APPENDIX TO *POISON ON PETS II*: PROJECT METHODS

Scope of Project

This project was designed to assess human exposure and health risk from the pesticides tetrachlorvinphos (TCVP) and propoxur from flea collars containing these chemicals. This was accomplished by quantifying pesticide residues on pet fur for TCVP and propoxur, and measuring concentrations of propoxur in air in a room containing a flea collar. Dose estimates for oral and dermal exposures were calculated and compared with levels EPA considers acceptable (non-cancer RfDs) because no appreciable harm is expected. Lifetime cancer was also calculated based on exposure levels.

Pesticide Residue Testing Methods

Study Subjects

Ten NRDC staff members from the San Francisco and Washington, D.C., offices volunteered a total of nine dogs and five cats to participate in the flea collar residue study. The first round of sampling was conducted in June 2007 involving all 14 pets. Two dogs from a single household also participated in a second round of sampling in October 2008. Volunteers were screened to prevent the participation of families with young children or pregnant women or households with prior use of flea collars. Each volunteer was given information about the goals, methods, and potential risks of the study and signed a consent form prior to participating. The pets varied in size and fur length. The dogs weighed between 30 and 80 pounds (average 60 pounds) and the cats weighed between 12 and 20 pounds (average 15 pounds). During the study period, animals were closely monitored for adverse reactions, and participation in the study was terminated following any signs of irritation or distress.[1] Participants were instructed to avoid bathing their pets for the duration of the study and were advised orally and in writing about keeping children and pregnant women away from the pets, washing their hands thoroughly after touching their pets, and bathing the pets at the completion of the study.

Prior to treatment, none of the pet owners reported prior use of flea control products containing TCVP or propoxur. Frontline® (fipronil) was the most commonly used product for flea and tick control (five pets). Other products employed by participants included Advantage® (imidacloprid) and Sentinel® (lufenuron). For six participants, mainly cats, there was no reported use of flea and tick control products prior to participation in the study.

Flea Collars

Four types of commercially available flea collars, containing two different pesticides, were employed in this study.[2] Table A-1 summarizes the information for each type of collar.

TABLE A-1: FLEA COLLARS EMPLOYED IN THIS STUDY

FLEA COLLAR	ACTIVE INGREDIENT(S) (AI)	PERCENT	NET WT. OF COLLAR (OZ.)	QUANTITY OF ACTIVE INGREDIENTS (g)
Hartz Advanced Care 3 in 1 Collar for Cats	TETRACHLORVINPHOS	14.55	0.6	2.47
	(S)-METHOPRENE	1.02		0.17
Hartz Advanced Care 2 in 1 Reflecting Flea & Tick Collar for Dogs	TETRACHLORVINPHOS	14.55	0.85	3.51
Zodiac Flea & Tick Collar for dogs	PROPOXUR	10.0	1.2	3.40
Bio Spot Flea and Tick Collar for dogs	PROPOXUR	10.0	1.2	3.40

Sampling Methods For Flea Collar Subjects

Study participants were assigned a flea collar containing either propoxur or tetrachlorvinphos (TCVP). Participants with more than one pet participating in the study were generally assigned the same type of collar for both pets. A total of 10 animals (five dogs and five cats) were assigned to the TCVP group, and four dogs were assigned to the propoxur group. All 14 animals were sampled for pesticide residue prior to the application of the collar and then again after three days; eight of these animals were sampled again at 14 days. Table A-2 summarizes the description of the study participants and treatment groups.

TABLE A-2: STUDY PARTICIPANT DESCRIPTION

ANIMAL	WEIGHT	FUR LENGTH	TYPE OF COLLAR	DURATION OF STUDY (DAYS)	PRIOR FLEA/TICK TREATMENT USED
CAT	N.A.	MED	TCVP	3	NONE
CAT	13	SHORT	TCVP	3	NONE
CAT	12	LONG	TCVP	3	NONE
CAT	15	SHORT	TCVP	14	IMIDACLOPRID (ADVANTAGE®)
CAT	20	LONG	TCVP	14	NONE
DOG	68	SHORT	TCVP	3	FIPRONIL (FRONTLINE®)
DOG	65	SHORT	TCVP	3	FIPRONIL (FRONTLINE®)
DOG	50	MED	TCVP	14	FIPRONIL (FRONTLINE®)
DOG	80	SHORT	TCVP	14	NONE
DOG	70	MED	TCVP	14	LUFENURON (SENTINEL®)
DOG	75	MED	PROPOXUR	3	LUFENURON (SENTINEL®)
DOG*	60	SHORT	PROPOXUR	14	FIPRONIL (FRONTLINE®)
DOG*	30	MED	PROPOXUR	14	FIPRONIL (FRONTLINE®)
DOG	45	MED	PROPOXUR	14	NONE

*PARTICIPATED IN SECOND ROUND OF SAMPLING

Flea collars were applied to the animals following instructions on the packaging, making certain that the collar was not applied too tightly and leaving 2–3 inches of collar following the buckle.

