ADDENDUM TO THE TOXICOLOGICAL PROFILE FOR DINITROCRESOLS

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CONTENTS

ADDENDUM for Dinitrocresols

Supplement to the 1995 Toxicological Profile for Dinitrocresols

Background Statement

This addendum to the Toxicological Profile for Dinitrocresols supplements the profile that was released in 1995.

Toxicological profiles are developed in response to the Superfund Amendments and Reauthorization Act (SARA) of 1986, which amended the Comprehensive Environmental Response, Compensation, and Liability Act of 1980 (CERCLA or Superfund). CERCLA mandates that the Administrator of ATSDR prepare toxicological profiles on substances on the CERCLA Priority List of Hazardous Substances and that the profiles be revised "no less often than once every three years". CERCLA further states that the Administrator will "establish and maintain inventory of literature, research, and studies on the health effects of toxic substances" [Title 42, Chapter 103, Subchapter I, § 9604 (i)(1)(B)].

The purpose of this addendum is to provide to the public and other federal, state, and local agencies a non-peer reviewed supplement of the scientific data that were published in the open peer-reviewed literature since the release of the profile in 1995.

Chapter numbers in this addendum coincide with those of the Toxicological Profile for Dinitrocresols (1995). This document should be used in conjunction with the profile. It does not replace it.

2. HEALTH EFFECTS

2.2 DISCUSSION OF HEALTH EFFECTS BY ROUTE OF EXPOSURE

2.2.2 Oral Exposure

2.2.2.5 Reproductive Effects

Takahashi et al. (2004) showed that exposure of sexually mature male rats to 4,6-dinitro-o-cresol (DNOC) resulted in reduced sperm motility and increased incidence of tailless sperm in the cauda epididymides. In a more recent study designed to elucidate the pathogenesis of tailless sperm, male rats were given daily oral doses of 0, 10, or 15 milligrams/kilogram DNOC for 5 days. Sperm were examined on days 1, 7, and 14 after the last dosing. On day 1, post-dosing peeled (loss of mitochondrial sheath at the proximal end of the mid shaft) sperm were observed in the caput epididymides. On day 7, the highest incidence of peeled sperm was found in the corpus epididymides. On day 14, the highest incidence of abnormal sperm was in the cauda epididymides, where the primary abnormality was taillessness. The authors concluded that exposure of the rats to DNOC causes partial loss of the mitochondrial sheath in the testicular elongated spermatids, resulting in tailless sperm in the cauda epididymides by day 14.

2.2.2.7 GENOTOXICITY

The mutagenicity of DNOC was tested in a number of strains of *Salmonella typhimurium*. DNOC was not mutagenic when tested in the presence or absence of metabolic activation. In addition, little or no effect on sister-chromatid exchanges (SCE) in human peripheral blood lymphocytes treated with DNOC was observed either in the presence or absence of metabolic activation. In contrast, DNOC showed dose-related toxic effects in both the presence and

absence of S9 mix in assays of unscheduled DNA synthesis (UDS activity) in human peripheral blood lymphocytes. In a study of male and female rats treated *in vivo* with DNOC, gender differences were noted. Dose-related increases in the frequency of chromosome aberrations and aberrant cells were observed in bone marrow cells of male rats treated. Female rats showed a statistically significant increase over controls in chromatid breaks at the middle dose tested, but with a lesser magnitude than male rats (Hrelia et al. 1994).

2.3 TOXICOKINETICS

2.3.3 Metabolism

DNOC induces apoptosis-like cell death in soybeans. The effects of DNOC on soybean suspensions included activation of capsase-3-like proteins and release of cytochrome c from mitochondria, confirming the apoptotic-like phenotype. The authors suggest that plants and animals may share similar pathways related to programmed cell death (Aranha et al. 2007).

