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Golfer Exposure to Chlorpyrifos and Carbaryl Following Application to Turfgrass

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Exposure of golfers to pesticides following their application to turfgrass is of concern to regulators, turfgrass professionals, and consumers. Multipathway exposures were evaluated for golfers on turfgrass treated with chlorpyrifos and carbaryl. Air concentrations and transferable foliar residues (TFRs) were measured to assess potential respiratory and dermal exposures, respectively. At the same time, exposure to individuals simulating the play of golf was determined by dosimetry and urinary biomonitoring. Individual golfer exposure was determined in 76 rounds of golf following eight applications of chlorpyrifos and two applications of carbaryl. Estimated exposures to golfers following full course and full rate applications of chlorpyrifos and carbaryl were 19−68 times below current U.S. EPA acute reference dose (Rfd) values, indicating safe exposures under U.S. EPA hazard quotient criteria. Dermal exposure was determined to be the dominant exposure pathway to golfers, accounting for ~60% of the chlorpyrifos absorbed dose and 100% of the carbaryl absorbed dose. This study also provides a set of transfer factors (TFs) that may be used to determine dermal exposure of golfers to pesticides using transferable residue data.

KEYWORDS: Biomonitoring; carbaryl; chlorpyrifos; dosimetry; golf; turfgrass

INTRODUCTION

Turf environments, including golf courses, parks, athletic fields, and home and commercial lawns, are an integral part of our landscape that provide aesthetic and recreational benefits as well as functional ones, such as reducing runoff and protection against soil erosion. To maintain these benefits, turf environments are managed, and chemical and nutrient application rates can exceed those used in agricultural settings (*1*). Insecticide use on lawns and golf courses is estimated at 240 and 1300 kg active ingredient/km^2, respectively, and herbicide use is estimated at 580 and 500 kg active ingredient/km^2, respectively (*2*). In contrast, insecticide and herbicide use on treated cropland average about 140 and 90 kg active ingredient/km^2, respectively (*3*). According to the 1998 and 1999 Pesticide Industry Sales and Usage Report, up to 85 million pounds of pesticide active ingredient were applied by consumers for residential pest control, while close to 15 million pounds of pesticide active ingredient were professionally applied to golf courses (*4*). There are more than 16 000 golf courses encompassing at least 2.4 million acres (*5*). Additionally, there are approximately 66 million private lawns in the U.S. (*6*).

This widespread and extensive use of pesticides has raised concerns regarding potential human exposure to individuals recreating on or otherwise using turf that has been treated with pesticides (*7, 8*). Because of the large amount of time people spend in turf environments, exposure to pesticides from treated turf is potentially a significant nondietary, nonoccupational exposure pathway. Resulting pesticide exposures may lead to an absorbed dose that is hazardous in and of itself, and/or it may contribute to the aggregate daily dose for those pesticides that individuals are also exposed to through food, water, or other residential exposures.

Chlorpyrifos and carbaryl, two widely used turfgrass insecticides in the northeastern U.S., were selected for this study because their toxicities and modes of action are well researched, their general toxicokinetics are available, and both have a suitable urinary metabolite to monitor.

In humans, chlorpyrifos undergoes oxidative desulfuration to form a more toxic intermediate chlorpyrifos-oxon (*9, 10*), which is rapidly hydrolyzed to 3,5,6-trichloro-2-pyridinol (TCP). The TCP−glucoronide conjugate is the main urinary metabolite, with a urinary half-life ($t_{1/2}$) of 27 h after dermal and oral exposure to chlorpyrifos (*9*). A dermal absorption rate for chlorpyrifos of 9.6% per 24 h has been determined (*11, 12*). The current U.S. Environmental Protection Agency (U.S. EPA) acute oral reference dose (Rfd) is 0.005 mg/(kg d). This Rfd is based on a no-observable adverse effect level (NOAEL) of 0.5 mg/(kg d) from an acute oral rat blood time-course study that reported significant (28−40%) plasma cholinesterace (ChE) inhibition at a peak time of 3−6 h following a single 1 mg/kg dose of chlorpyrifos (*13, 14*).