Pesticide residues were sampled using a protocol based on the methods used by Chambers et al. 2007.[3] Residues were collected on circular glass microfiber filters 11 cm in diameter moistened with a 3 percent surfactant solution containing sodium dioctyl sulfosuccinate, used to simulate human perspiration.[4] A rectangular area below the collar and around the animal's shoulders (250 cm^2 for dogs and 60 cm^2 for cats) was wiped thoroughly for one minute using moderate to firm strokes with the moistened filter paper to simulate petting. Care was taken to avoid wiping the flea collar or the fur directly beneath the collar. Wipe samples were stored in a closed wide-mouth jar kept at < 4^oC until analysis.

Two dogs from the same household participated in a second round of sampling with a flea collar containing propoxur. The second round of sampling was conducted using the same protocol, with the addition of supplemental samples taken to simulate petting that occurred across the collar itself. This modification was made in response to observations of the pet owner that contact with the collar was a frequent occurrence during normal pet handling. The results of this sampling were used as the basis of a high-exposure scenario in the cancer risk assessment in which half of the contact with the dog included touching the collar and half did not.

Analytical Methods for Wipe Samples

Pesticide residue levels were determined by Environmental Micro Analysis, Inc., Woodland, California, using EPA method 8141A for tetrachlorvinphos and method 8318 for propoxur. The filter paper wipes were extracted with acetone and analyzed for tetrachlorvinphos by gas chromatography with pulsed flame photometric detection. Propoxur analyses were conducted using high-pressure liquid chromatography with post column derivitization and fluorescence detection.

Two quality control samples were also analyzed for each batch run by the laboratory, a blank and a spike. A blank wipe that had not contacted a pet or any pesticide-treated surface was extracted and analyzed in the same manner as the samples. No pesticides were detected in the blank wipe. An additional wipe was spiked with a known amount of propoxur and tetrachlorvinphos standard, extracted and analyzed as above. Recoveries of propoxur and tetrachlorvinphos from the spiked samples were 99–103 percent and 86–92 percent, respectively.

Air Testing Methods

Indoor air testing was performed to determine whether the use of propoxur-containing flea collars could result in measurable levels of propoxur in the air. Propoxur-containing flea collars were chosen for this testing due to the volatility (vapor pressure = 1.29 mPa) of the chemical and the pronounced chemical odor associated with use of the flea collar. A scenario of someone sleeping with his or her pet was simulated by placing a collar in a small, closed room for eight hours with limited air exchange.[5] A Zodiac Flea & Tick Collar for large dogs, containing 10 percent propoxur at a total weight of 1.2 oz., was used for this experiment. This simulation was conducted without the use of a pet.

Sampling Methods

The air sampling was conducted in an unoccupied room approximately 38 m^3 in size. The sampling equipment was set up at a height of 90 cm (35 inches). A flea collar was activated and handled according to instructions on the package. Instead of being fastened around the neck of a pet, the collar was attached to a loop of string, allowing it to hang unobstructed at approximately the same height as the intake for the air sampling equipment. The collar was positioned 85 cm (33.5 inches) from the sampling equipment.

The sampling was conducted in accordance with EPA sampling method TO-10A, Determination of Pesticides and Polychlorinated Biphenyls in Ambient Air Using Low Volume Polyurethane Foam (PUF) Sampling Followed by Gas Chromatographic/Multi-Detector Detection (GC/MD).[6] The sampling equipment consisted of a personal sampling pump connected to a low-volume sampling PUF/Tenax tube (SKC catalog number 226-124) with flexible tubing. Immediately following the setup of the flea collar, the pump was activated and allowed to run for eight hours at a flow rate of 2.02 L/min. The door to the room was closed during the sampling period. At the end of the sampling period, the researcher reentered the room, removed the tube from the sampling equipment, wrapped it in aluminum foil and bubble wrap, and placed it in a closed glass jar. The jar was kept at < 4°C until analysis by Analytical Sciences Laboratory, Petaluma, California. A comparison blank sample was obtained using the same methodology in an adjacent room of the same size with no flea collar present.

Analytical Methods

The concentration of propoxur in the PUF tubes was determined by a commercial laboratory using the following methods. The samples were extracted with an ASE 100 Accelerated Solvent Extraction System (Dionex). Each PUF tube was placed in a stainless steel extraction cell and extracted with several aliquots of 1:1 methylene chloride/acetone at 100°C and 1,500 PSI. The extracts were then transferred to round-bottom flasks and concentrated with a rotary evaporator to a volume of 2 mL. The extracts were then analyzed by GC/MS operating in selective ion monitoring mode.

