MICHIGAN DEPARTMENT OF ENVIRONMENTAL QUALITY

INTEROFFICE COMMUNICATION

September 27, 2002

TO: File for tributyl phosphate (126-73-8)

FROM: Marco Bianchi

SUBJECT: Initial Threshold Screening Level

The initial threshold screening level (ITSL) for tributyl phosphate (TBP) is 22$\mu g/m^3$ based on an 8-hour averaging time.

The following references or databases were searched to identify data to determine the ITSL: IRIS-online, HEAST, NTP Management Status Report-online, RTECS, EPB-CCD, EPB library, CAS-online, NLM-online, IARC-online, NIOSH Pocket Guide, and ACGIH Guide.

Tributyl phosphate is a colorless, odorless liquid that is used as a plasticizer for cellulose esters, lacquers, plastics (especially vinyl resins), and as an antifoaming agent. Acute toxicity studies suggest that the chicken is the least sensitive species to TBP, rats and mice being more sensitive. Its oral administration to rats produces muscular weakness, dyspnea, coma and pulmonary edema, whereas its intraperitoneal administration causes paralysis in mice. Production workers have found this compound to be an irritant to the respiratory system and mucous membranes.

The single oral LD_{50} in rats for TBP is 3 g/kg, ranging from 1390 to 3000 mg/kg. The oral LD_{50} for mice ranges from 400 to 1240 mg/kg. Intraperitoneal LD_{50} values for TBP in rats and mice range from 800 to 1600 mg/kg and 100 to 200 mg/kg, respectively. The dermal LD_{50} value for TBP in the rabbit was greater than 3100 mg/kg. A single topical application of 500 mg TBP to intact or abraded skin of six rabbits produced severe irritation.

In an acute and subchronic neurotoxicity study, male and female adult Sprague-Dawley rats received TBP in corn oil by gavage in acute (single-dose) and subchronic (three-month) studies. Dosage levels in the acute study were 100, 325, and 1000 mg/kg; whereas, dose levels of 32.5, 100, and 325 mg/kg/day were administered in the subchronic study. Behavioral evaluations were performed in both studies, and neuropathological evaluations were performed in the three-month study only. Mean body weight decreases were statistically significant compared to controls for male rats at the high-dosage level only in the acute study. Transient changes only, attributable to the general toxicity of the material, were noted in forelimb grip strength and mean activity level during the first 24 hours after dosing for 1000 mg/kg rats. In the subchronic study, high-dose males and females had statistically significant body weight decreases; some mortality also was observed at this dosage level. The motor activity levels and qualitative and quantitative functional observational battery measurements were comparable between treatment and control groups, and there were no gross or neurohistopathological findings in the rats indicative of treatment-related effects. Based on these study results, TBP was not neurotoxic to rats following either acute or subchronic exposures.

Laham et al. (1983) gavage-dosed Sprague-Dawley rats (10/sex/group) with either 0.28 ml/kg or 0.42 ml/kg daily, for 14 days. Control rats were gavage-dosed tap water at 0.42 ml/kg. A significant reduction in conduction velocity of the caudal nerve was observed in high dose male rats. Electron microscopic examination of sciatic nerve showed morphological changes such as retraction of Schwann cell processes surrounding unmyelinated fibers in both sexes of high dose groups. In another short-term toxicity test, Laham et al. (1984) reported the results of a short-term toxicity study in which Sprague-Dawley rats were administered TBP by gavage at doses of 0.14 and 0.42 ml/kg for 14 days. No overt signs of toxicity were observed throughout the study. There were no significant differences in body weight between the test groups and their respective controls, but absolute and relative liver weights were significantly increased in the high-dose group (both-sexes). Histopathogical examination revealed a low incidence of degenerative changes in the seminiferous tubules of the high-dose group. Finally, in an 18-week follow-up study, Laham et al. (1985) administered TBP by gavage once a day (5 days/week) to Sprague-Dawley rats (12 rats/sex/group). Low-dose animals received 200 mg/kg/day throughout the study. High-dose animals received 300 mg/kg/day for the first 6 weeks and 350 mg/kg/day for the remaining 12 weeks. Histopathological examination of tissues revealed that all treated rats developed diffuse hyperplasia of the urinary bladder epithelium. Similar changes were not found in the control animals. No testicular changes were observed in the high-dose rats.

