

along marine and freshwater shorelines of King County, Washington. Over 3000 unique respondents were interviewed for their recreational or fish consumption preferences. Exposure distributions (e.g., event time and event frequency) were developed for fifteen unique recreational activities and for seafood consumption rates. The mean consumption rates for consumers of marine fish and shellfish and freshwater fish were 32, 22, and 10 grams/day, respectively. Results indicate that the consumption patterns of marine anglers from King County have remained consistent since the mid-1980s. Recreational activity and freshwater fish consumption information was previously unavailable for King County, WA and these surveys provided site-specific data for use in defining exposure. The survey results were incorporated into the exposure models of risk assessments (marine and freshwater) that were used in watershed planning activities in King County. In addition, the freshwater fish consumption survey results have been incorporated into a fish consumption advisory for Lake Washington, Seattle. These results provide current recreational and fish consumption exposure data for the general population living in King County, WA.

## 860 ESTIMATING SOURCE CONTRIBUTION INSIDE SCHOOL BUSES

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Many studies identifying levels of diesel-related air pollutants inside school buses have been reported, some claim to have been able to derive quantitative estimates of "self-pollution", i.e., source contribution from the engine powering the bus. In-cabin exposure to crankcase emissions of lube-oil products released from road draft tubes located inside the engine compartment (non-tail pipe emissions) have also been reported. Reports of exposure to student passengers from source contribution have resulted in heightened concern among parents, school officials, and regulatory agencies. The reliability and accuracy of quantitative estimates of source contribution is dependent upon the approach and methodology employed to measure source contribution.

To compare the results of prior source contribution studies, we conducted a study using three distinct methodologies to measure source contribution in two school buses operating on two school bus routes. Methods used in this study include an explicit fuel-borne tracer (iridium); measured DPM-related constituents; and a deuterated-alkane tracer placed into the lube oil. In addition to these methods, an on-board dilution tunnel was used to collect exhaust samples to derive estimates of DPM emission rates over an actual route. This study is unique in that it:

- (1) Used three separate methods to evaluate potential source contribution inside the same school bus;
- (2) Will provide quantitative estimates of potential exposure to non-tailpipe crankcase emissions; and
- (3) Will estimate DPM source contribution based on actual on-road measurements of DPM and crankcase emissions.

Samples are undergoing analyses and anticipated to be complete in January.

## 861 PYRETHROIDS EXPOSURE IN THE CANADIAN POPULATION: A CASE STUDY IN MONTREAL.

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Pyrethroids are widely used in agriculture, horticulture, in and around the house and for the treatment of head lice and fleas. The general population is therefore ubiquitously exposed to those pesticides. The aim of the present study was to determine the extent of pyrethroid exposure in an urban and suburban population such as that of the Montreal area through measurements of urinary metabolites. This study was approved by the ethics board on human research of the Université de Montréal. A total of 120 adults (43 males and 77 females) aged between 18-64 y.o. and of 120 children (60 males and 60 females) aged between 6-12 y.o. were randomly recruited by phone by a survey company. Their admissibility was verified and personal information was gathered. Adults were asked to provide a 24-h urine collection and children a (more or less) 12-h collection. They were also asked to fill out a questionnaire. The urines were kept in the refrigerator at all times by the participants and, once at the laboratory, frozen until the analysis. The urines were analyzed by HPLC-MS-MS for the following specific and semi-specific metabolites: 4-fluoro-3-phenoxybenzoic acid, *cis*-3-(2,2-dibromovinyl)-2,2-dimethylcyclopropane carboxylic acid, 3-phenoxybenzoic acid and *cis* and *trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane carboxylic acid. Roughly 80% of adults and children com-

pleted the study. The percentile distribution and geometric mean of metabolite concentrations were determined for each group. The results were similar to those obtained by the CDC ranging from below the detection limit to 3.25 µg/L. Based on the questionnaire data, the most common source of exposure appeared to be contaminated food as all the other sources, including residential use, were rarely reported in the questionnaires. The present case study provides greater insights for the management of pesticides exposure by Canadian public health authorities.

## 862

## ASSESSING EXPOSURE LEVELS OF CHILDREN TO FLEA CONTROL INSECTICIDES (CHLORPYRIFOS, TETRACHLORVINPHOS, AND PERMETHRIN) FROM THE FUR OF DOGS

M. K. Davis, M. Russak, J. W. Tyler, J. S. Boone, M. K. Ross and J. E. Chambers. Center for Environmental Health Sciences, Mississippi State University, Mississippi State, MS.

Organophosphorus and pyrethroid insecticides have been extensively used for control of fleas and other pests on pets, leading to potential exposures of children. In the present study, pet dogs were treated with over-the-counter collars containing chlorpyrifos (CP) or tetrachlorvinphos (TCVP), or a spot treatment with permethrin (PER). Dislodgable insecticide residues were quantified on cotton gloves used to pet the dogs for 5 minutes, and on cotton t-shirts worn by a child for a 4-hour (average) period at selected time points after insecticide application. First morning urine samples were also obtained from adults and children for metabolite quantification. Twenty-four dogs were used for the CP and TCVP treatments, and 15 dogs were used for the PER treatment. Transferable residues for all compounds were highest near the neck of the dogs and were lowest in areas most distant from the neck. During a 5-minute rubbing on the third week after application, the average amounts of CP and TCVP transferred from the fur of the neck (rubbing over the collar) to a glove were 469 ± 83 and 12,957 ± 2029 mg/glove, respectively, and the average amounts of CP and TCVP transferred from the fur of the back to a glove were 8 ± 2 and 81 ± 26 mg/glove, respectively. The average amount of PER transferred to a glove from the fur of the neck (location of treatment) 4 hours post application was 44,746 ± 19,314 mg/glove while the average amount transferred at 14 days post application was 991 ± 299 mg/glove. T-shirts worn by the child for 4 hours (average) on the day following application showed levels in ng/g shirt of 156 ± 83, 1678 ± 892, and 8297 ± 3421 for CP, TCVP, and PER, respectively. There were no significant differences between adults and children in the levels of urinary metabolites of CP; however, children typically had somewhat higher urinary levels of metabolites than adults. (Supported by EPA R828017)

## 863 DDE LEVELS BEFORE AND DURING HUMAN PREGNANCY: A LONGITUDINAL STUDY

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Background: p,p'-DDE (DDE) is a toxic metabolite of the insecticide DDT, persisting in soil and fatty tissue, and long outlasting active use. Mexico prohibited DDT in 1998 after decades of use. Some studies show DDE in pregnant women or in maternal milk damages child development.

