DEVELOPMENT OF HEALTH CRITERIA FOR SCHOOL SITE RISK ASSESSMENT PURSUANT TO HEALTH AND SAFETY CODE SECTION 901(g):

Dieldrin Evaluation

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Integrated Risk Assessment Branch
Office of Environmental Health Hazard Assessment
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Introduction

Health and Safety Code (HSC), Section 901(g), requires the Office of Environmental Health Hazard Assessment (OEHHA), in consultation with the appropriate entities within the California Environmental Protection Agency, to identify those chemical contaminants commonly found at school sites and determined by OEHHA to be of greatest concern based on child-specific physiological sensitivities. HSC 901(g) also requires OEHHA to annually evaluate and publish, as appropriate, numerical health guidance values (HGVs) for five of those chemical contaminants until the contaminants identified have been exhausted. HGVs established by this mandate are intended for use in the assessment of health risks at proposed or existing California school sites. At this time, OEHHA is focusing its evaluation on non-cancer effects of the identified chemicals, pending the completion of a new method for developing HGVs based on child-specific carcinogenic effects. Accordingly, current HGVs are in the form of a child-specific reference dose (chRD) or child-specific reference concentration (chRC).

The Introduction serves as a background for the technical evaluation of dieldrin. For those that are not familiar with this OEHHA program, it is advisable to review this chapter prior to analyzing the following technical report.

The technical chapter is a focused document that summarizes the evaluation of this chemical and discusses the appropriateness of developing a chRD. Recent reviews of the chemical by various entities, such as the U.S. Environmental Protection Agency (U.S. EPA), Agency for Toxic substances and Disease Registry (ATSDR), and/or California Department of Pesticide Regulation (CDPR), serve as a baseline for OEHHA to conduct additional literature searches. In the document, OEHHA identifies relevant information from the baseline and from literature searches for discussion. OEHHA will not reiterate basic data on environmental fate, pharmacokinetics, and pharmacodynamics that have been adequately covered in the cited baseline documents. Because the objective of the evaluation is to determine the appropriateness of establishing a chRD, which would then be applied for assessing health risk from oral or dermal exposure, non-cancer studies using an oral route of administration and studies that provide information regarding age-sensitivity are the primary focus of the OEHHA review.

It should be underscored that a chemical-specific risk assessment is not required to support the development of chRDs. The purpose of establishing these child-specific health criteria is to provide improved means for consultants of school districts or the Department of Toxic Substances Control (DTSC) to conduct school site-specific risk assessment. The process here is similar to that used by U.S. EPA in developing reference doses (RfDs) for superfund site risk assessment. Thus, OEHHA is not considering exposure issues here. They will be dealt with in the site-specific risk assessment, specifically in the exposure assessment portion which can be found in the "Guidance for Assessing Exposures and Health Risks at Existing and Proposed School Sites Pursuant to Health and Safety Code §901(f), February 2004." The appropriate chRDs will be applied

only if site-specific sampling and analysis indicate the occurrence of the corresponding chemicals. The consultants will have the option to use, for example, default dermal or oral bioavailability factors provided in the exposure assessment guidelines, or proposed a departure from the default based on supporting data.

Developing a chRD or chRC

Challenge

The use of appropriate HGVs and exposure parameters is essential to provide an unbiased assessment of potential health risks at an existing or a proposed school site. Since children have higher air, food and water intake relative to their body weight compared to adults; and have activity or behavioral patterns that may lead to higher exposure to environmental contaminants than adults, these higher intakes and unique activity patterns need to be considered in developing a set of child-specific exposure parameters for use in the risk assessment. OEHHA has analyzed these exposure parameters in issuing the report, Guidance for Assessing Exposures and Health Risks at Existing and Proposed School Sites (OEHHA, 2004).