In a study carried out to determine the delayed neurotoxicity potential of TBP, this compound was administered to groups of 20 adult hens at test doses equivalent to its LD_{50} of 1500 mg/kg. After the administration of the first dose, a waiting period of 21 days was allowed before administering the second dose to allow for the organophosphate induced delayed neuropathy (OPIDN). The results showed no delayed neurotoxicity in the hen since neither neurologic deficits nor histopathological changes characteristic of OPIDN was seen. In contrast, hens treated with a single oral dose of 750 mg/kg body weight of tri-o-cresyl phosphate, a known OPIDN, developed delayed neuropathy.

In a two-generation reproductive toxicity study, TBP was tested for reproductive toxicity in rats. Thirty weanlings/sex (F0) were exposed to TBP in the diet ad libitum at 0, 200, 700, or 3000 for 10 weeks and then randomly mated within groups for 3 weeks with continued exposure. F0 parents and 10 F1 weanlings/sex/dose were necropsied and adult reproductive organs, urinary bladders (both sexes), kidneys (males), and livers (females) were evaluated histologically. Thirty F1 weanlings/sex/dose continued exposure for 11 weeks and were bred as described above. F1 parents and F2 weanlings, 10/sex/dose, were then necropsied as described above. Adult toxicity was observed in both sexes and generations at 700 and 3000 ppm; observations included reduced body weights, weight gain and feed consumption, urinary bladder epithelial hyperplasia (both sexes), renal pelvis epithelial hyperplasia only at 3000 ppm (male kidneys), and centrilobular hypertrophy (female livers). At 200 ppm, transient reductions in body weight were observed in F0 and F1 females, with urinary bladder epithelial hyperplasia in F0 males and females and in F1 males. There was no evidence of reproductive toxicity, of reproductive organ pathology, or of effects on gestation or lactation at any dose tested. Postnatal toxicity was evidenced by consistent reductions in F1 and F2 pup body weights at 3000 ppm and by occasional weight reductions in F2 litters at 700 ppm, and was associated with maternal toxicity observed at these doses and times. Under the conditions of this study, a NOAEL was not determined for adult toxicity; the NOAEL for reproductive toxicity was at least 3000 ppm and the NOAEL for postnatal toxicity was approximately 200 ppm.

In a teratological study, TBP was administered orally to pregnant Wistar rats on days 7-17 of gestation at 0, 100, 200, 400, 800 mg/kg/day in a dose-finding study. Oral doses of 0, 62.5,

125, 250, or 500 mg/kg/day were administered in a subsequent teratological study. Caesarean sections were performed on day 20 of gestation. In the dose-finding study, all of the pregnant rats were killed by the treatment with TBP at 800 mg/kg/day. In the teratological study, salivation and depression of body weight gain, adjusted body weight gain and food consumption were observed at the higher doses of TBP. There were no significant differences between the groups in the incidence of dead or resorbed fetuses, the number of living fetuses and the body weights of living fetuses of both sexes. The incidence of rudimentary lumbar rib increased significantly at 500 mg/kg/day. There were two cases of malformation; a fetus with deformity of fore-and hind-limbs at 400 mg/kg/day in the dose-finding study and conjoined twins exhibiting three fore-limbs and four hind-limbs at 125 mg/kg/day in the teratological study. These malformations were rare in the background data of teratology, and the incidence of fetuses with malformations was not increased significantly. According to the study investigators, TBP was considered not to be teratogenic in this study.