Two quality control samples were also analyzed, a blank and a spike. A blank PUF tube that had never been exposed to pesticides was extracted and analyzed in the same manner as the samples. No pesticides were detected in the blank tube. An additional tube was spiked with a known amount of propoxur standard, then extracted and analyzed as above. The recovery of propoxur from the spiked sample was 82 percent.

Estimation of Levels of Concern for Toddler Pesticide Exposure

Pesticide residues on pets and on hands following the petting of an animal present opportunities for exposure via dermal absorption and ingestion. Young children, who engage in significant hand-to-mouth activity, have the highest risk of exposure to pesticide residues from flea collars via ingestion. Using two exposure scenarios, we estimated the level of pesticide residue on a child's hand that would correspond to the reference dose for non-cancer effects established by EPA for tetrachlorvinphos and propoxur.[7,8] This calculation was based on formulas used to estimate the ingested and dermal dose to a toddler from the residues measured after the petting simulation, accounting for both direct hand-to-mouth activity and contact with objects or food that are then placed in the mouth. EPA's safety evaluations for tetrachlorvinphos and propoxur in flea collars rely on outdated data and unsafe assumptions for estimating children's exposure to pesticides used in flea collars. Therefore, we supplemented or replaced EPA methodology where necessary to conduct a more thorough analysis that better captures the vulnerabilities of young children. Our calculations include two scenarios that represent an average level and a high level of contact with a pet.

General Parameters

Duration of Exposure

The duration of exposure reflects the time (hours per day) that a child would be expected to spend with a pet wearing a flea collar. Review of the published literature and other risk assessments revealed very little data describing this parameter. EPA's exposure assessment for residential post-application exposure to pesticides in flea collars in both the Organophosphate Cumulative Risk Assessment (2006) and the draft N-Methyl Carbamate Cumulative Risk Assessment

(2007) rely on a study by Freeman et al. (2001) that included only three children with pets.[9,10,11] Based on the results of this study, EPA assumed a triangular distribution with a minimum value of 0.03 hrs./day and a maximum value of 1.03 hrs./day of time spent with a pet. These numbers are much smaller than the estimate used in the risk assessment performed for dichlorovos (DDVP) in flea collars and the EPA SOPs for Residential Exposure Assessments, which both assume that a child plays with a pet for 2 hrs./day.[12] However, none of these numbers reflect what we would consider a high-end scenario, which might involve a child who spends many hours with a pet (including sleeping) or multiple pets. We utilized the estimate from the DDVP risk assessment for our average exposure scenario and 8 hrs./day of exposure to capture the high end scenario.

Toddler Weight

EPA risk assessments typically attempt to capture the risks to toddlers defined as children 1–6 years old and assumed to weigh 15 kilograms. In reality, children at the younger end of this range are likely to have much greater hand-to-mouth activity and also weigh less than 15 kilograms.[13,14] Therefore, the risks of pesticide exposure to this vulnerable subgroup is often greatly underestimated. We used data from the EPA (1997) Exposure Factors Handbook to estimate the weight of 1- and 2-year-olds by taking the mean of reported median weights of male and female 1- and 2-year-olds.[15] Using this weight, we also derived a scaling factor (11/15 = 0.74) to adjust the other body surface area parameters to reflect the smaller body of these toddlers.

GENERAL PARAMETERS

			AVERAGE		*HIGH*	
PARAMETER		**UNIT**	**VALUE**	**SOURCE**	**VALUE**	**SOURCE**
GENERAL	DURATION OF EXPOSURE	HRS/DAY	2.0	EPA (2006) DDVP RED	8.0	AUTHOR'S DETERMINATION OF HIGH END TO INCORPORATE SLEEPING W. PET
	TODDLER (1 - 2-YEAR OLD) WEIGHT	KG	11.0	EPA (1997) EXPOSURE FACTORS HANDBOOK	11.0	EPA (1997) EXPOSURE FACTORS HANDBOOK
	REFERENCE DOSE, PROPOXUR	MG / KG-DAY	.005	EPA (1997) PROPOXUR RED	.005	EPA (1997) PROPOXUR RED
	REFERENCE DOSE, TETRACHLORVINPHOS	MG/KG-DAY	.04	EPA (2006) RED TETRACHLORVINPHOS	.04	EPA (2006) RED TETRACHLORVINPHOS

Dermal Exposure

EPA's exposure assessment for residential post-application exposure to pesticides in flea collars in both the Organophosphate Cumulative Risk Assessment (2006) and the draft N-Methyl Carbamate Cumulative Risk Assessment (2007) is based on dislodgeable residues on pet fur and a transfer coefficient empirically derived from a small study of pet groomers. This method does not adequately reflect children's exposure to pesticide residues left by flea collars. Therefore, we utilized the following equation, based on the DDVP risk assessment, which more thoroughly encompasses the various components of exposure expected following use of a flea collar.