The principal metabolic pathways for carbaryl are naphthyl ring hydroxylation and carbamic acid ester hydrolysis (*15*). The carbamic acid ester moiety of carbaryl can be hydrolyzed by

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direct enzymatic action of a plasma albumin fraction and in cytosol by β-type esterases, carboxylesterases (*16*), aliphatic esterases, and cholinesterase (*17*). 1-Naphthol is the main metabolite in humans and accounts for more than 85% of urinary metabolites of carbaryl (*18*). Urinary excretion of 1-naphthol occurs rapidly, mostly within 24 h after absorption, as glucuronide or sulfate conjugates (*17*). Most animals excrete between 65 and 75% of the radioactivity in the urine within 24 h (*15*). Biological monitoring by measuring the urinary excretion of 1-naphthol has been conducted (*18*); however, most exposure studies with carbaryl-exposed workers have investigated the suppression of cholinesterase activity. Several risk assessment end points are generally used to evaluate carbaryl exposure hazard, including an acute and chronic dietary Rfd of 0.01 mg/(kg d) and a NOAEL of 1.0 mg/(kg d) (*19*). The NOAEL is used for both acute and chronic dietary end points, as well as for short- and intermediate-term oral and inhalation end points. A dermal penetration rate of 8.3% is used based on the U.S. EPA carbaryl reregistration decision (*19*).

In the current study, golfer exposure is evaluated by measuring residue transfer from insecticide-treated turf using the "California roller" device (*20, 21*), personal air samples, whole body dosimeters, and biomonitoring techniques. Dosimetry and biomonitoring, together with concurrently collected foliar transferable and airborne residue data, provide a novel and complete database on the magnitude of golfer exposure. Relevant transfer factors (TFs) specific to golfers that can be used to predict absorbed dermal dose (ADD) without the need for future human volunteers are also determined.

MATERIALS AND METHODS

Reagents. Analytical standards of chlorpyrifos, TCP, carbaryl, and 1-naphthol were obtained from U.S. EPA National Pesticide Standard Repository (Forte Mead, MD). Isotopically labeled TCP ($^{13}C_2{}^{15}$N-3,5,6-TCP) was a gift from Dow AgroSciences (Indianapolis, IN). Deuterated 1-naphthol (1-naphthol-d_7) was obtained from Cambridge Isotope Laboratories, Inc. (Andover, MA). Triphenyl phosphate (TPP), 2-phenoxybenzoic acid (2-PBA), and the derivitizing agent, *N*-methyl-*N*-[*tert*-butyldimethylsilyl] triflouroacetamide, were purchased from Aldrich (Milwaukee, WI). β-Glucuronidase from *Helix pomatia* (467 000 units/g) was purchased from Sigma Chemical Co. (St. Louis, MO).

Pesticide Applications. All pesticide applications and subsequent exposure trials were conducted between 2001 and 2004. A 100 m × 20 m rectangular bentgrass turfgrass plot was established at the University of Massachusetts Turfgrass Research Center in South Deerfield, MA, for the concurrent collection of transferable foliar, dosimetry, and biomonitoring data. The plot was maintained as a golf course fairway (mowed at a height of 1.3 cm three times a week and irrigated as needed to prevent drought stress). Dursban Pro (23.5% chlorpyrifos, Dow AgroScience, Indianapolis, IN) was applied at the maximum U.S. EPA-approved label rate of 1.12 a.i. kg/ha. Sevin SL (43.0% carbaryl, Lesco, Strongville, OH) was applied at the maximum, U.S. EPA-approved label rate of 7.85 a.i. kg/ha. Immediately following all applications, 1.3 cm of postapplication irrigation was applied to water-in the pesticides according to label recommendations. Pesticide applications were scheduled when weather was predicted to be seasonal, without precipitation, and having wind speeds <10 mph.

Volunteer Golfer Activities. Exposure to volunteers simulating the play of golf was determined by dosimetry and biomonitoring. Individual golfer exposure was determined in 76 rounds of golf following applications of chlorpyrifos (eight applications) and carbaryl (two applications). Each experiment consisted of eight volunteers (one foursome for dosimetry and a second foursome for biomonitoring) simulating the play of an 18-hole round of golf in 4 h. In the standardized round of golf, each player walked 6500 yards, hit a ball 85 times, and took 85 practice swings (170 total). Clubs were rotated in an appropriate way depending on the yardage of individual holes, balls were teed-up, divots replaced, and clubs wiped clean between shots using a golf bag towel as needed. The basic exposure scenario consisted of a single 4 h round of golf starting 1 h after the completion of postapplication irrigation. One application was performed in the evening (8 p.m.) with relevant exposure samples, including golfer dosimetry and biomonitoring, collected the next morning. An exposure scenario simulating the application of chlorpyrifos to only 9 holes was also conducted.