In a dietary oncogenicity study TBP was administered in the diet at concentrations of 0, 200, 700 and 3000 ppm to groups of 50 male and 50 female Sprague-Dawley rats for two years. Body weights and food consumption were measured weekly for the first 13 weeks and monthly thereafter. Hematology was performed at 12, 18 and 24 months; urinalyses were performed at 3 weeks and at 3, 6, 12 and 18 months. All surviving animals were euthanized after 24 months of treatment. Macroscopic postmortem examinations were performed on all animals; complete histopathological evaluation was performed on control and high dose animals; target organs were examined in all dose groups. Significant decreases in body weight gain occurred in males and females receiving the 3000 ppm concentration and a slight decrease in weight gain occurred in females receiving the 700 ppm concentration. The only clinical sign attributed to TBP was an increased incidence of red discoloration of the urine in some high-dose males. Survival, hematology and urinalysis parameters were unaffected by treatment at any concentration. A dose-related increase in the incidence and severity of urinary bladder hyperplasia and the incidence of urinary bladder papillomas was evident in male and female rats receiving the 700 and 3000 ppm concentrations. Transitional cell carcinomas were present in six of 49 males and two of 50 females, and a squamous cell carcinoma was present in one of 49 males in the group that received 3000 ppm. The oncogenic effects showed a clear threshold of 700 ppm in the diet. The NOEL (no observable effect level) for chronic toxicity was 200 ppm. Mean intake of TBP was 9 and 12 mg/kg/day for males and females, respectively, receiving 200 ppm; 33 and 42 mg/kg/day for males and females, respectively, receiving 700 ppm, and 143 and 182 mg/kg/day for males and females, respectively, receiving 3000 ppm. TBP was negative in genotoxicity tests, suggesting that the tumors are induced by nongenotoxic mechanisms.

In a companion dietary oncogenicity study, TBP was administered in the diet at concentrations of 0, 150, 1000 or 3500 ppm to groups of 50 male and 50 female CD-1 mice for 18 months. Survival, clinical signs and hematology parameters were unaffected by treatment at any concentration. Initial weight losses and significant decreases in body weight gain occurred in males and females receiving the high dose (3500 ppm) of TBP in diet. A significant dose-related increase in absolute and relative liver weights was seen in male and female mice that received the two highest dietary concentrations of TBP (1000 and 3500 ppm). The incidence of hepatocellular adenomas was significantly increased in male mice treated with 3500 ppm TBP in diet. No other tumors were associated with TBP administration in this study. The NOEL for chronic toxicity was 150 ppm, or 28.9 mg/kg/day for females and 24.1 mg/kg/day for males. Although rats treated chronically with TBP have exhibited urinary bladder hyperplasia and urinary bladder papillomas and transitional cell carcinomas, no urinary bladder alterations attributed to TBP administration occurred in this study.

The American Conference of Governmental Industrial Hygienist (ACGIH) documentation stated that workers exposed to 15 mg/m^3 of TBP complained of nausea and headache. TBP was shown to be an irritant to human skin, mucous membranes, eye, and the respiratory tract. *In vitro* studies on isolated human skin showed that TBP possessed a high capacity for percutaneous penetration. The ACGIH documentation concluded that because little toxicity data is available to assign a TLV to TBP, they will use a similar chemical to do so. They state that *the LD_{50} of TBP is about the same as that for triphenyl phosphate; but, contrary to triphenyl phosphate, TBP irritates skin and is a narcotic. TBP also is a weak cholinesterase inhibitor as compared with the congener triphenyl phosphate. Given the absence of epidemiologic data on TBP, the same TLV for both TBP and triphenyl phosphate seems justified and a TLV of 0.2 ppm is considerably less than the concentrations reportedly associated with worker complaints of nausea and headache.* The ACGIH set a Threshold Limit Value (TLV) for TBP at 0.2 ppm or (2.2 mg/m^3).

Toxicity testing for TBP has resulted in varied outcomes depending upon species, duration of exposure and target organ. Acute testing produced a wide range of oral LD_{50}s; from 1,390-3,000 mg/kg in rats, to 400-1,490 mg/kg in mice. Some short-term studies showed that TBP depresses body weight. However, other short-term studies showed no depression of body weight but histological evidence of degenerative changes in the seminiferous tubules. Further short-term studies indicated no changes in seminiferous tubules, but diffuse hyperplasia of the urinary bladder epithelium. In mutagenicity studies, equivocal results have been obtained in the Ames test in the presence or absence of metabolic activation. However, E. coli tests, Salmonella microsome tests, and recessive lethal mutation tests in Drosophila melanogaster all indicate that TBP is non-mutagenic. TBP produces only mild plasma cholinesterase depression in rats. Short-term exposure resulted in the depression of caudal nerve conduction velocity and equivocal morphological changes in the Schwann cells of peripheral nerves. Chickens dosed with high levels of TBP showed no evidence of ataxia or nerve and brain histopathology. These data demonstrate that TBP does not produce delayed neuropathy in the chicken.