With respect to evaluating non-cancer risk by comparing the potential chemical exposure against the corresponding health criteria in the school setting, HGVs in the form of child-specific reference doses or concentrations should be used. Until the inception of the HSC 901(g) program, these child-specific HGVs were not available. Instead, existing reference doses or concentrations for non-cancer endpoints, which were based on adult human or animal data, were mostly used. The Food Quality Protection Act of 1996 (http://www.epa.gov/opppsps1/fqpa/) was an attempt to address the issue of child sensitivity. In addition to the traditional interspecies and intra-species uncertainty factors, it mandated a safety factor of 10 for the protection of children unless data existed to indicate that children were not more sensitive than adults. Thus, a question has been raised that the intra-species uncertainty factor of 10 would not adequately protect children because it was mainly designed to account for genetic variability such as metabolizing isoenzyme variations.

A case can be made for the development and application of child-specific HGVs based on studies in young animals or epidemiological analysis of pertinent data rather than relying solely on a safety factor or uncertainty factor. While locating the appropriate data are a challenge, OEHHA has strived to do so because children can be more (or less) susceptible to chemical effects due to pharmacodynamic and pharmacokinetic differences between them and adults, and thus empirical data in the young would be preferable. U.S. EPA and the March of Dimes sponsored a workshop -- Identifying Critical Windows of Exposure for Children's Health -- in September 1999 to systematically review the state of knowledge on prenatal and postnatal exposures and subsequent outcomes (Selevan *et al.* 2000). The workshop focused on the nervous, immune, respiratory, reproductive, and endocrine systems—organ systems that are still undergoing development and maturation in children and thus deemed to be highly vulnerable to chemical insults. Workshop participants noted that data pertaining to children's sensitivities to environmental contaminants during various critical developmental periods are limited. In particular,

little attention has been given to studying peripubertal/adolescent exposures or adult consequences from childhood exposure. Thus, the state of scientific knowledge pertaining to chemical effects on children is and will continue to be a limiting factor in OEHHA's ability to develop child-specific HGVs for these contaminants.

The evaluation of empirical data in the young can be a complex task. Vulnerability of the young often depends on the organ system in question and its developmental stage. There are critical periods of structural and functional development during both prenatal and postnatal life, including adolescence. During its critical period(s), a particular structure or function is most sensitive to disruption due to interactions between a toxicant and target tissues that are undergoing biochemical changes. Damage may not be evident until a later stage of development (DeRosa et al., 1998; Bigsby et al, 1999). The brain, for example, is an organ with distinct neurodevelopmental stages that occur in temporally distinct time frames across different regions, so the specific chemical, dose, and time of exposure during development determine if a specific function in the brain will be altered (Faustman et al, 2000).

Differences also exist between children and adults with respect to their absorption, distribution, metabolism, and elimination of chemical contaminants. For example, absorption may be different in neonates because of the immaturity of their gastrointestinal tract and their larger skin surface area in proportion to body weight (Morselli et al. 1980; NRC, 1993); the gastrointestinal absorption of lead is greatest in infants and young children (Ziegler et al. 1978). Distribution of xenobiotics may be different; for example, infants have a larger proportion of their bodies as extracellular water, and their brains and livers are proportionately larger (Altman PL, 1974; Fomon, 1966; Fomon *et al*. 1982; Owen G.M., 1966; Widdowson E.M., 1964). The infant also has an immature blood-brain barrier (Adinolfi, 1985) (Johanson, 1980) and probably an immature blood-testis barrier (Setchell B.P., 1975). Many xenobiotic metabolizing enzymes have distinctive developmental patterns. At various stages of growth and development, levels of particular enzymes may be higher or lower than those of adults, and sometimes unique enzymes may exist at particular developmental stages (Komori *et al*. 1990; Leeder and Kearns, 1997; NRC, 1993; Vieira *et al*. 1996). Whether differences in xenobiotic metabolism make the child more or less susceptible also depends on whether the relevant enzymes are involved in activation of the parent compound to its toxic form or in detoxification. There may also be differences in excretion, particularly in newborns, who all have a low glomerular filtration rate and have not developed efficient tubular secretion and resorption capacities (Altman PL, 1974; NRC, 1993; West J.R., 1948). Children and adults may differ in their capacity to repair damage from chemical insults.