Objective: Develop models describing change in maternal DDE from before pregnancy through the third trimester to estimate timing and sources that could influence fetal exposure. Methods: We recruited non-pregnant women planning pregnancy from one urban and three farming areas in Morelos State, Mexico during legally-required prenatal counseling after they signed an IRB-approved informed consent. We analyzed blood from the pre-nuptial session and from each trimester of pregnancy for wet basis of serum DDE and p,p'-DDT by electron capture gas-liquid chromatography. Questionnaires assessed sociodemographic data, dietary habits, and changes in maternal weight before and during pregnancy. We modeled change in DDE over the four time periods using mixed models. Results: 297 women supplied data. Geometric mean DDE was 11.1 ppb (range: 0.05–185.1). Only one DDE sample was found at the detection limit. 757 DDT samples from the same women were measured at the detection limit (0.005 ppb). Third trimester DDE was up to 20% higher (p<0.04) than in prior phases. Urban women had up to 1.2 times less DDE (p<0.0005) than women in farming areas. Women with prior breast-feeding history had 45% lower DDE than women without. Each doubling of age and weight was associated with a 63% (p=0.001) increase and a 55% decrease (p=0.003) in DDE, respectively, through all phases. Self-reported diet was not significantly related to DDE. Conclusions: Measurable levels of DDE remain in pregnant women years after DDT use stopped. Older women living in agricultural areas have higher levels, especially during the third trimester. Diet may no longer be a source of DDT residues in this population, though this apparent absence requires continued vigilance.

## **NCER Assistance Agreement Final Report Executive Summary**

**Date of Final Report:** August 28, 2006

**EPA Agreement Number:** R828017

**Title:** Assessing levels of intermittent exposures of children to flea control insecticides from the fur of dogs.

**Investigators:** Janice E. Chambers, J. Scott Boone (partial), John W. Tyler (partial), Carolyn R. Boyle, Mark Russak (partial)

**Institution:** Mississippi State University

**Research Category:**

**Project Period:** 4/10/2000 - 4/9/2005

**Description and Objective of Research:** The majority of insecticide residues reported are from drinking water, food, surfaces and dust. Information has not been available for determining the levels of insecticide from topical flea control products on pet dogs that might be dislodged and transferred to people, especially children. These dermal exposures could easily become oral exposures when children place their contaminated hands in their mouths.

Organophosphorus insecticides or synthetic pyrethroids are among the most common types of insecticides used for flea control with over-the-counter products. Our calculations from a previous study, from dislodgable residues on white cotton gloves used to pet the dogs, estimated that transfer of these dislodgable residues could result in exposure levels exceeding the adult reference dose ( $R_fD$ ), which does not account for the greater sensitivity of children. There are a very large number of dog-owning households in the United States (about 37%) and about half of pet-owning households have children in them. Consequently, the opportunity for large numbers of children to contact flea control insecticides on pets is high. Because of this lack of information and the likelihood of appreciable insecticide residues being present on dog fur, this project was designed to determine the dislodgable residues from dog fur onto white cotton gloves (obtained by a 5 minute petting of the dog on a defined area of the fur) from flea control collars containing either chlorpyrifos or tetrachlorvinphos (TCVP) or permethrin from a spot-on treatment. Additionally, the residues on a white cotton tee shirt worn for 4 hours by a child in the household of the dog were obtained, as were the insecticide metabolites in the urine of these children and also adults in the same household.

**Summary of Findings:** Our calculations from a previous study indicated the time at which peak dislodgable residues would be obtained from the chlorpyrifos collar product, and they also indicated that dislodgable residues were similar

throughout the sampling period after about 2 weeks following placement of the collar. Therefore this study took samples during the first 3 weeks after collar placement. Results indicated about 1 µg chlorpyrifos on the gloves prior to collar placement, and average residues of about 8, 317 and 469 µg on the gloves used to pet the back, the neck and the neck over the collar. Residues on tee shirts were significantly greater after the collar was placed on the dog than before. Because chlorpyrifos is used widely, the urinary metabolite TCP routinely is observed in much of the population and TCP was present in the urine of both the adults and the children at about 8 and 13 ng/ml, respectively. There were slight increases in urinary TCP in both adults and children after placement of the collar, but these were not statistically significant compared to the pretreatment values. Few statistical correlations were found between time of contact with the dog, fur length, fur density, glove residues, tee shirt residues and urinary TCP levels. A moderate correlation was found between tee shirt residues and urinary TCP (non-creatinine adjusted) in children.

Previous studies with a TCVP collar indicated that the peak time for dislodgable residues from the dog's fur was 7 days with a decline after that. Therefore in this study, samples were taken between days 5 and 12 post-treatment. There were no residues of TCVP from the fur or the tee shirts prior to the placement of the collars. The TCVP collar yielded substantial residues on the gloves following placement of the collars on the dogs, approximately 22,000 µg/glove for over the collar samples. There was a 30% decline in detectable TCVP residues obtained from the over the collar samples during the 7 days of the test interval. The residues from the back of the dog were considerably lower, about 80 µg/ glove, and did not vary appreciably during the test interval. There were no TCVP residues on the tee shirts prior to placement of the collar. A wide range of tee shirt residues were obtained during the course of the study, from about 5 to 6500 ng/g shirt. There were no residues of the metabolite trichloromandelic acid in the urine of either adults or children in the pre-treatment samples. Following placement of the collar, maximum metabolite levels were observed on day 11 post-treatment for children (about 200 ng/ml) and on day 12 for adults (about 100 ng/ml).

Transferable residues of permethrin from the spot-on treatment over the region to which the treatment had been applied were highest at 4 hours after application (about 160,000 µg/glove) and these residues declined 99% by 20 days post treatment. All of the observed tee shirt residues were significantly greater than pre-treatment levels and decreased throughout the duration of the study. Urinary metabolite levels are not finalized at the time of this report.

In all cases, the data were highly variable, reflecting the diversity of individual dogs, children and adults in their contact and behavior with the dogs, and the variability of the sampling.

**Conclusions:** Overall variability among all of these studies was high, and was expected because of the use of a variety of dogs, families and samplers. This information should be informative to any probabilistic risk assessment calculations. Different patterns were observed with the two different collars, with the chlorpyrifos collar yielding a more consistent pattern of dislodgable residues and urinary metabolites over time, and the TCVP collar yielding a peak with a reduction of residues over time. Children had higher urinary levels of the metabolites of chlorpyrifos and TCVP than the adults tested.

Because of the widespread use of chlorpyrifos, people had the chlorpyrifos metabolite, TCP, in their urine prior to the placement of the flea collar on the dog. There was a slight but not statistically significant increase in urinary TCP after placement of the collar. If this increase is real, this seems to be a trivial amount compared to the baseline levels of urinary TCP. Although dislodgable residues of chlorpyrifos were observed in both gloves used to rub the dogs and in tee shirts worn by the children, these residues did not seem to result in appreciable increases in urinary TCP, and suggests that chlorpyrifos is not absorbed well following this type of exposure. Therefore it appears that exposure to chlorpyrifos from use of this chlorpyrifos-containing flea collar on dogs adds little, if any, to the overall chlorpyrifos exposure level of either adults or children.

Because TCVP is not used widely, people did not have residues of it or its metabolite prior to the collar placement. High levels of dislodgable residues were obtained shortly after the collar placement, and these dislodgable residues declined quickly. However, because there is no baseline occurrence of this compound or its metabolite, the relevance of the presence of TCVP in dislodgable residues or speculations about how readily TCVP is absorbed cannot be made from these data. However, this source of exposure should be taken into account during aggregate and cumulative risk assessments.