Chronic toxicity testing by dietary administration has shown that the target organ for the rat is the urinary bladder, while for the mouse it is the liver. For rats, a dose-related increase in the incidence and severity of urinary bladder hyperplasia and the incidence of urinary bladder papillomas was evident in male and females receiving 700 and 3000 ppm concentrations. Transitional cell carcinomas were present in males and females receiving 3000 ppm. According to the study investigators, the oncogenic effects of this compound appeared to show a clear threshold of 700 ppm in the diet; with incidence rates of papillomas/ carcinomas of 0, 0, 2, 30 for male rats dosed at 0, 200, 700, or 3,000 ppm. Likewise, in mice a significant dose-related increase in absolute and relative liver weights was seen in male and female mice that received the two highest dietary concentrations of TBP (1000 and 3500 ppm), but not at the 150 ppm dose. The incidence of hepatocellular adenomas was significantly increased in male mice treated with 3500 ppm, but no other tumors were associated with TBP administration in the study. A two-generation rat study revealed similar results, with a NOAEL for reproductive toxicity of 3000 ppm and a NOAEL for postnatal toxicity at 200 ppm. Finally, a teratogenicity study found a NOAEL for maternal toxicity at >700 ppm and fetal toxicity at >3000 ppm.

According to the Air Toxics Rules, in the absence of EPA RfCs, RfDs, and cancer slope factors, an ITSL or an IRSL is typically derived from human or lifetime animal studies; unless there is evidence to show that using an occupational exposure limit would better fit an inhalation route of exposure or provide a greater margin of safety. After reviewing data for TBP, it was determined that a TLV-based ITSL may be more appropriate for this particular compound. The TLV for TBP of 2.2 mg/m^3 is based on the similar congener, triphenyl phosphate. The TLV is set to prevent

eye, respiratory, and skin irritation from worker exposures. In comparison, oral and dietary rat studies showed the urinary bladder to be the target organ; with sub-chronic studies causing urinary bladder hyperplasia leading to urinary bladder carcinoma in the 2-year chronic dietary studies. Mice, on the other hand, developed hepatocellular adenomas after a 2-year TBP dietary study. Nothing in the literature suggested an association between inhalation exposure and urinary bladder carcinogenicity or hepatocellular adenomas. Another aspect of TBP toxicity is the threshold potential of rat urinary bladder carcinogenicity. Urinary bladder carcinogenicity was only found in groups of rats given a dietary concentration of greater than 700 ppm (30-50 mg/kg/day) after two years of administration. Because this effect was found in only one test specie and at a threshold concentration, the study investigators concluded that it appears to be specific to the rat and should not pose an increased risk to humans. However, as a comparison, if an IRSL were to be derived from the rat or mouse dietary study using the Global82 linear multistage model assuming no threshold, the resultant one-in-a-million risk would equal 0.5-2 ug/m^3 annual averaging for rats. An ITSL based on the TLV and adjusted to an annual averaging time would equal 2-3 ug/m^3. This value is very similar to the calculated IRSL and would be lower than the SRSL. Likewise, if an RfD were to be derived from the dietary study (accounting for dose adjustments and uncertainty factors), it would equal 90 ug/m^3 annual averaging. Again, an ITSL based on the TLV and adjusted to an annual averaging time would equal 2-3 ug/m^3. Therefore, it seems appropriate to use the TLV to derive an ITSL for TBP.

The ITSL was determined as follows:

ACGIH TLV = 2.2 mg/m^3

2.2 mg/m^3 ÷ 100 = 0.022 mg/m^3

$$0.022 \text{ mg/m}^3 \times \frac{1000 \text{ ug/m}^3}{1 \text{ mg/m}^3} = 22 \text{ ug/m}^3$$

The ITSL for tributyl phosphate = 22 ug/m^3 based on 8 hr. averaging.

References:

1. Documentation of Threshold Limit Values and Biological Exposure Indices. 1992. Tributyl phosphate. American Conference of Governmental Industrial Hygienists (ACGIH), 6th Edition.

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