OEHHA faces an additional challenge when evaluating chemicals that are potential endocrine disruptors. The topic of endocrine disruption during development has been the subject of much scientific and regulatory debate (Colborn *et al*. 1993a; Colborn *et al*. 1993b; Cranmer *et al*. 1984; US EPA, 1998). While not all chemicals selected for the OEHHA review are endocrine disruptors, the endocrine disruptors do pose a greater concern because not only could they directly impact the maturation and proper functioning of the endocrine system, they could also interfere with hormonal signal

transduction that leads to abnormal growth and functioning of other target organs (e.g., immune and nervous systems) in school children. Exposure to endocrine disruptors during critical "programming" periods in development, in contrast to exposure during adulthood, may produce irreversible effects on the reproductive, nervous, and/or immune systems (Bigsby *et al*. 1999). In adulthood, these endocrine disruptors might only produce reversible effects by participating in the "seesaw" process of stimulation and feedback inhibition.

Given the complexity of hormone signaling processes, it is also not surprising to find the evaluation of the dose and response relationship to be another challenge. The shape of the dose response curve may not be linear, but rather shaped like an upright U or an inverted U (Markowski *et al*. 2001; vom Saal *et al.* 1997). This makes data interpretation difficult when the study does not include sufficient treatment doses to span the entire range of interest.

In summary, the use of a study in children or young animals as the basis for a child-specific HGV is preferred. In cases when epidemiological studies involving an adult population, or studies involving adult animals, are used, the challenge is to integrate other experimental studies that suggest a greater sensitivity in the young with adult studies to justify the application of appropriate safety factors.

Process

In June 2002, OEHHA issued a report, "Development of Health Criteria for School Site Risk Assessment Pursuant to Health and Safety Code, Section 901(g): Identification of Potential Chemical Contaminants of Concern at California School Sites," documenting the process by which OEHHA identifies chemicals and presenting a compilation of 78 chemicals (OEHHA, 2002). The compilation, whose sole purpose is to provide OEHHA staff with a manageable list of chemicals to work from, has no regulatory status and is a living document – chemicals may be added or removed as new information becomes available.

The chRD development process begins with the prioritization of chemicals from the compilation described in the June 2002 report. OEHHA has employed the following criteria, recognizing that often the availability of health effect data may be the overriding consideration in the selection of chemicals for evaluation.

1. Chemicals having a strong indication of their presence at school sites according to monitoring studies or other reliable sources.

2. Chemicals cited to have possible adverse effects in three or more of the systems that are undergoing critical development during childhood: the nervous, immune, respiratory, reproductive, or endocrine systems.

3. Chemicals that other OEHHA programs have identified as a concern.

From a public health protection standpoint, the OEHHA scientists working on health guidance values for children as mandated by Health & Safety Code 901(g) have adopted the following procedures in evaluating and developing chRDs or chRCs. First, in order to protect children from infancy through the time they leave school, chRDs must consider school-aged children up to age 18, and infants and toddlers in daycare facilities located at school sites. Second, OEHHA opts to consider the most sensitive species and endpoints in our evaluations. When evaluating various studies that use different test parameters to measure the same endpoint such as the nervous system, the lowest LOAEL (lowest observed adverse effect level) or NOAEL (no observed adverse effect level) from these studies would be selected. Third, the paucity of data has underscored the reality that the databases for sensitive endpoints may be incomplete. An uncertainty factor for database deficiency will be considered when there is sufficient information to strongly suggest child-specific sensitivity but insufficient quantitative data from young animal studies to permit the use of these data. Fourth, quantifying differences in susceptibility between a developing organ system and a mature one are hampered by the availability of studies that intentionally compare an effect in young animals with one in adult animals. Available data are mainly from developmental toxicity studies that limit dosing to the mother during pregnancy. OEHHA staff have deemed that these studies can be used for development of a child-specific health guidance value (chRD or chRC) if it is reasonable to assume that the effect of the chemical on the target organ in the offspring animal would likely occur on the same target organ undergoing development after birth in humans. If studies that include gestational dosing of the mother and lactational dosing of the pups (a protocol of the U.S. EPA Developmental Neurotoxicity Health Effects Test) are available, OEHHA will also consider these studies acceptable for establishing a chRD or chRC if the development of the critical organ system continues to occur during childhood.