Relatively high amounts of permethrin occur as dislodgable residues shortly after the placement of the spot-on treatment. However, these levels dissipate substantially relatively quickly. Because the urinary metabolite data are incomplete at this time, conclusions cannot be drawn yet.

Overall, it appears that over-the-counter flea control remedies can result in dislodgable residues that have the potential to cause exposure of people. The likelihood of this resulting in internalization of the insecticide does not appear to be consistent among products. The exposure resulting from flea control products should be taken into account in aggregate and cumulative risk assessments. While there is insufficient information from these studies to accurately assess the actual exposure levels from these products, it is probably wise to exercise caution in contacting pets recently treated with similar products because apparently high levels of dislodgable residues occurred shortly after product application on some of the dogs.

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**Supplemental Keywords:** flea and tick collars, residential exposure, companion animal parasite control, exposure assessment

**Relevant Web Sites:** <http://www.cvm.msstate.edu/cehs/index.htm>



Journal List > Environ Health Perspect > v.109(11); Nov 2001

Environ Health Perspect. 2001 November; 109(11): 1109–1114.

PMCID: PMC1240470

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### Research Article

## Transferable residues from dog fur and plasma cholinesterase inhibition in dogs treated with a flea control dip containing chlorpyrifos.

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### Abstract

We studied chlorpyrifos, an insecticide present in a commercial dip for treating ectoparasites in dogs, to estimate the amount of transferable residues that children could obtain from their treated pets. Although the chlorpyrifos dip is no longer supported by the manufacturer, the methodology described herein can help determine transferable residues from other flea control insecticide formulations. Twelve dogs of different breeds and weights were dipped using the recommended guidelines with a commercial, nonprescription chlorpyrifos flea dip for 4 consecutive treatments at 3-week intervals (nonshampoo protocol) and another 12 dogs were dipped with shampooing between dips (shampoo protocol). The samples collected at 4 hr and 7, 14, and 21 days after treatment in the nonshampoo protocol averaged 971, 157, 70, and 26 microg chlorpyrifos, respectively; in the shampoo protocol the samples averaged 459, 49, 15, and 10 microg, respectively. The highest single sample was about 7,000 microg collected at 4 hr. The pretreatment specific activities in the plasma of the dogs were about 75 nmol/min/mg protein for butyrylcholinesterase (BChE), and 9 nmol/min/mg protein for acetylcholinesterase (AChE). BChE was inhibited 50-75% throughout the study, and AChE was inhibited 11-18% in the nonshampoo protocol; inhibition was not as great in the shampoo protocol. There was no correlation ( $p < 0.05$ ) between length of hair and residues measured that would explain the residue differences among dogs. Transferable residues had largely dissipated during the three weeks after treatment, with the largest decrease occurring during the first week. Greater plasma ChE inhibition was observed at 7 days than at 4 hr, probably reflecting the bioactivation of chlorpyrifos to chlorpyrifos-oxon. Plasma cholinesterase activity did not return to control levels during the 3-week period. The differences between the shampoo and nonshampoo protocols were explained by differences in the techniques of the dip applicators.

### Full Text

The Full Text of this article is available as a PDF (516K).

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1: J Expo Sci Environ Epidemiol. 2007 Nov;17(7):656-66. Epub 2007 Mar 28.

Links

**Assessing transferable residues from intermittent exposure to flea control collars containing the organophosphate insecticide chlorpyrifos.**

**Chambers JE, Boone JS, Davis MK, Moran JE, Tyler JW.**

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Children can be exposed to pesticides from numerous residential sources such as carpet, house dust, toys and clothing from treated homes, and flea control remedies on pets. In the present studies, 48 pet dogs (24 in each of two studies) of different breeds and weights were treated with over-the-counter flea collars containing chlorpyrifos (CP), an organophosphorus insecticide. Transferable insecticide residues were quantified on cotton gloves used to rub the dogs for 5 min and on cotton tee shirts worn by a child (Study 2 only). First morning urine samples were also obtained from adults and children in both studies for metabolite (3,5,6-trichloro-2-pyridinol) quantification. Blood samples were obtained from treated dogs in Study 1 and plasma cholinesterase (ChE) activity was monitored. Transferable residues on gloves for all compounds were highest near the neck of the dogs and were lowest in areas most distant from the neck. Rubbing samples (over the collar) at two weeks post-collar application contained 447+/-57 microg CP/glove while samples from the fur of the back contained 8+/-2 microg CP/glove. In Study 2, cotton tee shirts worn by children at 15 days post-collar application for 4 h showed CP levels of 134+/-66 ng/g shirt. There were significant differences between adults and children in the levels of urinary metabolites with children generally having higher urinary levels of metabolites than adults (grand mean+/-SE; 11.6+/-1.1 and 7.9+/-0.74 ng/mg creatinine for children and adults, respectively, compared to 9.4+/-0.8 and 6.9+/-0.5 ng/mg creatinine before collar placement). Therefore, there was little evidence that the use of this flea collar contributed to enhanced CP exposure of either children or adults.

PMID: 17392689 [PubMed - indexed for MEDLINE]

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- An observational study of the potential for human exposures to pet-borne diazinon residues following flea collar use. [J Expo Sci Environ Epidemiol. 2006]
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## Transferable Residues From Dog Fur and Plasma Cholinesterase Inhibition in Dogs Treated with a Flea Control Dip Containing Chlorpyrifos

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We studied chlorpyrifos, an insecticide present in a commercial dip for treating ectoparasites in dogs, to estimate the amount of transferable residues that children could obtain from their treated pets. Although the chlorpyrifos dip is no longer supported by the manufacturer, the methodology described herein can help determine transferable residues from other flea control insecticide formulations. Twelve dogs of different breeds and weights were dipped using the recommended guidelines with a commercial, nonprescription chlorpyrifos flea dip for 4 consecutive treatments at 3-week intervals (nonshampoo protocol) and another 12 dogs were dipped with shampooing between dips (shampoo protocol). The samples collected at 4 hr and 7, 14, and 21 days after treatment in the nonshampoo protocol averaged 971, 157, 70, and 26  $\mu\text{g}$  chlorpyrifos, respectively; in the shampoo protocol the samples averaged 459, 49, 15, and 10  $\mu\text{g}$ , respectively. The highest single sample was about 7,000  $\mu\text{g}$  collected at 4 hr. The pretreatment specific activities in the plasma of the dogs were about 75 nmol/min/mg protein for butyrylcholinesterase (BChE), and 9 nmol/min/mg protein for acetylcholinesterase (AChE). BChE was inhibited 50–75% throughout the study, and AChE was inhibited 11–18% in the nonshampoo protocol; inhibition was not as great in the shampoo protocol. There was no correlation ( $p \leq 0.05$ ) between length of hair and residues measured that would explain the residue differences among dogs. Transferable residues had largely dissipated during the three weeks after treatment, with the largest decrease occurring during the first week. Greater plasma ChE inhibition was observed at 7 days than at 4 hr, probably reflecting the bioactivation of chlorpyrifos to chlorpyrifos-oxon. Plasma cholinesterase activity did not return to control levels during the 3-week period. The differences between the shampoo and nonshampoo protocols were explained by differences in the techniques of the dip applicators. **Key words:** acetylcholinesterase, butyrylcholinesterase, chlorpyrifos, dog fur, flea control, organophosphate insecticide, pesticide monitoring, transferable residues. *Environ Health Perspect* 109:1109–1114 (2001). [Online 19 October 2001] <http://ehpnet1.niehs.nih.gov/docs/2001/109p1109-1114boone/abstract.html>

Exposure of children to pesticides is a major health concern. Children could be exposed to pesticides from carpet, house dust, and toys from treated houses, from clothing of parents who are farm workers and pesticide applicators, and from playing outside in treated lawns and gardens (1–5). One overlooked but important potential source for pesticide exposure to children is pets treated with parasite control products. These products often contain carbamate, pyrethroid, and organophosphate insecticides.

