February 13, 2002

Public Information and Record Integrity Branch
Information Resources and Services Division (7502C)
Office of Pesticide Programs
Environmental Protection Agency
1200 Pennsylvania Ave., NW
Washington, DC 20460

RE: docket control number PF-1045; 2,4-dichlorophenoxyacetic acid (2,4-D)

On January 11, 2002 the Industry Task Force II on 2,4-D Research Data ("2,4-D Industry Task Force") submitted comments to the EPA in response to World Wildlife Fund (WWF) comments submitted to the EPA on November 21, 2001. In essence, the Industry Task Force disagreed with WWF's arguments that a FQPA safety factor should be applied to 2,4-D to take into account both quantitative and qualitative developmental sensitivity. More specifically, the Task Force stated that a study demonstrating increased quantitative developmental sensitivity was re-evaluated in May 1996 and re-interpreted to show no increased developmental sensitivity. In addition, the Task Force disagreed with WWF's position that 2,4-D potentially exhibits "qualitative" increased susceptibility because it has been found to decrease thyroid hormone levels (hypothyroidism). Developmental hypothyroidism is a condition that is a well-documented in experimental animals models and humans to have adverse effects on brain development, including effects on intelligence and auditory function.

**EPA should require a developmental neurotoxicity study for pesticides that cause hypothyroidism**

The Industry Task Force II on 2,4-D Research Data comment indicated that a developmental rat study that was originally interpreted to show a developmental LOEL lower than a maternal LOAEL (i.e. evidence of increased quantitative susceptibility) was re-evaluated and the developmental and maternal LOELs were determined to occur at the same dose. As described by the 2,4-D Industry Task Force, WWF based its arguments on an EPA Federal Register (Federal Register: September 5, 1997; Vol. 62, No. 172; pp. 46900-46907) notice that did not discuss this change.

The broader issue beyond this particular study revision is that EPA should not continue to ignore the implications thyroid disrupting chemicals have for fetuses, infants and children. The link between developmental hypothyroidism and long term cognitive effects in both humans and laboratory animals is well-established. The Endocrine Disruptor Screening and Testing Advisory Committee (EDSTAC) stated that one of the most obvious deficiencies of the current

**World Wildlife Fund**
1250 Twenty-Fourth St., NW Washington, DC 20037-1132 USA
Tel: 202.293.4800 Fax: 202.887.5293
www.worldwildlife.org

*Affiliated with World Wide Fund for Nature*

multigenerational study is the lack of endpoints specific for thyroid disruption and suggested the inclusion of several endpoints to address this deficiency (Table 1). The EPA has largely put off endocrine disruptor regulation pending the outcome of the Endocrine Disruptor Screening Program. In the meantime, a developmental neurotoxicity study should be required for pesticides that elicit hypothyroidism, such as 2,4-D. The absence of a developmental neurotoxicity study for 2,4-D represents a significant data gap and should be reflected in the FQPA safety factor determination.

Although the developmental neurotoxicity study will likely not characterize many aspects of thyroid disruption, it should be more informative regarding developmental susceptibility than the rat or rabbit developmental studies, which are designed primarily to assess teratogenicity. The increased sensitivity of developmental neurotoxicity studies over the developmental rat and multigeneration study has already been demonstrated. A 1998 EPA draft report titled 'A Retrospective Analysis of Twelve Developmental Neurotoxicity Studies Submitted to the USEPA Office of Prevention, Pesticides and Toxic Substances (OPPTS)' found that the developmental neurotoxicity study resulted in a lower NOEL for many of the chemicals discussed when compared to the developmental rat or rat reproduction study (Table 10, page 36).

**Specific responses to comments made by the Industry Task Force II on 2,4-D Research Data**

1) WWF's own characterization of the causal data support is "insufficient evidence".

There is only insufficient evidence to know whether 2,4-D causes hypothyroidism in <u>pregnant dams and offspring</u>; there is more than sufficient evidence to show that 2,4-D causes hypothyroidism in adult male and female rats. In fact, there is no reason to believe 2,4-D would <u>not</u> cause hypothyroidism in pregnant dams.

2) The extensive toxicity profile available for 2,4-D provides scant evidence for an effect via the endocrine system.

This statement is perplexing as it appears to disregard thyroid effects found in several registrant studies (Table 2).

3) Observed effects on thyroid system function and pathology were limited to high-dose treatments, and were associated with clearly delineated NOELs that were higher than other well characterized non-endocrine target organ effects of 2,4-D.

We note that the thyroid effects described in registrant studies typically occurred at the study LOEL (Table 2).

4) Chronic studies in multiple species consistently indicate that the kidney is the primary target organ for 2,4-D toxicity. Other effects seen at higher doses, such as those associated with the thyroid system, are attributable to secondary mechanisms.

The fetal brain will not differentiate between primary and secondary effects. If a chemical causes hypothyroidism, it has the potential to adversely effect neurological development regardless of other target organs.