Finally, these prenatal and perinatal studies are frequently part of a series of studies to elucidate a "mechanism of toxicity." These studies may not have used a large number of animals or dose ranges. However, due to the critical windows in which cell proliferation and differentiation are occurring in specific organ systems during childhood, a study in young animals is usually preferred over one in adults, even adult humans. With corroborating studies showing a mechanism of action and biological plausibility, OEHHA will consider using these studies as appropriate. However, data from adult animals may be used, if they are from high quality studies and if there are data to provide a means of inference to vulnerability of development in young animals so that an appropriate uncertainty or safety factor can be applied.

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Dieldrin

Summary

OEHHA has identified dieldrin as a contaminant of concern pursuant to Health and Safety Code Section 901(g). In an updated review of available literature, OEHHA has found additional information that exposure to dieldrin during the childhood neurological developmental period could irreversibly impact the system of nerve cells that use dopamine as its neurotransmitter. Progressive adverse effects on the nigrostriatal dopamine system from early-life exposure might contribute to an early onset of Parkinson's disease. While this developmental neurotoxicity may be a very sensitive endpoint, available data do not permit a determination of the lowest dose for this effect. Accordingly, OEHHA is not proposing a child-specific reference dose (chRD) for dieldrin. Instead, OEHHA recommends the use of the U.S. Environmental Protection Agency (EPA) reference dose (RfD), or the Agency for Toxic Substances and Disease Registry (ATSDR) minimal risk level (MRL), both of which have a value of 5×10^{-5} mg/kg-day, for assessing the non-cancer risk of dieldrin at school sites.

What is dieldrin?

Dieldrin was used extensively as an insecticide on crops such as corn and cotton from the 1950s until 1970 (ATSDR, 2002). The U.S. Department of Agriculture canceled all uses of dieldrin, as well as aldrin (a structurally similar pesticide) in 1970. In 1972, however, U.S. EPA approved aldrin and dieldrin for killing termites. Use of aldrin and dieldrin to control termites continued until 1987 when the manufacturer voluntarily canceled the registration for use in controlling termites.

Besides being manufactured directly, dieldrin can be derived from aldrin. In the environment, aldrin is readily oxidized into dieldrin. Likewise, aldrin is metabolized into dieldrin once it enters the body.

What information indicates dieldrin is of concern pursuant to Health & Safety Code Section 901 (g)?

OEHHA has identified dieldrin as a contaminant of concern pursuant to HSC 901(g) (OEHHA 2002). Dieldrin persists in the environment because it is resistant to biotransformation and abiotic degradation. Being lipophilic, dieldrin also bioconcentrates and biomagnifies through the terrestrial and aquatic food chains. From reviewing school site risk assessments through 2004, the Department of Toxic Substances

Control noted that dieldrin had been found in almost 10 percent of the school sites in California (S. Fair, 2004, personal communication). This highlights the environmental persistence of dieldrin, as it has not been used since 1987.