Organophosphate (OP) compounds have been used commonly as insecticides in and around households, on farm and domestic animals, and on agricultural lands. These compounds are used widely in the United States because of their relatively low mammalian toxicity, their short half-lives, and their ease of use. These insecticides have been used residentially for the control of termites, ants, roaches, ticks, fleas, and other insect and arachnid pests. Thus, there is increased opportunity for children to be exposed by multiple routes, and these aggregate exposures could contribute to significant toxicity. Identification of all possible routes of exposure and the quantification of the magnitude of these exposures may

contribute to a more accurate calculation of pesticide risk, and thereby decrease the reliance on uncertain default assumptions in risk assessment.

The amount of exposure data to OP insecticides for children is limited. Most of the risk calculations are for adults and may not consider the potentially greater absorption and sensitivity of children, poorer personal hygiene, potentially lower capacity for detoxication, developing organ systems, and a greater body surface area to volume ratio. Human potential exposures to organophosphate compounds have been documented from several sources, such as carpeting and household dust (2). Children have also been poisoned by OP insecticides through exposure to contaminated items, such as bed linens, clothing, and burlap sacks (3–5). However, the amount of exposure to organophosphate compounds from flea and tick control products on pets, such as dogs and cats, has not been documented.

A demographic survey of companion animals by the American Veterinary Medical Association (6) indicated that in 1991 approximately 34.6 million households (36.5%) in the United States owned a dog or dogs, a number essentially unchanged

from 1987. More households had dogs as pets than other types of animals. There was a mean of 1.52 dogs per dog-owning household, yielding an estimated national population of 52.5 million dogs. Fifty percent of pet-owning households were parental households with children; in comparison, 40% of total households have children. The authors of that study projected that there would be 53.6 million dogs in the United States in 1998 (7). These dogs could be a source of exposure to millions of children who live in the same environment and come into direct contact with dogs treated with flea control products. Also, millions of cats and other pets or domestic animals, such as horses or cows, are also treated for insect pests and could serve as additional sources of intermittent insecticide exposure to children. The control of fleas and ticks on pets is a high priority for pet owners. Dips, flea and tick collars, treatment of yards with powders, sprays, and granular forms of insecticides, and the use of sprays and foggers in the home are all insecticide methods used by pet owners to control pests associated with pets.

Chlorpyrifos is one of the most commonly used organophosphate insecticides in the United States. This broad-spectrum insecticide effectively controls a wide variety of insects, primarily as a contact poison, and is used in flea control products such as flea collars, sprays, and, until recently, as a dip. When this study was conducted, chlorpyrifos

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The authors thank H.D. Canrwell for the use of the animal facilities, S. Waldrop and T. Couch for the cholinesterase assays, K. Lebbin-Hanson for the chemical analyses, C. Boyle for statistical advice, C. Langston for advice on kinetic calculations, and C. Bates, C. Buisson, D. Dunaway, T. Kothstein, A. Smith, and C. Zickus for treatment and sampling of the dogs.

The research was supported by a U.S. Environmental Protection Agency grant, EPA R-825170-01-0, and by the Mississippi Agriculture and Forestry Experiment Station (MAFES) under project number MISV-3403 and the College of Veterinary Medicine, Mississippi State University. This article is MAFES publication number J-9617 and Center for Environmental Health Sciences publication number 102.

Received 17 November 1999; accepted 11 April 2001.

was used in dip products, but this formulation has been withdrawn by the manufacturer.

This study was designed to determine the amount of chlorpyrifos that could be transferred to humans from dogs treated with a flea and tick dip. We quantified transferable residues from cotton gloves, which were used to rub the fur of the back of the dogs for 5 min according to a standard protocol and at set intervals after application of the dip. We investigated the time course of dissipation of these residues after initial treatment, considered the influence of different fur lengths on transferable residues, and studied the effects of bathing the dogs between dippings. We also determined the amount of plasma cholinesterase inhibition in the dog as a biomarker of insecticide absorption and persistence within the dog.

We also performed a test (reproducibility test) to determine whether rubbing with a cotton glove removed a significant amount of pesticide from the dog, which would bias subsequent samples. The reproducibility test determined residues after 7 days on two locations after an initial residue test on only one location at 4 hr.

## Methods

**Chemicals.** We obtained the dip, shampoo, and eye drops from a veterinary supply company. The Adams Flea and Tick Dip (chlorpyrifos 3.84%, 2 oz/gal, SmithKline Beecham Animal Health) and the shampoo (Mycodex Pearlescent) were manufactured by SmithKline Beecham (West Chester, PA). The eye drops (Akwa Tears) were manufactured by Akorn, Inc. (Abita Springs, LA). All solvents used for the pre-extraction and extraction of the gloves were pesticide grade. The chlorpyrifos standard was obtained from Chem Service and was greater than 99% pure. All other reagents were purchased from Sigma Chemical Company (St. Louis, MO).

**Animals.** Care and use of the dogs were in accordance with the *Guide for the Care and Use of Laboratory Animals* (8) in a facility accredited by the Association for Assessment and Accreditation of Laboratory Animal Care, overseen by J.E. Harkness, University Laboratory Animal Veterinarian, Office of Research, Mississippi State University. The collection of blood, collection of transferable residues, and the dipping of the dogs occurred in rooms in the College of Veterinary Medicine Animal Health Center. The dogs were monitored by the directing veterinarian of the project (J. Tyler) and veterinary students. Dogs were selected from pet owners at the College of Veterinary Medicine, Mississippi State University, and met the following criteria: They had had no known organophosphate compound exposure one month before

inception of the project; the dog must weigh > 10 lbs; and the owner must comply with the experimental protocol, including housing the dog at the College of Veterinary Medicine when needed and not using any other organophosphate compound during the study. Dogs used in the study are described in Table 1. Dogs were tested in two groups of six for each protocol because of space and time constraints. The dogs represent many sizes, ages, and inside/outside conditions. There were 12 replicates (individual dogs) for each of the two protocols.

**Protocols.** We employed two protocols in this study, one not involving shampooing between dippings (nonshampoo protocol) and the second involving shampooing immediately before each dipping (shampoo protocol). We designed the latter protocol to determine whether intermediate shampooing would reduce potential accumulation of residue levels.