5) No consistent or treatment-related pattern of low dose endocrine-modulation toxicity is suggested.

WWF never suggested that 2,4-D caused thyroid toxicity at low doses. The low dose question is a separate issue from the increased susceptibility issue.

In summary, we strongly urge EPA to apply at least a 3X FQPA Safety Factor to 2,4-D to take into account the implications of thyroid toxicity for the neurological development of embryos, fetuses, infants, and children. Hypothyroidism clearly represents a qualitative increased susceptibility for children, especially those already at risk for congenital hypothyroidism, which is estimated to be 1 in 4,000 by the Thyroid Foundation of America.

We appreciate the opportunity to provide these comments in response to the Industry Task Force II on 2,4-D Research Data petition.

Sincerely,

Kristina Thayer, PhD
Program Scientist
Wildlife and Contaminants Program
(202) 822-3473

Theo Colborn, PhD
Senior Program Scientist and Director
Wildlife and Contaminants Program
(202) 778-9643

**Table 2. Comparison of Current Endocrine Related Endpoints Assessed in Multigenerational Studies and Additional Endpoints Recommended by EDSTAC**

| Test | current endocrine related endpoints | recommended endocrine related endpoints |
|---|---|---|
| 2-generation mammalian reproductive toxicity | **Estrogen Agonist/Antagonist**<br>gonad development (size, morphology, weight)<br>accessory sex organ (ASO) development (weight ± fluid, histology)<br>acquisition of vaginal patency (VP)<br>preputial separation (PPS)<br>fertility<br>fecundity<br>time to mating<br>mating and sexual behavior<br>ovulation<br>estrous cyclicity<br>gestational length<br>abortion<br>premature delivery<br>dystocia (difficult delivery)<br>spermatogenesis<br>epididymal sperm numbers and morphology<br>testicular spermatid head counts<br>daily sperm production (DSP)<br>efficiency of DSP<br>gross and histopathology of reproductive tissues<br>anomalies of genital tract<br>viability of conceptus in utero (prenatal demise)<br>survival and growth of offspring<br>maternal lactational behaviors (e.g. nursing, pup retrieval, etc.)<br>**Androgen Agonist/Antagonist**<br>altered apparent sex ration (based on anogenital distance)<br>malformations of the urogenital system<br>altered sexual behavior<br>changes in testis and ASO weights<br>effects on sperm numbers, morphology, etc.<br>retained nipples in male offspring<br>altered AGD (now triggered from VP/PPS)<br>male fertility<br>agenesis of prostate<br>changes in androgen dependent tissues in pups and adults (not limited to sex accessory glands)<br>**Thyroid Agonist/Antagonist**<br>growth, body weight<br>food consumption, food efficiency<br>developmental abnormalities<br>perinatal mortality<br>testis size and DSP<br>vaginal patency<br>preputial separation | Es**trogenic/Androgenic**<br>ASO function (secretory products)<br>nipple development and retention<br>androgen and estrogen levels<br>LH and FSH levels<br>testis descent<br>**Thyroid**<br>neurobehavioral deficits<br>TSH, T4<br>thyroid weight and histology (e.g. goiter)<br>pinna detachment<br>surface righting reflex<br>eye opening<br>acquisition of auditory startle<br>negative geotaxis<br>mid-air righting reflex<br>motor activity on PND 13, 21<br>postwean includes motor activity on PND21 and postpuberty ages (sex difference)<br>learning and memory PND60 (acitive avoidance/water maze, brain weight (absolute), whole and cerebellum brain histology. |

**Table 2: Thyroid Effects Noted in the California Department of Pesticide Regulation Toxicological Summary for 2,4-D (http://www.cdpr.ca.gov/docs/toxsums/toxsumlist.htm)**

| Study Design | Thyroid Effects (LOEL) |
|---|---|
| combined chronic toxicity/oncogenicity (rat)<br>study NOEL = 5 mg/kg/d<br>(0, 5, 75, and 150 mg/kg/d tested) | ↑ thyroid gland weight (75 mg/kg/d)<br>↓ T4 (♀,♂), strongest effect in ♀(75 mg/kg/d)<br>slight ↓ thyroglobulin (♀) (LOEL not explicit) |
| subchronic toxicity (rat)<br>study NOEL = 15 mg/kg/d<br>(0, 1, 15, 100, and 300 mg/kg/d tested) | ↓ thyroid hormone, especially T4 (100 mg/kg/d) |
| subchronic toxicity (rat)<br>study NOEL = 18.1 mg/kg/d<br>(0, 1.2, 18.1, 120, and 361 mg/kg/d tested) | ↓ thyroid hormone, especially T4 (120 mg/kg/d)<br>thyroid follicular cell hypertrophy (361 mg/kg/d) |

## Documentation


Makris S, Raffaele K, Sette W, Seed J. A retrospective analysis of twelve developmental neurotoxicity studies submitted to the USEPA Office of Prevention, Pesticides, and Toxic Substances (OPPTS). US EPA, 1998.