ATSDR reviewed the health effects of dieldrin (ATSDR, 2002). Due to its high lipophilicity, dieldrin has been detected in breast milk (Polishuk et al., 1977); and shown to cross the blood-brain barrier and remain in brain tissues (ATSDR, 2002). People exposed to large amounts of dieldrin experienced convulsions, some had kidney damage and some died. Exposure to moderate levels of dieldrin led to headaches, dizziness, irritability, vomiting, or uncontrollable muscle movements. Some sensitive people seemed to have developed an autoimmunity in which dieldrin caused the body to destroy its own blood cells. Results from animal studies showed that dieldrin caused similar effects on the nervous system and on the kidneys to those seen in people. Additional effects on the liver and immune system were also observed in animal studies.

The nervous system is a primary target organ of dieldrin. Dieldrin causes hyperexcitation of the central nervous system and generalized seizures (convulsions). It was believed that the hyperexcitatory effects was a result of a generalized activation of synaptic activities (Joy, 1982). However, the role of dieldrin in blocking inhibitory activity within the brain has received a great deal of attention as the probable mechanism underlying the central nervous system excitation. Based on good correlations of effects from the molecular level to whole animal toxicity, the preponderance of evidence indicates that the convulsing and other excitatory effects of dieldrin are a consequence of the blocking action on the $GABA_A$ receptor-chloride channel complex (Ikeda et al., 1998; Liu et al., 1997a; Liu et al., 1997b; Narahashi et al., 1998). The investigation into the effect of dieldrin on $GABA_A$ receptor subunit mRNA expression reported that dieldrin increased β3 subunit transcripts by 300 percent and decreased γ2S and γ2L transcripts by 50 and 40 percent, respectively (Liu, Morrow et al. 1997b). This molecular study suggests that dieldrin could pose a risk to the brain by altering gene expression and the GABAergic circuitry.

There is increasing evidence that dieldrin could also affect the dopaminergic system, which in turn could trigger a neurodegenerative process that results in the manifestation of Parkinson's disease (Di Monte et al., 2002; Kanthasamy et al., 2005). The mechanism of action is not completely understood. Dieldrin could act directly on the dopaminergic system or the dopaminergic effect could be mediated by the effect on GABA because γ2 subunits of the $GABA_A$ receptor are highly expressed in the dopaminergic neurons of the substantia nigra pars compacta (Okada et al., 2004). Fleming was one of the pioneers that demonstrated a significant correlation between dieldrin and Parkinson's disease (Fleming et al., 1994). Dieldrin was detected in six of 20 brains from Parkinson's disease patients and in none of the 14 age-matched control brains. Another study demonstrated a significantly higher concentration of dieldrin in the caudate nucleus of the striatum obtained post-mortem from Parkinsonian patients as compared to controls (0.515 μg/g versus 0.283 μg/g lipid) (Corrigan et al., 1998). This latter information suggests that dieldrin may preferentially target the nigrostriatal system. In reviewing data pertaining to the effect of dieldrin on dopamine and its transporters in pre-synaptic terminals of

nigrostriatal neurons, on oxidative stress and mitochondrial dysfunction, and on caspases activities, Kanthasamy has advanced a model that describes dieldrin-induced apoptosis in dopamingeric neurons of the substantia nigra pars compacta (Kanthasamy et al., 2005). The hallmark of Parkinson's disease is progressive and selective dopamingeric neuron loss in the substantia nigra. After more than 50 percent of neuronal loss in the subtantia nigra and 75 percent depletion of striatal dopamine content, patients start to exhibit the clinical symptoms, including resting tremor, bradykinesia, rigidity, and postural instability (Steece-Collier et al., 2002). While age is an indisputable risk factor for the disease (1.5 million elderly individuals in U.S. or approximately 2% of the population over the age of 50, and the prevalence increases to 5% by the age of 85 (Kanthasamy et al., 2005)), exposure of children to dieldrin could initiate the neurodegenerative process at a relatively young age and exacerbate the loss of dopamingeric neurons in the substantia nigra pars compacta, causing an early onset of Parkinson's disease.