**Petting.** The dogs were rubbed in anterior and posterior directions (back and forth motions) in a firm pattern but not one that would cause discomfort to the dog. The dogs seemed to enjoy the attention and petting. The dogs were petted in a 10 × 4 inch area marked with a readily removable adhesive tape on the back just caudal to the neck for 5 min (fur samples) with 100% cotton gloves that had been precleaned. The gloves were inverted when removed and placed into a clean, labeled glass jar.

**Collection of samples.** Handlers who dipped the dogs wore eye protection, shoulder-length rubber gloves, and a rubber-coated cotton apron. Dogs were bathed one day before the start of the study and were dipped according to the directions on the product label. Eye drops were placed into the eyes of the dogs before dipping. The dip was diluted to 2 oz/gal and sponged over the dog until the undercoat was wet (approximately 2 min). A fresh dip was prepared for each subsequent dog and dipping; each dog's coat was saturated with the dip, and no dip solution was reused. The dogs were placed in cages and dried with cage dryers on high fan setting with low heat (approximately 2 hr for complete drying).

For the nonshampoo protocol, we collected fur samples before dipping (0 hr), 4 hr after dipping, and at 1, 7, 14, and 21 days after the dip. At 21 days the dogs were sampled (21-day sample), dipped, dried, and sampled 4 hr after the dipping (4-hr sample) and at 7, 14, and 21 days later. This process was completed 2 more times, so there were four sequential dippings per replicate. We collected blood samples in evacuated blood collection tubes with no anticoagulant. Blood samples, taken at the same time as fur samples, were centrifuged after collection and stored at 4°C overnight, and cholinesterase activity was determined within 24 hr after collection.

For the shampoo protocol, we collected the fur samples before dipping (0 hr), at 4 hr

**Table 1.** Dogs used in the transferable residue and cholinesterase experiments, including breed, weight, fur length, and percent change from average within protocol at 4 hr.

Dog, <sup>a</sup> breed (sex)	Weight (lbs)	Fur length	Percent of average <sup>b</sup>
<b>Nonshampoo protocol</b>			
1 German shepherd (F)	77.2	medium	47.39
2 Labrador x Greyhound (F)	48.5	short	135.74
3 Mix (F)	19.0	long	57.14
4 Labrador x Mix (M)	62.3	long	69.31
5 Boston terrier x Mix (F)	22.5	short	62.99
6 Border collie x Greyhound (F)	44.5	medium	89.62
7 Rottweiler (F)	43.0	short	65.17
8 Labrador (F)	74.3	short	301.77
9 Mix (F)	23.1	long	78.90
10 Rottweiler x Mix (M)	73.3	short	109.74
11 Mix (F)	18.7	short	58.09
12 Rottweiler x Mix (F)	72.5	short	124.15
<b>Shampoo protocol</b>			
13 Rottweiler (F)	107.1	short	199.29
14 Rottweiler (F)	109.5	short	175.32
15 Border collie (M)	55.5	long	22.25
16 Mix (F)	33.3	short	26.13
17 Labrador (F)	73.5	short	47.46
18 Boxer (M)	86.1	short	46.46
19 Mix (M)	48.0	short	167.78
20 Schnauzer x Poodle (F)	13.4	med/long	68.73
21 Spitz x Poodle (M)	13.2	long	52.14
22 Weimaraner (F)	85.0	short	147.64
23 Weimaraner (M)	88.0	short	135.50
24 Husky x Mix (F)	33.9	long	111.30

Abbreviations: F, female; M, male; med, medium; Mix, mixed breed; x, cross breed.

<sup>a</sup>Dogs identified by number. <sup>b</sup>This number represents the percent of the average over four dippings at 4 hr from each protocol shown in Tables 2 and 3. For example, dog 1 averaged 47% of the total average (1,229 µg), or 578 µg over the four dippings at 4 hr.

and 1, 7, 14, and 20 days after dip, and the day after shampooing (21 days). The dogs were shampooed the day before each dipping, fur samples were taken on day 20, and the dogs were shampooed and allowed to dry overnight at home. At 21 days the dogs were sampled (to check for shampoo removal of chlorpyrifos), dipped, dried, and sampled at 4 hr after dip. The sampling was repeated (4 hr and 7, 14, 20 and 21 days) for a total of four dips. Blood samples were collected at the same time points as the fur samples, except a postshampoo blood sample between dips was not taken (day 21).

**Reproducibility test.** We developed this test to check collection of samples to make sure the decrease seen in the levels of chlorpyrifos over time was caused by the dissipation and not removal by the rubbing; we conducted two tests. In this study, 4 dogs were dipped, dried, and then sampled along the dorsal cervicothoracic spine (back/neck) in a 40-square-inch area at 4 hr as described previously. At 7 days the dogs were again petted along the dorsal cervicothoracic spine (back/neck), the same area as the 4-hr sample. Also at 7 days an additional fur sample was obtained from the fur along the dorsal lumbosacral region (back/tail) in a 40-square-inch area for comparison. If the sample from the dorsal cervicothoracic spine region at 7 days was not significantly different from that of the dorsal lumbosacral region at 7 days, this would indicate that the rubbing did not cause the decrease over time; the 4-hr rubbing had no significant effect on the sample collection at subsequent times.

In the reproducibility test, there was no significant difference (general linear model, least significant difference,  $p \leq 0.05$ ) between the back/neck sample and the back/tail sample at 7 days (back/neck,  $93 \pm 12 \mu\text{g}$ ; back/tail,  $92 \pm 10 \mu\text{g}$ ); samples at 4 hr had originally been  $1,113 \pm 195 \mu\text{g}$  at the back/neck area.

**Determination of chlorpyrifos on gloves.** The gloves used for rubbing were 100% cotton. They were washed once with laundry detergent, three times without detergent, pre-extracted for 8 hr with methylene chloride, dried, and stored in glass jars washed and prerinsed three times with water, acetone, and petroleum ether. After sampling, the gloves were extracted with petroleum ether using an Accelerated Solvent Extractor (ASE) by Dionex (Sunnyvale, CA). The operating conditions were heat for 5 min at  $100^\circ\text{C}$  and 1,500 psi; static for 2 min; flush 50% of volume; static for 2 minutes; purge with nitrogen for 150 sec; and a final purge for 60 sec. For every 20 samples three additional gloves were spiked with 20  $\mu\text{L}$  hexane, 10  $\mu\text{g}$  chlorpyrifos, or 1,000  $\mu\text{g}$  chlorpyrifos

at the time of sampling and stored and extracted with the samples.