4. PRODUCTION, IMPORT/EXPORT, USE, AND DISPOSAL

4.3 USE

DNOC has been used in Central and Southern Africa for the control of locusts (Byaruhanga 1999). DNOC was tested for activity against *Mycobacterium tuberculosis*. It was found to be comparable or superior to ethambutol, which is used in combination with other drugs for the prevention and treatment of *Mycobacterium avium* complex and drug-resistant tuberculosis (Garcia-Garcia et al. 2005).

5. POTENTIAL FOR HUMAN EXPOSURE

5.3 ENVIRONMENTAL FATE

5.3.2 Transformation and Degradation

The bacterium *Sphinomonas sp* UG30 is a pentachlorophenol-degrading strain that can degrade several nitrophenolic compounds, including DNOC (Cassidy et al. 1999).

The kinetics of degradation of seven nitrophenols, including DNOC, by hydrogen peroxide photolysis, the Fenton reagent, and photo-Fenton, were studied. Of the three methods, treatment with the Fenton reagent was found to be the cheapest and most effective. Use of the *Daphnia magna* acute toxicity test showed that the Fenton reagent led to complete detoxification of the nitrophenols (Goi and Trapido 2002).

5.3.2.3 Sediment and Soil

In a study of the specific adhesion of nitroaromatic pesticides to clay minerals, Haderlein et al. (1995) found a high adsorption coefficient for DNOC to clay when the ionizable cation in the clay is K+. A mixed culture of microorganisms isolated from soil contaminated with pesticides and from activated sludge were used to study the aerobic biodegradation of DNOC in batch culture and in fixed-bed column reactors. Between 65 and 84% of DNOC nitrogen was released into the medium as nitrate, whereas 61% of ^{14}C-labeled DNOC was recovered as $^{14}CO_2$ (Gisi et al. 1997). In a column experiment using aquifer material and about 25 micrograms/liter DNOC and aerobic conditions, it was found that the DNOC was significantly retarded by sorption. After a lag period of 80 days, the DNOC degraded quickly with 0 order rate constant of 1.3–2.6 micrograms/L/day (Tuxen et al. 2000). The observation of a long lag period for the degradation

of DNOC was also found by Broholm et al. (2001) in a study of the sorption and degradation of DNOC under aerobic conditions in a sandy aquifer in Vejen, Denmark. The authors noted significant and spatially variable sorption of DNOC (Kd range 0.10–0.98 liter/kilogram) due to specific binding of DNOC to clay materials. The spatial variation was mainly due to a pH effect, with stronger sorption at lower pH, whereas the effect of cation composition on the solid matrix appeared to have a negligible effect.

Sorption of DNOC to three different smectites was studied by use of FTIR, HPLC, and quantum chemical methods. DNOC has a high affinity to smectite due to site-specific interactions with exchangeable cations and non-specific van der Waals interactions with the siloxane surface. The FTIR-derived sorption isotherm of DNOC sorbed to K-SWy-2 smectite was in good agreement with the isotherm derived from HPLC measurements. The molar absorptivity value of DNOC sorbed to K-SWy-2 was 1.43 x 107 +/- 0.3 centimeters^2/mol. With the FTIR method, the limit of detection of DNOC was ~5 microgram DNOC/gram (clay). The authors concluded that the sorption isotherms and the molar absorptivity provide a direct link between the macroscopic sorption results and the FTIR spectra (Johnston et al. 2002).

5.4 LEVELS MONITORED OR ESTIMATED IN THE ENVIRONMENT

Although the use of DNOC was cancelled in the United States in 1987 and in European countries between 1999 and 2004, it is still found in the environment because of its slow environmental degradation. In addition, there are still stocks of this chemical available, mainly in underdeveloped countries. Consequently, this chemical is included in studies that assess the presence of pesticides and herbicides in the environment.