$$\text{DOSE (MG / KG - DAY)} = \frac{\left\{\begin{array}{l}\text{MEASURED}\\\text{CONC. ON WIPE}\\\text{(MG / CM}^2\text{)}\end{array}\right\} \times \left\{\begin{array}{l}\text{SURFACE AREA}\\\text{IN CONTACT}\\\text{WITH PET (CM}^2\text{)}\end{array}\right\} \times \left\{\begin{array}{l}\text{LOADING}\\\text{FREQUENCY}\\\text{(EVENTS/HR)}\end{array}\right\} \times \left\{\begin{array}{l}\text{FRACTION}\\\text{ABSORBED}\end{array}\right\} \times \left\{\begin{array}{l}\text{TIME SPENT}\\\text{WITH ANIMAL}\\\text{(HRS/DAY)}\end{array}\right\}}{\textit{WEIGHT OF TODDLER (KG)}}$$

Surface Area

The surface area reflects the area of the body in contact with the pet and therefore containing any pesticide residues. EPA's exposure assessment for residential post-application exposure to pesticides in flea collars in both the Organophosphate Cumulative Risk Assessment (2006) and the draft N-Methyl Carbamate Cumulative Risk Assessment (2007) do not give values for this parameter. We assume in our average exposure scenario that a child touches the pet with the palms of both hands and the lower forearms. In the high-end scenario we utilize the "pet hug" scenario described in the DDVP risk assessment. Both of these measurements were scaled to reflect a 1- 2-year-old child.

Loading Frequency

The loading frequency reflects the number of times the child touches the pet. EPA's exposure assessment for residential post-application exposure to pesticides in flea collars in both the Organophosphate Cumulative Risk Assessment (2006) and the draft N-Methyl Carbamate Cumulative Risk Assessment (2007) does not give values for this parameter, and the DDVP risk assessment assumes only one hug per day. In the absence of other data, we estimated a loading frequency using our best judgment for what might be a reasonable expectation for child/pet interactions.

Fraction Absorbed

We used EPA reported values for the fraction of pesticide absorbed through the skin from the TCVP Reregistration Eligibility Decision (RED) and the Propoxur Reregistration Eligibility Decision (RED).

DERMAL EXPOSURE PARAMETERS

			AVERAGE		*HIGH*	
PARAMETER		UNIT	VALUE	SOURCE	VALUE	SOURCE
DERMAL	SURFACE AREA IN CONTACT WITH PET	CM^2	412.0	SURFACE OF BOTH PALMS PLUS LOWER FOREARMS FROM EPA (1997) EXPOSURE FACTORS HANDBOOK SCALED FOR 1- 2-YR-OLD	1380.0	PET HUG FROM EPA DDVP RED SCALED FOR 1- 2-YR-OLD
	LOADING FREQUENCY TCVP FRACTION	EVENTS/HR	4.0	AUTHORS' JUDGMENT	4.0	AUTHORS' JUDGMENT
	FRACTION ABSORBED – TCVP	N/A	0.096	EPA (2006) TCVP RED	0.096	EPA (2006) TCVP RED
	FRACTION ABSORBED– PROPOXUR	N/A	0.2	EPA (1997) PROPOXUR RED	0.2	EPA (1997) PROPOXUR RED

Oral Exposure

Direct Hand-to-Mouth Activity

Direct hand-to-mouth activity is estimated by the following equation taken from the U.S. EPA Standard Operating Procedures (SOPs) for Residential Exposure Assessments, which has been used by EPA to estimate pesticide exposures to toddlers and young children associated with the use of flea collars containing tetrachlorvinphos and dichlorvos.[16,17,18]

$$\text{DOSE (MG / KG - DAY)} = \frac{\left\{\begin{matrix}\text{MEASURED}\\ \text{CONC. ON WIPE}\\ (\text{MG / CM}^2)\end{matrix}\right\} \times \left\{\begin{matrix}\text{SURFACE AREA}\\ \text{OF FINGERS OR}\\ \text{HANDS}\\ \text{MOUTHED (CM}^2)\end{matrix}\right\} \times \left\{\begin{matrix}\text{FREQUENCY OF}\\ \text{IMOUTHING}\\ \text{ACTIVITY}\\ \text{(EVENTS/HR)}\end{matrix}\right\} \times \left\{\begin{matrix}\text{SALIVA}\\ \text{EXTRACTION}\\ \text{FACTOR}\end{matrix}\right\} \times \left\{\begin{matrix}\text{TIME SPENT}\\ \text{WITH ANIMAL}\\ \text{(HRS/DAY)}\end{matrix}\right\}}{\textit{WEIGHT OF TODDLER (KG)}}$$