During method development, we quantified gloves from dogs and applied various concentrations from 1 to 5,000  $\mu\text{g}$  of chlorpyrifos to different gloves to check for recovery rates and extraction parameters. All spiked gloves yielded 90–100% recovery. After collection, the sampling gloves and the spiked gloves were stored at  $4^\circ\text{C}$  until extraction. After extraction by the ASE, the extract was evaporated and transferred to hexane under a nitrogen stream using an N-EVAP (1–3 mL; Organomation Associates, Inc., Northborough, MA), and then was transferred to graduated test tubes, and the volume was adjusted to 10 mL. We analyzed the extract by gas chromatography/electron capture detection using an Rtx-5 Amine column (30 m, 0.53 mm ID, 1.0  $\mu\text{m}$  df; Restek Corporation, Bellefonte, PA). Injector temperature was  $280^\circ\text{C}$ ; oven temperature was  $180\text{--}200^\circ\text{C}$  (ramp 2 degrees/min for 5 min); and detector temperature was  $325^\circ\text{C}$ . The recovery rate for the spiked gloves throughout the study was 90–100%. The level of detection (LOD) and level of quantitation (LOQ) were 0.6  $\mu\text{g}/\mu\text{L}$  and 2.7  $\mu\text{g}/\mu\text{L}$  for 1  $\mu\text{L}$  injections. The LOD and LOQ were determined by the methods described by Taylor (9), and we used spiked gloves.

**Cholinesterase assay.** The cholinesterase (ChE) assay was a modification of the procedure described in Chambers and Chambers (10), which is based on Ellman et al. (11). We determined cholinesterase by four different means using various combinations of inhibitors and substrates to investigate plasma enzymes. We diluted the plasma in

0.05 M Tris-HCl buffer, pH 7.4 at  $37^\circ\text{C}$ . We used 14 test tubes for each plasma sample. Each test tube contained 0.05 M Tris-HCl buffer, pH 7.4 at  $37^\circ\text{C}$ , and the plasma sample. We added eserine sulfate to four of the test tubes to determine noncholinesterase-mediated hydrolysis of acetylthiocholine (ATCh) iodide. To four of the test tubes we added tetraisopropyl pyrophosphoramidate (iso-OMPA) to determine nonbutyrylcholinesterase (BChE)-mediated hydrolysis of butyrylthiocholine (BTCh) iodide. We added ATCh iodide (final concentration 1 mM) to seven of the tubes, which were vortexed and returned to a  $37^\circ\text{C}$  shaking water bath for 15 min. The seven test tubes comprised 3 for uninhibited assays, 2 with eserine sulfate, and 2 with iso-OMPA. We assayed a parallel set of tubes with BTCh iodide (final concentration 1 mM). We added sodium dodecyl sulfate (SDS) and 5,5'-dithio-bis(2-nitrobenzoic acid) (DTNB) (final concentrations of 0.44% and 0.02%, respectively) to each tube to stop the reaction (SDS) and for the chromogen (DTNB) to react with the thiocholine released from ATCh or BTCh hydrolysis; tubes were vortexed and the absorbance was read at 412 nm. We determined acetylcholinesterase (AChE) activity by subtracting the eserine sulfate readings from the iso-OMPA readings in the acetylthiocholine iodide set. We determined total ChE activity by subtracting the eserine sulfate readings from the uninhibited readings in the ATCh set. We determined butyrylcholinesterase activity with ATCh by subtracting the iso-OMPA readings from the uninhibited readings in the ATCh set. We

**Table 2.** Transferable residues of chlorpyrifos from fur after four consecutive treatments of dogs with flea control dip during the nonshampoo protocol.

Time	Amount on glove ( $\mu\text{g}$ ) <sup>a</sup>	95% Confidence intervals		Range	
		Lower	Upper	Lower	Upper
Pretreatment	0.1583	0.0697	0.3600	< 0.03	0.73
4 hr	971.18	491.93	1917.34	157	6,999
7 days	157.00	79.52	309.96	4	2,584
14 days	70.11	35.51	138.39	1	2,472
21 days	26.63	13.49	52.57	1	1,469

<sup>a</sup>Geometric means are significantly different at  $p \leq 0.05$ . Each point is the average of 12 dogs (replicates) over all of the four treatments.

**Table 3.** Transferable residues of chlorpyrifos from fur after four consecutive treatments of dogs with flea control dips during the shampoo protocol.

Time	Amount on glove ( $\mu\text{g}$ ) <sup>a</sup>	95% Confidence intervals		Range	
		Lower	Upper	Lower	Upper
Pretreatment	0.1079	0.0560	0.2078	< 0.03	0.97
4 hr	458.78	278.93	754.40	17	2,674
7 days	49.26	29.96	81.02	9	479
14 days	14.86	9.03	24.48	1	190
20 days	9.62	5.84	15.83	1	130
Shampoo					
21 days	2.98	1.78	5.00	< 0.03	72

<sup>a</sup>Geometric means are significantly different at  $p \leq 0.05$ . Each point is the average of 12 dogs (replicates) over all of the four treatments.

determined BChE activity with BTCh by subtracting the iso-OMPA readings from the uninhibited readings in the BTCh set. We determined protein by the method of Lowry et al. (12) for standardization.

**Statistics.** We analyzed data using general linear model analysis of variance (ANOVA) for a randomized complete block design with a control and a 4 × 4 (nonshampoo protocol) or a 4 × 5 (shampoo protocol) factorial arrangement of treatment and time after treatment application. The individual dog was the block; multiple observations at each time point were averaged. We examined the residuals from each ANOVA using frequency histograms and normal probability plots; the data were logarithmically transformed and reanalyzed if the normality assumption appeared to be substantially violated. This was true for the residue data but not for the ChE data. If significant effects were found, we separated the means using the Least Significant Difference Test. We performed calculations on a personal computer using the SAS System for Windows, Version 8.0 (SAS Institute Inc., Cary, NC); all statistical tests used the 0.05 level of significance. There was no interaction between treatment and time for the residue studies. There was an interaction between treatment and time for the 4-hr ChE measurements because of the initial inhibition onset. We determined terminal half-life of dissipation of transferable chlorpyrifos residues from linear regression of each sample from 7 to 21 days.

## Results

Table 1 describes the dogs used in this study, including different breeds, weights, and fur length. There was no correlation between transferable residues and fur length at  $p \leq 0.05$ . The type of fur and amount of undercoat were not quantified. In the nonshampoo protocol, dog #8 had the highest transferable residue, 6,999  $\mu\text{g}$  after dip 1, and in the shampoo protocol, dog #13 had the highest transferable residue, 2,676  $\mu\text{g}$  after dip 4 at 4 hr. Comparable breeds of dogs within each protocol did not exhibit the high residues found with dogs #8 or #13. In neither of the protocols did a single dog exhibit consistently the lowest transferable residue as was seen with the high residues.

In both protocols, the levels of chlorpyrifos dissipated quickly after each of the four dippings. Tables 2 and 3 contain the averaged transferable residues and ChE inhibition of all four dippings of the nonshampoo and shampoo protocols, respectively. Figure 1 shows the amount of transferable residue for each dipping in succession for the nonshampoo and shampoo protocols. For the nonshampoo and shampoo protocols, the terminal half-lives of dissipation of transferable chlorpyrifos

residues from 7 to 21 days followed first-order kinetics and were 5.38 days and 5.40 days, respectively.