5.4.1 Air

Nitrophenol formation within the atmosphere was studied at a remote sampling site with low direct impact from rural areas. The levels of phenol and four nitrophenols, including DNOC, were measured in the gas and liquid phases of clouds at the summit of Great Dun Fell over a 20-day period. The authors concluded that mononitrophenols are derived mainly from car exhaust, while dinitrophenols are formed from secondary nitration within the atmosphere (Luttke et al. 1997). They used data from five cloud events at the Great Dun Fell to measure the partition coefficients of phenol and four nitrophenols. The observed partition coefficient for DNOC was found to be consistent with the assumption of an equilibrium between the gas and liquid phases, as described by Henry's law.

Analysis of air samples from three locations in France, namely Strasburg (urban area), Schiltigheim (suburban area), and Erstein (rural area), was undertaken to assess the special and geographic variations of concentrations and the effect of traffic emissions on the level of phenols and nitrophenols, including DNOC. Samples were collected during the four seasons and at various times over a 24-hour period, and they were analyzed by GC/MSD. Partitioning of phenolic compounds between gas and particle phases appeared to be correlated with vapor pressure (Morville et al. 2006).

5.4.2 Water

Over a three-year period in Bavaria, Germany, rainwater was collected at eight locations and analyzed monthly for the level of nitrophenols. The levels of DNOC varied from near zero to about 140 micrograms/month*m^2 (Schussler and Nitschke 2001). Over an 18-month period,

rainwater was collected monthly at two stations in Denmark and analyzed for 79 compounds, including DNOC. DNOC was detected in 93% of the samples at one site and in 100% at the other; the levels ranged from “not detected” to 1053 nanograms/liter (median 464.5 nanograms/liter) and 149 to 1038 nanograms/liter (median 546 nanogram/liter), respectively (Asman et al. 2005).

6. ANALYTICAL METHODS

6.2 ENVIRONMENTAL SAMPLES

Uzer et al. (2006) developed two distinct spectrophotometric methods to assay for DNOC in soil samples. The authors propose that these methods are cost-effective tools which can be used to screen for DNOC in field studies prior to applying higher cost methodologies. Another study compared soil organic matter (SOM) and K-saturated reference smectite clay (SWy-2) as substrates for the adsorption of pesticides. DNOC adsorbed better to SWy-2 and better than the other pesticides tested. The conclusion was that expandable soil clays have the potential to be an equal or dominant sorptive phase compared to SOM for certain pesticides, including DNOC (Sheng et al. 2001). Ohfuji et al. (1997) developed a gas chromatographic method for the determination of DNOC in citrus fruits by methylation by using trimethylsilyldiazomethane. The detection limit was 0.005 microgram/gram. The recovery of DNOC in citrus fruit was between 73 and 85%. Methods for the detection of DNOC include liquid chromatography with electrochemical detection (Galeano-Diaz et al. 2000); solid phase extraction-spectrophotometry (Uzer et al. 2006); atmospheric pressure chemical ionization mass spectrometry-mass spectrometry with methanol and acetonitrile as the organic modifiers (Geerdink et al. 1999); gas chromatography-mass spectrometry (GC-MS) with negative ion chemical ionization (Nakamura et al. 2001); solid phase microextraction and GC-MS (Jaber et al. 2007); capillary high

performance liquid chromatography with uv detection (San Andres et al. 2000); capillary electrophoresis and capillary electrochromatography (Sovocool et al. 1999); pulsed amperometric detection with poly(dimethylsiloxane)-fabricated capillary electrophoresis microchips (Ding and Garcia 2006); and high flow, high resolution ion mobility spectrometry (Wu et al. 2002).

7. REGULATIONS AND ADVISORIES

The National Institute for Occupation Safety and Health's (NIOSH) recommended exposure limits (REL) and the Occupational Safety and Health Administration's (OSHA) permissible exposure limits (PEL) for DNOC is a time-weighted average 0.2 milligram/meter^3 (skin) [NIOSH Pocket Guide 2009]. The TLV 8-hour time weighted average (TWA) is 0.2 milligram/cubic meter (skin) [ACGIH 2008].

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