More recent data, which were not available at the time of ATSDR's review, suggest that children may differ from adults in their susceptibility to health effects from dieldrin exposure. Richardson et al. reported that perinatal exposure to dieldrin, did not produce overt toxicity to the dams or offspring at the doses tested, but nevertheless resulted in alterations of the dopaminergic system, which in turn increased its susceptibility to the parkinsonism-inducing neurotoxin MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) administered later in life (Richardson et al., 2006). This study not only epitomizes the concept of critical windows in developmental exposure, but also illustrates that low-dose exposure can create a "silent state of dysfunction" that cannot be easily detected by conventional toxicological testing, and that exposure could render children more sensitive pharmacodynamically to subsequent chemical insults.

In summary, exposure to dieldrin residues in the school setting may increase children's risk of developing Parkinson's disease, which is the second most common neurodegenerative disorder in the U.S. (Kanthasamy et al., 2005). Dieldrin could render school children more sensitive to subsequent insults by other environmental contaminants with a mode of action similar to MPTP, and/or cause direct neuron loss. These irreversible effects on the nigrostriatal system could lead to an early onset of Parkinson's disease.

What are the existing health guidance values for dieldrin?

Since dieldrin has been withdrawn from registration for almost 20 years, OEHHA only notes the following three health guidance values:

U.S. EPA Reference Dose (RfD)

U.S. EPA has established an RfD of 0.00005 mg/kg-day for dieldrin (U.S.EPA, 1990), which was based on a 2-year rat feeding study (Walker et al., 1969). Walker et al. administered dieldrin (recrystallized, 99% active ingredient) to Carworth Farm "E" rats (25/sex/dose; controls 45/sex) for 2 years at a dose of 0, 0.005, 0.05 or 0.5 mg/kg-day. Body weight, food intake, and general health remained unaffected throughout the 2-year period, although at 0.5 mg/kg-day all animals became irritable and exhibited tremors and

occasional convulsions. No effects were seen in various hematological and clinical chemistry parameters. At the end of 2 years, females in the 0.05 or 0.5 mg/kg-day treatment group had increased liver weights and liver-to- body weight ratios ($p<0.05$). Histopathological examinations revealed liver parenchymal cell changes including focal proliferation and focal hyperplasia. These hepatic lesions were considered to be characteristic of exposure to an organochlorine insecticide. The LOAEL was identified as 0.05 mg/kg-day and the NOAEL as 0.005 mg/kg-day. U.S. EPA applied an uncertainty factor of 100 (10 for intraspecies variability and 10 for interspecies extrapolation) to the NOAEL to compute the RfD.

ATSDR Minimal Risk Level (MRL)

ATSDR has established a MRL of 0.00005 mg/kg-day (ATSDR, 2002). ATSDR employed the same study used by U.S. EPA and also applied a 100-fold safety factor to the NOAEL in deriving the MRL.

FAO/WHO Provisional Tolerable Daily Intake (PTDI)

In its background document for developing dieldrin guidelines for drinking-water quality (WHO, 2003), the World Health Organization (WHO) indicated that an acceptable daily intake (ADI) of 0.0001 mg/kg-day was recommended by the joint meeting of the Food and Agriculture Organization of the United Nations (FAO) and WHO in 1977 (FAO/WHO, 1978). This ADI was reaffirmed at a subsequent joint meeting in 1994 and renamed as a provisional tolerable daily intake (PTDI) of 0.0001 mg/kg-day because aldrin and dieldrin were no longer used as pesticides (FAO/WHO, 1995). The PTDI of 0.0001 mg/kg-day was used as the basis for developing the water quality guidelines.

WHO's 2003 document indicated that the PTDI was based on NOAELs of 1 mg/kg of diet in the dog and 0.5 mg/kg of diet in the rat, which are equivalent to 0.025 mg/kg of body weight per day in both species. An uncertainty factor of 250, which included a safety factor of 2.5 based on concern about carcinogenicity observed in mice, was applied to the 0.025 mg/kg-day NOAEL to derive the PTDI. This is the extent of information available because a copy of the original analysis cannot be located.