In both protocols, the plasma ChE was inhibited maximally at 7 days (Tables 2 and 3, average of all four dippings). Figures 2, 3, 4, and 5 show the inhibition of ChE following each of the four dippings for the two protocols. In both protocols, the pretreatment BChE activity was 7- to 10-fold higher than AChE activity (Tables 2 and 3; Figures 2 and 3). The BChE was inhibited to the greatest extent, whereas AChE in both the protocols was minimally inhibited (Tables 2 and 3; Figures 2 and 3). The inhibition of the measured activity of total ChE and BChE (using ATCh) reflects the same pattern of inhibition as BChE using BTCh (Tables 2 and 3; Figures 4 and 5).

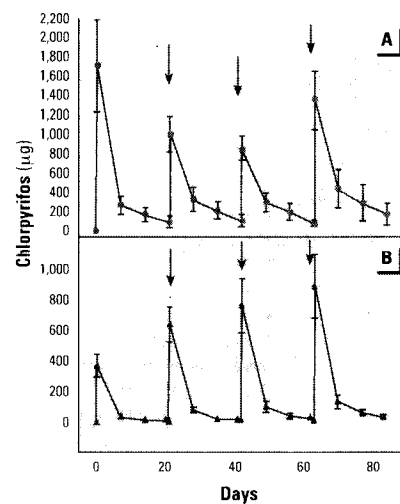
## Discussion and Conclusions

By designing this experiment to use a variety of breeds of dogs and several people for the treatments and sampling, we anticipated that variability in chlorpyrifos residues from dipping (differences in formulation and applicators), dog physiology (differences in breeds and coats), dog activity (rolling in yard, swimming), and petting (pressure, motion, and skin conditions) would reflect the range of values that pet owners might expect to encounter when using a chlorpyrifos dip in the recommended manner for routine pet flea and tick control. The differences among

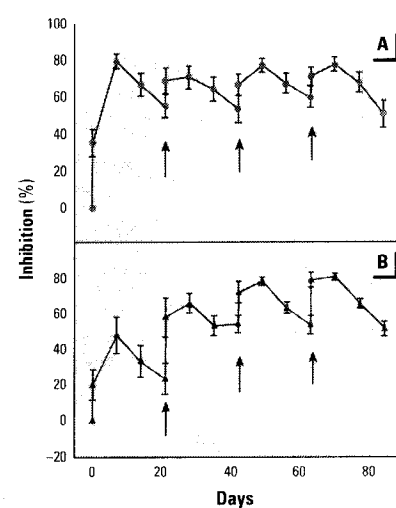
the dogs, as seen from the percent change in Table 1, demonstrate the differences in amounts of transferable residues that might be expected from flea control products caused by the diversity of the animals and handlers. The differences between the shampoo and nonshampoo experiments probably derived from the differences in fur saturation during dipping and in rubbing pressure among samplers. The two protocols were conducted by two different groups of workers and used 12 different dogs in each protocol.

There was a 73–87% average decrease in transferable residue by 7 days after dipping in both protocols. This rapid decrease was not anticipated in the original protocol design, and subsequent studies should include time points between 4 hr and 7 days to fully understand residue dissipation from dog fur. Within the shampoo protocol there was an average decrease of 32% in chlorpyrifos residues after shampooing, but this did not affect the level of cholinesterase inhibition. Although this percent decrease following shampooing appears large, the amount of transferable chlorpyrifos had declined about 97% 20 days from the initial 4-hr levels. The shampooing was not significant in preventing accumulation of chlorpyrifos or decreasing cholinesterase inhibition because the transferable residue had decreased to almost negligible levels by the end of the time period between approved dippings.

The reproducibility test showed that the decrease in transferable residues is a result of



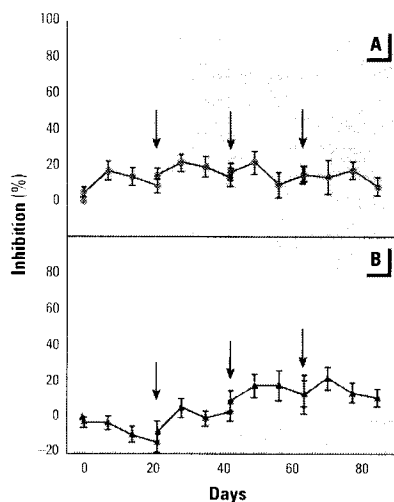
**Figure 1.** Transferable residues of chlorpyrifos from cotton gloves used in petting dogs for 5 min over a 40-square-inch area. The residues are from 12 dogs treated with four sequential dippings of chlorpyrifos every 21 days (A) without shampooing between dippings and (B) with shampooing between dippings. The sample times are 4 hr, 7 days, 14 days, and 21 days after dipping. Arrow represents times of subsequent dippings. The graph represents geometric means with 95% confidence intervals.



**Figure 2.** Percent inhibition of BChE (using BTCh as a substrate and iso-OMPA as an inhibitor) from the plasma of 12 dogs treated with four sequential dippings of chlorpyrifos every 21 days (A) without shampooing between dippings and (B) with shampooing between dippings. The sample times are 4 hr, 7 days, 14 days, and 21 days after dipping. Arrow represents times of subsequent dippings. The graph represents geometric means with 95% confidence intervals.

decline in the levels by degradation or wear and not the result of removal by the sampling procedure. These data showed that the use of cotton gloves for sampling transferable residues leads to reproducible results and that the gloves do not remove all of the pesticide available for exposure.

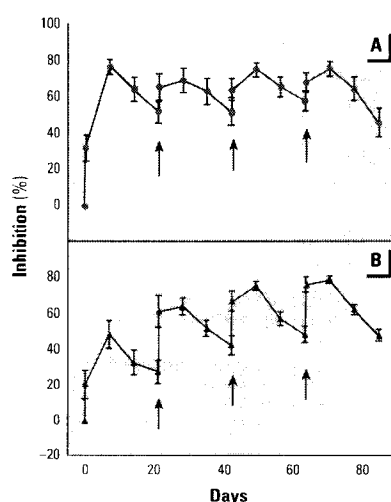
Maximum observed inhibition of plasma ChE activities measured in both studies occurred at 7 days and not at 4 hr probably because of the required activation of the parent compound. Maximum inhibition may have occurred at 2, 3, or 4 days, but blood samples at these time points were not taken because of the experimental design. As with the residue monitoring, subsequent studies should monitor the blood ChE between 4 hr and 7 days. The BChE in the plasma gave the highest enzyme activity and would serve as the most effective biomarker of insecticide action and persistence after an exposure to chlorpyrifos flea and tick control in dogs. Measuring AChE would not be a useful indicator of chlorpyrifos exposure in dogs because AChE levels are low and apparently not as sensitive to inhibition as BChE. Measuring BChE using BTCh gave the highest activity and is the most useful of the several ChE measurements performed for determining decreases in ChE levels after exposure to chlorpyrifos. The study demonstrated also that BChE hydrolyzed ATCh better than AChE hydrolyzed BTCh, and was the reason that inhibition of total ChE using ATCh reflects the inhibition pattern of BChE.