Indirect Hand-to-Mouth Activity

Indirect hand-to-mouth activity reflects exposures to pesticides that occur when a child with pesticides on his or her hand touches food or an object that is then placed in the mouth. Although EPA's risk assessment methodologies do not include this route of exposure, the California Environmental Protection Agency Office of Environmental Health Hazard Assessment (OEHHA) and numerous studies recognize that this is a potential route of exposure to environmental contaminants.[19,20] To quantify this exposure, we adapted OEHHA's equation for indirect hand-to-mouth activity, developed as a component of the exposure assessment guidelines for adult exposure to lead in fishing tackle.

$$\text{DOSE (MG / KG - DAY)} = \frac{\left\{\begin{matrix}\text{MEASURED}\\ \text{CONC. ON WIPE}\\ (\text{MG / CM}^2)\end{matrix}\right\} \times \left\{\begin{matrix}\text{SURFACE AREA}\\ \text{IN CONTACT}\\ \text{WITH OBJECT}\\ (\text{CM}^2)\end{matrix}\right\} \times \left\{\begin{matrix}\text{FREQUENCY OF}\\ \text{INDIRECT HTM}\\ \text{ACTIVITY}\\ \text{(EVENTS/HR)}\end{matrix}\right\} \times \left\{\begin{matrix}\text{TRANSFER}\\ \text{FACTOR}\end{matrix}\right\} \times \left\{\begin{matrix}\text{TIME SPENT}\\ \text{WITH ANIMAL}\\ \text{(HRS/DAY)}\end{matrix}\right\}}{\textit{WEIGHT OF TODDLER (KG)}}$$

Surface Area

The surface area mouthed reflects the area of the body that a child puts in his or her mouth. We utilized EPA's methods employed in the Organophosphate (OP) Cumulative Risk Assessment (CRA) that assumed mouthing of the area of three fingers. For calculating indirect hand-to-mouth exposures, the surface area is the area of the hand that touches the mouthed object or food. Since developmentally toddlers are less likely to have advanced fine motor skills and more likely to use their entire hand to hold an object or food, we used the area of the palm of a single hand.[21] Both of these measurements were scaled to reflect a smaller/younger toddler.

Frequency of Mouthing Activity

EPA's exposure assessment for residential post-application exposure to pesticides in flea collars in the Organophosphate Cumulative Risk Assessment (2006) relies on a study by Freeman *et al.* (2001) of 19 children ages 3–12. This ignores the increased vulnerability of younger children, who have been found to have increased rates of hand-to-mouth activity.[22] Therefore, we used the frequency of hand-to-mouth activity reported in a recent meta-analysis performed by Xue *et al.* (2007), incorporating the mean and 95th percentile reported for 1- and 2-year-olds for our average and high-end scenarios, respectively. For the indirect hand-to-mouth exposure assessment, we used the reported values from Ko *et al.* (2007), which observed direct and indirect hand-to-mouth activity for 37 children 1–5 years old and found a statistically significant positive correlation between object/food-in-mouth frequency and blood lead levels.[23]

Transfer Factors

EPA risk assessments and guidance materials provide a number of different estimates (ranging from 0.5 to 1) to describe the efficiency of saliva in transferring pesticides from the hand to the mouth, known as a saliva extraction or direct transfer factor. We used the value selected by OEHHA (0.5) in its guidelines for lead in fishing tackle, which was based on the removal efficiency literature for three pesticides. The indirect transfer factor describes the percent of pesticide

ingested as a function of the loading level on the hand due to placement of food or objects contaminated as a result of handling in the mouth. For this parameter we used the value described in OEHHA's guidelines for exposure assessment for lead in fishing tackle because other estimates are unavailable.

Additional Assumptions

The following additional assumptions were made: 1) The hands are washed after playing with the pet, thus ending the opportunity for further dosing. This assumption is unstated but implicit in the way the calculation is set up. 2) The measured concentration on the wipe is equivalent to the dislodgeable residue that a person would experience from petting the animal.

ORAL EXPOSURE PARAMETERS

			AVERAGE		*HIGH*	
PARAMETER		UNIT	VALUE	SOURCE	VALUE	SOURCE
ORAL	SURFACE AREA MOUTHED	CM^2	15.0	EPA (2006) OP CRA -FINGERS (SCALED 1-2 YEAR OLD)	15.0	EPA (2006) OP CRA -FINGERS (SCALED 1-2 YEAR OLD)
	FREQUENCY OF DIRECT MOUTHING ACTIVITY	PER HR	19.6	XUE ET AL. (2007) - MEAN (1-2 YEAR OLD)	63.0	XUE ET AL. (2007) - 95TH PERCENTILE
	SALIVA EXTRACTION FACTOR/DIRECT TRANSFER FACTOR	N/A	0.5	OEHHA GUIDELINES - PB IN FISHING TACKLE	0.5	OEHHA GUIDELINES - PB IN FISHING TACKLE
	SURFACE AREA IN CONTACT W. OBJECT	CM^2	65.0	PALMAR SURFACE OF ONE HAND (SCALED 1-2 YEAR OLD)	65.0	PALMAR SURFACE OF ONE HAND (SCALED 1-2 YEAR OLD)
	FREQUENCY OF INDIRECT HTM ACTIVITY	PER HR	15.0	KO ET AL. (2007) MEAN OBJECT/FOOD IN MOUTH	66.0	KO ET AL. (2007) 90TH PERCENTILE OBJECT/FOOD IN MOUTH
	INDIRECT TRANSFER FACTOR	NA	0.25	OEHHA GUIDELINES - PB IN FISHING TACKLE	0.25	OEHHA GUIDELINES - PB IN FISHING TACKLE

