup weight and reduced viability at 301/321 mg/kg/day (Ryan et al. 2001).

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Significantly reduced water consumption was also reported in the Ryan et al. (2001) study, particularly in the 301/321 mg/kg/day males and females. A specialized 13-week neurotoxicity study in rats reported decreased motor activity in females dosed with 360 mg/kg/day, but not with 107 mg/kg/day (Beyrouly 1998). However, the most significant findings among the intermediate-duration database were reported in a 28-day study in mice (Hsieh et al. 1992). These investigators found hematological and neurochemical effects in mice at 1.8 mg/kg/day and immunological effects at ≥ 6.2 mg/kg/day. Hsieh et al. (1992) dosed CD-1 mice (five per dose group) with phenol in the drinking water for 28 days. At termination, there was a dose-related decrease in red cell counts, statistically significant at all dose levels. The hematocrit was decreased only at the highest dose level. In the absence of a change in hematocrit, the decrease in red blood cells may have been due to macrocytosis, but the study did not provide sufficient information to evaluate this possibility. Ryan et al. (2001), in a two-generation study, conducted a comprehensive evaluation of hematological parameters in rats exposed to up to 321 mg/kg/day for 10 weeks and found no significant alterations. Similarly, in the inhalation experiments of U.S. Air Force (1961) in mice exposed continuously for 90 days, no hematological alterations were observed. Hsieh et al. (1992) also reported significant dose-related alterations in various neurotransmitters in the brain (i.e., dopamine, norepinephrine). In the absence of clinical effects, this is difficult to interpret. Beyrouly (1998) exposed rats to doses of 308 mg/kg/day of phenol for 13 weeks and found no significant neurological alterations, although neurochemical evaluations were not conducted. Hsieh et al. (1992) also found a significant decrease in antibody response to immunization with SRBCs at ≥ 6.2 mg/kg/day, detected by two different assays (plaque-forming cell assay and antibody titer). At the highest dose level tested, 33.6 mg/kg/day, lymphoproliferative responses to T and B cell mitogens were also significantly suppressed. Ryan et al. (2001) also conducted the plaque-forming cell assay to SRBC (but not the antibody titer) and evaluated lymphoreticular organs of rats in the two-generation study and found no significant alterations, although it is not uncommon to find differences in immune responses between rats and mice. The Hsieh et al. (1992) study was not used for derivation of an intermediate-duration oral MRL largely due to the unconfirmed nature of findings observed at relatively very low doses and because only five mice comprised each dose group. NTP is currently conducting a comprehensive series of tests to evaluate the potential immunotoxicity of phenol in mice. As reported in EPA (2002), preliminary results demonstrated immunosuppression (reduced antibody response), confirming the results of Hsieh et al. (1992). ATSDR will review the NTP report when it becomes available and will then decide on a course of action regarding an intermediate-duration MRL.

A chronic-duration oral MRL for phenol was not derived. The only chronic-duration animal studies are the NCI (1980) 103-week studies in rats and mice. NCI (1980) evaluated clinical signs, organ weights,

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and gross and microscopic appearance of organs and tissues. The lowest doses tested were 322 mg/kg/day in rats and 590 mg/kg/day in mice. Under the conditions of the study, phenol showed essentially no systemic toxicity, but neither hematology nor clinical chemistry tests were conducted. The only reported effect was a significant decrease in body weight in male (≥ 322 mg/kg/day) and female (≥ 721 mg/kg/day) rats associated with significant decreases in water intake; food consumption was comparable among all groups. It would not be appropriate to use the LOAEL of 322 mg/kg/day as the basis for an MRL since the effect (reduced final body weight) was likely due to decreased water intake. An additional reason for not deriving a chronic-duration oral MRL for phenol is the intermediate data from Hsieh et al. (1992) suggesting that immunosuppression may be the most sensitive effect, which leaves open the possibility that it could do the same in longer-term studies. Tests of immunocompetence were not conducted in the standard 2-year bioassays available.