**Figure 3.** Percent inhibition of AChE (using ATCh as a substrate and eserine sulfate as an inhibitor) from the plasma of 12 dogs treated with four sequential dippings of chlorpyrifos every 21 days (A) without shampooing between dippings and (B) with shampooing between dippings. The sample times are 4 hr, 7 days, 14 days, and 21 days after dipping. Arrow represents times of subsequent dippings. The graph represents means with 95% confidence intervals.

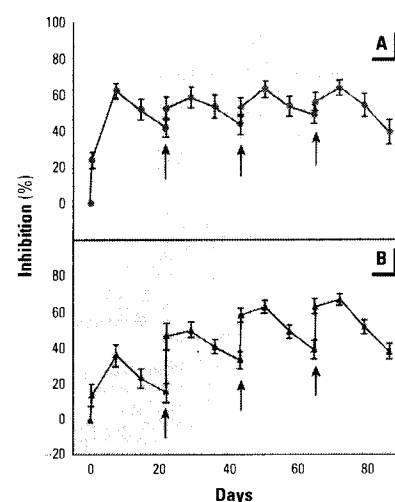
The ChE activity in the blood was depressed substantially after the first treatment, recovered incompletely by the time of the next treatment, and became substantially inhibited again with each subsequent treatment. Inhibition was maintained at 60–80% throughout the experiment. These results were surprising. Although the status of the dog was not the goal of the experiments, there is some concern that dogs with this level of inhibition would be vulnerable to additional exposures to anticholinesterases. Although this information would be used to caution dog owners, chlorpyrifos dips are no longer on the market.

The information gained from this study shows that humans are at greatest potential risk of exposure to chlorpyrifos shortly after dipping. Because children spend time with the pets and are potentially more sensitive to exposures than are adults, plausible exposure levels to calculate risk are needed. As an example, calculating from the 4-hr geometric means, if a child played with a dog for 5 min over an 80-square-inch area (twice the surface area sampled in this study), and the transfer was comparable to that transferred to our experimental gloves, the child could be exposed to an average of 0.9–1.9 mg of chlorpyrifos. With an absorption rate of 3% (13) the child could have a potential dosage of 0.027–0.057 mg. In a 25-kg (55 lb) child the absorbed dosage could be 0.0011–0.0023 mg/kg. In an 80-kg (175 lb) adult the absorbed dose could be 0.0003–0.0007 mg/kg.



**Figure 4.** Percent inhibition of BChE (using ATCh as a substrate and iso-OMPA as an inhibitor) from the plasma of 12 dogs treated with four sequential dippings of chlorpyrifos every 21 days (A) without shampooing between dippings and (B) with shampooing between dippings. The sample times are 4 hr, 7 days, 14 days, and 21 days after dipping. Arrow represents times of subsequent dippings. The graph represents means with 95% confidence intervals.

The 21-day repeated exposure no-observed-effect level (NOEL) for chlorpyrifos is 0.03 mg/kg/day and the reference dose (RfD) is 0.003 mg/kg/day (14,15). The numbers calculated above for the potential exposure from a flea dip are in the range of the NOEL and RfD, but the child would probably not rub the dog with as much pressure as the workers were trained to do, and would probably be cautioned not to play with the dog for the first day after dipping. However, the 5 min data obtained here are believed to be an appropriate surrogate for a much longer handling by a child. Within 1 week the residues declined substantially. At 7 days after the initial dipping using the geometric means, the exposure to the child in this same scenario would decrease to 0.00006–0.0002 mg/kg. Also the numbers calculated from this example are from one time point, and the NOEL and RfD are calculated for 21 days. By extrapolating the geometric mean data throughout the study on a per day basis, the 21-day exposure would range from 0.005 to 0.012 mg/kg, which is 0.0002 to 0.0006 mg/kg/day. With the worst-case-scenario data (the highest level at each time point independent of individual dog) the dosage would be 0.0077 mg/kg/day. The worst-case-scenario level is below the NOEL but slightly above the RfD. If time points such as 1, 3, and 5 days were taken to predict more accurately the decline of transferable residues, then the predicted exposure would be lower. Also, the habits of



**Figure 5.** Percent inhibition of total cholinesterase (using ATCh as a substrate and eserine sulfate as an inhibitor) from the plasma of 12 dogs treated with four sequential dippings of chlorpyrifos every 21 days (A) without shampooing between dippings and (B) with shampooing between dippings. The sample times are 4 hr, 7 days, 14 days, and 21 days after dipping. Arrow represents times of subsequent dippings. The graph represents means with 95% confidence intervals.

children vary with age, the amount of contact with body areas other than hands and forearms, the amount of clothing, and length of time the child plays with the dog. The absorption potentials vary greatly with the site of exposure from 8.6% to 50% for parathion (16). Also, the amount of occlusion, skin damage, and skin binding of the chemical will determine the risk of exposure (17).

We chose the use of cotton gloves as a measurement device for potential exposure to increase the reliability and reproducibility over hand washes. Hand wash protocols vary in time, number of washes, and the wash itself (2-propanol, 10% ethanol/water, 10% isopropanol/water, and 95% ethanol/water) (18–22). Because this was an experimental study and not a field monitoring study, we could not wash one sampler's hands several times over a few hours with a solvent because of skin damage and absorptive problems of the chemical, and we did not wish to expose the samplers to the chemical directly, especially when the treatment was fresh. It has been stated that gloves overestimate hand exposure up to 5 times, but that study did not test for the percent recovery of hand washes (23). A test for percent recovery of hand washes would include exposing a person's hand to a known amount of compound, waiting for a specified amount of time, washing the area, and calculating percent recovery. In many published articles, the percent recovery of compounds was not assessed, and exposure could be underestimated by up to 5-fold with chlorpyrifos (24) and 3-fold with captan (25). The percent recovery and residue removal of chlorpyrifos was not consistent and decreased with decreasing levels of loading (24). Also, skin absorption makes standard hand washing procedures inefficient at removing compounds (17). If hand washes are to be employed, care must be taken to estimate accurately the amount of residue recovered with standardized methods for removal efficiency. If glove dosimeters are slightly higher than hand washes, the more conservative numbers will help risk management better protect our children. For industry, the glove extractions are a conservative, quick, and relatively inexpensive test.

The data generated by this research are useful in determining the amounts of pesticides that children could be exposed to from pets treated for flea and tick control with a dip formulation. It is unknown how representative these data will be for other types of formulations still on the market; our laboratories are currently assessing transferable insecticides from other dips and flea collars using similar methodology. However, more information is needed regarding behavior of children, including their play time with pets and their hand-to-face and hand-to-mouth patterns. Many more types of insecticidal agents with different formulations and application techniques need investigation. The differences between the length of fur and amount of transferable residues were not significant, but we may need further evaluation of fur composition differences among dogs, i.e., undercoat. The data generated from this study demonstrate that BChE measurements are effective biomarkers of insecticide action and persistence following chlorpyrifos flea and tick control in dogs.

There is no perfect measurement technique for determining the amount of pesticide exposure, nor is there a perfect method for determining risk. Given the lack of information in this area and the large number of pesticides in use, the cotton glove dosimeter model is a quick, reliable, and very useful tool in determining potential pesticide exposure.

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