The safe level is calculated by combining the three formulas as follows:

$$\text{MEASURED CONC. ON WIPE (MG / CM}^2\text{)} = \frac{\left\{\text{REFERENCE DOSE}\right\} \times \left\{\text{TODDLER WEIGHT}\right\}}{\left\{\text{DIRECT ORAL DOSE}\right\} + \left\{\text{INDIRECT ORAL DOSE}\right\} + \left\{\text{DERMAL DOSE}\right\}}$$

Cancer Risk Assessment

We conducted a separate analysis, using a combination of EPA and California EPA OEHHA risk assessment methodology, of the cancer risk due to the residues from flea collars containing propoxur and tetrachlorvinphos. Because

propoxur is listed under California Proposition 65 as known to the state to cause cancer, we used exposure assessment methods consistent with risk assessments performed by entities considered authoritative bodies under Proposition 65. We assumed that the pesticide residue levels measured after 14 days reflect the average daily levels to be expected over the duration of use of the collar (150-180 days), after which the collar is replaced, as suggested by label instructions. In addition, in the propoxur assessment, we evaluated two exposure scenarios: one that assumes no direct contact with the flea collar and one that incorporates the risks of everyday pet contact that may include inadvertent contact with the collar.

We calculated the lifetime cancer risk to an adult pet owner using standard methodologies from U.S EPA and California EPA. The cancer risk calculation is based on the Cancer Potency Factor (q*) for each chemical. We divided the standard adult body weight of 70 kilograms by the q* and multiplied it by 1/100,000 to determine the Proposition 65 No Significant Risk Level (NSRL) for propoxur, and by 1/1,000,000 to determine the level that U.S. EPA considers to be a significant cancer risk for both pesticides.[24,25]

For propoxur, the q* of 0.0037 mg/kg-day taken from the 1997 U.S. EPA Registration Eligibility Document (RED) generated a Proposition 65 NSRL of 0.189 mg/day (189 µg/day).[26,27] This exposure, if sustained on a daily basis for a lifetime, would be projected to generate one excess cancer per 100,000 people; a one-per-million cancer risk would be expected at 18.9 µg/day. For TCVP, the q* value of 0.00183 was taken from the 2006 U.S. EPA Registration Eligibility Document, and would generate a risk of one excess cancer per 100,000 people at a dose of 0.383 mg/day (383 µg/day), and a one-per-million risk at 38.3 µg/day.[28] Using a combination of Cal/EPA and U.S. EPA exposure assessment methods, we calculated the estimated daily dose for an adult living with a pet that wears a flea collar. Comparison of this dose with the NSRL provides an estimate of the number of excess cancers per 100,000 people and the degree to which Proposition 65 listed substances pose a significant cancer risk under California law. Comparison with the one-per-million risk number indicates the degree to which this substance would be considered a significant cancer risk by the U.S. EPA.

However, this methodology fails to account for the increased cancer risk posed by exposures during early life stages, when the rapidly growing body is more likely to be permanently impacted by cancer-causing chemicals. To incorporate the increased susceptibility of children to carcinogens like propoxur and tetrachlorvinphos, we utilized the methods described in EPA's Supplemental Guidance for Assessing Susceptibility From Early-Life Exposure to Carcinogens to calculate lifetime risk, assuming that an individual is exposed as both a child and an adult to a pet wearing a flea collar.[29] This Supplemental Guidance recommends adjusting for the added cancer risk associated with early life stage exposures by using a 10× factor for 0 to <2-year-olds and a 3× factor for 2 to <16. Lifetime risk is calculated as a time-weighted sum of the risks calculated for each age category. Since exposure levels would be expected to differ between young children and adults, we calculated them separately and incorporated these values into the assessment. Consistent with standard EPA risk assessment methodology, the exposure level for young children includes ages 0 to <6. Adult exposure levels were assumed for the remaining age groups (ages 6 to 70). The cancer risk for an individual age group was calculated according to the following formula:

> Cancer risk = [cancer potency factor] × [dose rate] × [Age Dependent Adjustment Factor] × [lifetime exposure weighting factor = (number of years in group/70)]

Dermal Exposure: Based on EPA Organophosphate Cumulative Risk Assessment (CRA) Methodology

The EPA CRA calculation of dose rate is the product of the residue concentration multiplied by a transfer coefficient and

the time spent with the pet.[30] EPA derived an average transfer coefficient for adults of 1817 cm^2/hour and scaled it for children to 673 cm^2/hour. EPA chose values of time spent with the pet ranging from a most-likely value of 0.108 hours (6 minutes) to 1.025 hours per day.[31] For this calculation we used the higher EPA estimate of 1.025 hours per day.