Is OEHHA recommending a child-specific reference dose for dieldrin?

From its literature search and review, OEHHA identified the 2006 Richardson et al. study for further review to determine if it could serve as the basis for establishing a chRD for dieldrin. Eight-week-old C57BL/6J mice were used in this study (Richardson et al., 2006). Female mice were dosed with 0, 0.3, 1, or 3 mg/kg of dieldrin every three days for two weeks prior to introduction of male mice for breeding. Dosing continued on the same schedule throughout gestation and lactation, and ended upon weaning of the pups (postnatal day 22). Mice were then housed separately by litter and sex. At 12 weeks of age, offspring of both sexes from control and dieldrin-treated groups received two

subcutaneous injections of saline or 10 mg/kg MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine, parkinsonism-inducer) 10 hours apart.

Richardson et al. observed no overt toxicity to the dams or offspring from all treatment groups. The animals exhibited no change in weight gain, no tremors, and no overt behavioral abnormalities. At 12 weeks of age, dieldrin was detected in striatal tissues of offspring from the 3 mg/kg treatment group; however, dieldrin residues were not found in the lower doses and control groups. To examine the effects of perinatal dieldrin exposure on the dopaminergic system, two biomarkers, the dopamine transporter (DAT) and the vesicular monoamine transporter 2 (VMAT2), were assayed just prior to administration of MPTP. DAT levels were significantly increased by 30 percent ($P<0.01$), 41 percent ($P<0.001$), and 52 percent ($P<0.001$) in male offspring of dams exposed to 0.3, 1 or 3 mg/kg of dieldrin, respectively. DAT levels were also increased in female offspring by 36 percent ($P<0.01$), 42 percent ($P<0.01$), and 61 percent ($P<0.001$) from the same treatment groups. Similarly, VMAT2 levels were increased in male offspring by 16 percent, 16 percent, and 27 percent ($P<0.01$); and in female offspring by 29 percent ($P<0.05$), 38 percent ($P<0.05$), and 59 percent ($P<0.01$). These results indicate that the effects on the dopaminergic system may be irreversible because they are also seen in those offspring that no longer have detectable levels of dieldrin in the striatal tissues.

As discussed above, the Corrigan study suggests that dieldrin may preferentially target the nigrostriatal system. To further test this hypothesis, Richardson et al., in addition to measuring DAT and VMAT2 levels, had quantified but did not observe any change in striatal GABA transporter levels or in cortical norepinephrine and serotonin transporter levels. These results support the model that dieldrin preferentially targets the nigrostriatal system.

To further define the critical period of vulnerability to dieldrin, Richardson et al. measured levels of NURR1, a factor which regulates DAT and VMAT2 mRNA transcriptions, and of DAT and VMAT2 mRNAs in 1-day-old and 12-week-old brains of offspring of dieldrin-treated females. NURR1, and DAT and VMAT2 mRNA levels were significantly increased in the 12-week-old brain but not in the 1-day-old brain. From this the authors concluded that the observed increased in DAT and VMAT2 (protein) levels may be a result of the amplification of DAT and VMAT2 mRNAs mediated through the dieldrin-induced expression of NURR1. More importantly, these findings suggest that the effects of dieldrin on the dopaminergic system are most likely the result of lactational exposure, and the critical window of vulnerability is probably postnatal rather than prenatal.