Hand-to-Mouth Exposure Based on OEHHA Methodology

Although adults can be exposed to propoxur from flea collars via direct hand-to-mouth (HTM) activity (e.g., nail biting) as well as indirect activity (e.g., eating), EPA did not calculate any hand-to-mouth exposures for adults in the risk assessments of flea control products. However, OEHHA has developed a methodology for assessing adult HTM exposure to lead in fishing tackle for purposes of Proposition 65.[32] This method can easily be adapted for use with pesticides in flea collars.

The calculation for adult direct HTM exposure is the product of the propoxur loading on the hand, the surface area of the part touching the mouth, a transfer factor, the rate of HTM contact, and the number of hours spent with the pet.[33] With minor modifications as stipulated by OEHHA, this same equation can be used to calculate indirect HTM exposures as well as the direct HTM pathway. For this equation, we used the measured wipe values from our study at 14 days, both including and excluding direct contact with the collar. The surface area (SA), transfer factor (F), and contact frequency (I) are taken directly from the OEHHA guideline document. For time spent with the pet, we used the EPA assumption of 1.025 hours per day from the Organophosphate Cumulative Risk Assessment.

PARAMETERS FOR ADULT CANCER RISK ASSESSMENT

PARAMETER	VALUE	UNIT	SOURCE
F_{DIRECT} DIRECT HTM TRANSFER FACTOR	0.5	NA	(OEHHA 2008) Guidelines for HTM Lead Exposure from fishing tackle
F_{DIRECT} INDIRECT HTM TRANSFER FACTOR	0.25	NA	(OEHHA 2008) Guidelines for HTM Lead Exposure from fishing tackle
SA_D SURFACE AREA CONTACTED FOR DIRECT TRANSERS FOR MEN	19.0	CM^2	(OEHHA 2008) Guidelines for HTM Lead Exposure from fishing tackle
I_D DIRECT HTM CONTACT FREQUENCY	9.0	PER HOUR	(OEHHA 2008) Guidelines for HTM Lead Exposure from fishing tackle
SA_I SURFACE AREA CONTACTED FOR INDIRECT TRANSFERS FOR MEN	190.0	CM^2	(OEHHA 2008) Guidelines for HTM Lead Exposure from fishing tackle
I_I INDIRECT HTM CONTACT FREQUENCY	1.0	PER HOUR	(OEHHA 2008) Guidelines for HTM Lead Exposure from fishing tackle
DURATION OF EXPOSURE	1.025	HRS	EPA (2006) OP CRA (maximum value)
NSRL	189.0	µg/DAY	For Propoxur

For direct and indirect HTM exposure for children, we used the same values from our non-cancer risk assessment with a few minor modifications to cover the 0 to <6 age range and to be consistent with OEHHA's adult exposure methodology.

PARAMETERS FOR CHILDREN'S CANCER RISK ASSESSMENT

PARAMETER	VALUE	UNIT	SOURCE
F_{DIRECT} DIRECT HTM TRANSFER FACTOR	0.5	NA	(OEHHA 2008) Guidelines for HTM Lead Exposure from fishing tackle
F_{DIRECT} INDIRECT HTM TRANSFER FACTOR	0.25	NA	(OEHHA 2008) Guidelines for HTM Lead Exposure from fishing tackle
SA_D SURFACE AREA CONTACTED FOR DIRECT TRANSFERS FOR CHILDREN	20.0	CM^2	EPA (2006) OP CRA - fingers
I_D DIRECT HTM CONTACT FREQUENCY	19.6	PER HOUR	Xue et al. (2007) - mean (1 - 2 y.o)
SA_I SURFACE AREA CONTACTED FOR INDIRECT TRANSFERS FOR CHILDREN	88.0	CM^2	Surface of one palm; derived from EPA SOP where 350 cm = SA of two hands
I_I INDIRECT HTM CONTACT FREQUENCY	15.0	PER HOUR	Ko et al. (2007) mean object/food mouth
DURATION OF EXPOSURE	1.025	HRS	EPA (2006) OP CRA (maximum value)

CANCER RISK FROM EXPOSURE TO PROPOXUR INCORPORATING CHILDREN'S EXPOSURES

AGE CATEGORY	WEIGHT (KG)	EXPOSURE (MG/DAY)		DOSE MG/KG - DAY		CANCER POTENCY FACTOR	AGE DEPENDENT ADJUSTMENT FACTOR (ADAF)	LIFETIME EXPOSURE WEIGHTING FACTOR	CANCER RISK	
		FUR ONLY 14 DAYS	COLLAR CONTACT INCL. 14 DAYS	FUR - ONLY 14 DAYS	COLLAR CONTACT INCL. 14 DAYS				FUR - ONLY 14 DAYS	COLLAR CONTACT INCL. 14 DAYS
0 TO < 2	11	0.65	6.48	0.06	0.59	0.0037	10	0.03	6.25E-05	6.23E-04
2 TO < 6	15	0.65	6.48	0.04	0.43	0.0037	3.0	0.06	2.75E-05	2.74E-04
6 TO < 16	70	1.06	10.55	0.02	0.15	0.0037	3.0	0.14	2.40E-05	2.39E-04
16 TO 70	70	1.06	10.55	0.02	0.15	0.0037	1.0	0.77	4.32E-05	4.30E-04
TOTAL - LIFETIME								1	1.57E-04	1.57E-03
TOTAL - LIFETIME PER MILLION									157.2	1566.0

Analysis of California Poisoning Data

The California Poison Control System (CPCS) is a 24-hour emergency telephone consultation service providing evaluation of poisoning and treatment advice to health care professionals as well as the public. The CPCS provides approximately 325,000 case consultations per year. The CPCS staff generates a computerized chart for all consultations in accordance with the required criteria of the American Association of Poison Control Centers (AAPCC).

The CPCS conducted a search of its case management database for consultations involving products used for flea control. The search was conducted using Visual Dotlab, version 4.3.1 (WBM Software, Fresno, California). CPCS records for 1998–2007 (10 years) were searched for "flea" or "flee" in the substance field or the history field. All moderate, major, and death-outcome cases along with a sample of 40 of 472 minor-outcome cases were reviewed by the study principal investigator. Based on this review, 4,688 cases were retrieved from the CPCS database, 4,490 cases were included in the study, and 198 cases were deleted from the study for the following reasons: They involved an intentional exposure, did not involve a flea control product (e.g., the history contained the phrase "flee the scene" and was unrelated to a flea product), involved adverse reactions to a drug or food and therefore were inappropriate for study, or involved non-human patients (e.g., pets). CPCS staff estimated that there was a 2 per 40 (5 percent) contamination rate of the minor-effect cases with non-flea-product cases.

The MS Access database provided to NRDC by CPCS contained data on 4,490 cases, each assigned a unique ID number. NRDC conducted another round of review to remove cases not related to flea control products. Cases where the toxicant was coded as an animal bite, human drug or medication, food, and non-pesticide chemicals were reviewed carefully, and only those where the information in the text descriptor fields suggested an exposure to a flea control product were retained in the final file. Due to the common use of shampoos and cleaners for flea control, cases coded as cleaning agents were not removed. This screening process resulted in the removal of an additional 168 cases for a final tally of 4,322 cases attributed to flea control products. This data set was queried for those cases where the word *collar* was found in the text description fields (called mdx_desc or verbatimdesc in the database). A total of 287 cases met these criteria and were designated cases attributed to flea collars.

Descriptive statistics were generated for both of these data sets to include the following fields: age of the patient, route of exposure, and active ingredient of the product. Unfortunately, non-specific codes were used for many of the active ingredients, such as, "veterinary pesticide," "other insecticide," "unknown insecticide," or a blank field. In order to add specificity, two pesticide brand names that are known to contain only carbamates, Zodiac and Sargeant's flea collars, were queried in the text description fields. The resulting 42 cases were added to the carbamate category and removed from other categories. In addition, similar pesticide class categories were combined to generate larger categories.

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[21] Calculation based on the assumption that the surface area of one palm equals one-fourth of the surface area of both hands given in the EPA (1997) SOP.

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[24] Food Quality Protection Act (FQPA) PUBLIC LAW 104–170—AUG. 3, 1996

[25] NSRL (μg/day) = (10^{-5}× 70 (kg))/q* (1 × 10^{6} excess cancer/mg/kg-day).

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[27] The cancer potency factor is based on inhalation rather than dermal or ingestion exposure. However, it is standard risk assessment practice to use a default assumption that the cancer potency via various exposure pathways is equivalent for risk assessment purposes.

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[30] Dose rate (mg/day) = (residue concentration (mg/cm^2)) × (transfer coefficient (cm^2/hr)) × (time spent with pet (hrs/day))

[31] The EPA's estimate is based on a study of three children with pets. Other studies have indicated that people spend much more time with their pets, so this is likely to be an underestimate.

[32] OEHHA. Proposition 65 Interpretive Guideline No. 2008-001: Guideline for Hand-to-Mouth Transfer of Lead Through Exposure to Fishing Tackle Products. California Office of Environmental Health Hazard Assessment, March 2008.

[33] Intake (μg) = (L (μg/cm^2)) × (SA (cm^2)) × (f) × (× (λcontacts/hr)) × (t (hrs)).