Richardson et al. observed that the alteration of DAT and VMAT2 levels by dieldrin had exacerbated the toxicity of MPTP. Striatal dopamine levels were used as measure of neuronal toxicity and an indicator for Parkinson's disease potential. In offspring of dieldrin-treated females, MPTP caused significantly greater reductions of dopamine in the male offspring (74 percent, 76 percent and 74 percent; all $P<0.05$) than in male offspring of controls. However, no appreciable difference between dieldrin-treated and control female offspring was noted. The increase in MPTP toxicity (marked by the

reduction of striatal dopamine) is expected with the increase in DAT levels because DAT facilitates neuronal entry of MPTP(Di Monte, 2003; Gainetdinov et al., 1997). The positive association of MPTP toxicity and VMAT2 levels, however, is unexpected because the increase in VMAT2 should have facilitated the sequestration of MPTP in vesicles, rendering it less available to exert its toxic effects (Di Monte, 2003; Miller, 2006; Staal and Sonsalla, 2000). In interpreting the data, the authors felt that the higher ratio of DAT to VMAT2 (both exhibited an increase but DAT's increase was greater) would be more pertinent in predicting the susceptibility of dopamine neurons to degeneration. In all, the data show that developmental dieldrin exposure has increased the susceptibility or sensitivity of dopminergic neurons to chemical insults later in life.

Pup dose (instead of maternal dose) is more applicable when evaluating the dose-response for consideration of establishing a chRD. However, dieldrin concentrations in milk and milk consumption rates were not measured. Thus, lactational pup doses cannot be estimated and maternal doses are used in this evaluation. Richardson et al. did not observe a NOAEL from the maternal dose range tested. A LOAEL of 0.3 mg/kg-3 days or approximately 0.1 mg/kg-day on a weekly basis was noted. In comparing the liver endpoint from the Walker study (used by U.S. EPA to establish the RfD and by ATSDR to derive the MRL) to the developmental neurotoxicity endpoint from the Richardson et al. study, it appears that the liver is a more sensitive endpoint. Table 1 provides a comparison that shows that a chRD based on the Richardson et al. study is less health protective than criteria based on liver weight endpoints. At this time, it is unclear where the NOAEL for the developmental neurotoxicity endpoint lies and the 10X for LOAEL-to-NOAEL conversion may or may not have underestimated the actual NOAEL. Additional postnatal studies will be necessary to pinpoint the "true" NOAEL for this developmental neurotoxicity endpoint. OEHHA will evaluate the merit of establishing a chRD when the new information becomes available.

Table 1

	Health Criteria (mg/kg-day)	Inter-species Factor	Intra-species Factor	LOAEL-to-NOAEL Factor	Modifying Factor	LOAEL* or NOAEL** (mg/kg-day)	Study	Endpoint
U.S. EPA RfD	0.00005	10	10	NA	NA	0.005**	2 yr rat	Increased liver weights and liver hyperplasia
ATSDR MRL	0.00005	10	10	NA	NA	0.005**	2 yr rat	Increased liver weights and liver hyperplasia
FAO/WHO PTDI	0.0001	10	10	NA	2.5	0.025**	Rat & dog	Not indicated
hypothetical chRD	0.0001	10	10	10	NA	0.1*	Perinatal mice	Dopamine system alteration

In conclusion, the Richardson study is highly relevant in the context of OEHHA's review pursuant to HSC §901(g). The study has demonstrated that dieldrin can adversely affect the developing brain. It also indicates the postnatal brain may be more vulnerable, which means infant and toddlers of daycare centers at certain school sites, as well as school children, may be sensitive receptors. It further illustrates that the epigenetic effect of dieldrin on the dopaminergic system is possibly irreversible, resulting in the increased susceptibility of the dopaminergic neurons to chemical insults later in life. While this developmental neurotoxicity appears to be a very sensitive endpoint, OEHHA cannot determine the lowest dose for this effect because the pup dose information is not available and a NOAEL is not observed in the maternal dose range tested. OEHHA is not proposing a chRD based on the maternal LOAEL from the Richardson et al. study, especially when the resulting health criteria would be less health protective to children. Accordingly, OEHHA is recommending the use of the RfD or MRL when assessing non-cancer risk of dieldrin at school sites in California.

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