Table 1. Selective Ion Monitoring Mass Spectometry (SIM-MS, 70 eV) for Target Analytes and Internal Standards

analyte	mass ions (*m*/*z*)[a]
TPP (internal standard)	99
chlorpyrifos	197, 258, 314
carbaryl	115, 116, 144
TBDMS Derivatives	
TCP	254, 256
$^{13}C_2{}^{15}$N-3,5,6-TCP (internal standard)	261
1-naphthol	201, 258, 185
1-naphthol-d_7 (internal standard)	208

[a] Quantifier ions are underlined.

Volunteers were recruited by word of mouth from the UMASS Environmental Toxicology and Risk Assessment Program (School of Public Health) and the Department of Veterinary and Animal Science. A protocol approved by the Human Subjects Review Committee, UMASS, including informed consent, was reviewed with potential participants at an orientation meeting prior to their participation (OGCA # 100A0438).

Exposure Determined by Dosimetry. Participants in the dosimetry group wore a whole body dosimeter (WBD) consisting of a long-sleeved shirt and long pants (Universal Overall Corp, Chicago IL) made of a single layer of white, sanforized, 100% cotton, a double layer of cotton gloves (VWR Scientific), and veil (19 cm × 36 cm, 200-thread count cotton fabric) attached with safety pins to the back of a baseball cap. Participants changed to fresh pair of double gloves at the 2 h mark. Suits and veils were removed at the end of the golf round, and the suits sectioned as follows for analysis: lower arms, upper arms, torso, lower legs, upper legs/waist, and gloves.

Insecticide residues were extracted from the WBD sections by rotary-shaking with hexane for 2 h. Following extraction, one-half of the hexane volume was removed and partitioned with a 15% NaCl solution at a 2:1 ratio for 1 min. The hexane layer was collected through Whatman # 1 filter paper containing 25 g of anhydrous sodium sulfate, reduced under vacuum, and the final volume adjusted as needed (10−25 mL) for analysis using an Agilent Technologies 6890 gas chromatograph equipped with a nitrogen−phosphorus detector (GC/NPD), a 7683 automatic sampler (Agilent Technologies, Inc., Wilmington, DE), and a fused silica DB-5 liquid phase, 30 M × 0.25 mm i.d., 0.25 μm film thickness capillary column (J & W Scientific)..

Inhalation exposure was measured using personal air sampling pumps (AirChek 52, SKC, Eighty Four, PA) calibrated to a flow of 2.0 L/min with XAD-2 (140/270 mg) glass fiber air sampling tubes (OVS tubes, SKC) attached to the volunteers' collars. Pesticide residues were desorbed for 1 h with 2.0 mL toluene containing 2 μg/ml triphenyl phosphate (TPP) as an internal standard and analyzed by gas chromatography/mass spectrometry (GC/MS) using a Hewlett-Packard GC (model 5890 Series II) equipped with a MSD (model 5971), and an automatic sampler (model 7673). The capillary column was a fused silica DB-5 liquid phase, 30M × 0.25 mm i.d., 0.25 μm film thickness (J & W Scientific). A deactivated, double-gooseneck, injection port liner (Hewlett-Packard) was used for splitless injection. Helium carrier gas had a linear velocity of approximately 30 cm/s. The injector temperature was 250 °C, and the transfer line was 300 °C. The injection volume was 2.0 μL, and the inlet time purge was 1.0 min. The oven was temperature programmed from 90 °C (held for 2.0 min) to 300 °C (held for 1.0 min) at 10 °C/min. The MSD was operated using the electron impact ionization (70 eV) mode in selected ion-monitoring mode (SIM). The ions monitored for each analyte are listed in **Table 1**.

No pesticide residues were detected in the rear sorbent plug of any field sample, indicating that chlorpyrifos and carbaryl were not breaking

though the front sorbent plug of the personal air sampler tubes. The relative response, determined as the peak area of the quantitation ion from target insecticide divided by the peak area of the TPP internal standard, was used to generate calibration curves (internal standard quantitation). Calibration curves included at least 7 points at varying concentrations, were linear ($R^2 > 0.99$) for each analyte, and ranged from 10 to 250 ng/mL. The absorbed dermal dose (ADD) of each insecticide was calculated by summing residues collected on the WBDs and multiplying by the appropriate dermal penetration rate: 9.6% for chlorpyrifos and 8.3% for carbaryl. To estimate the absorbed inhaled dose (AID), insecticide residues collected by air sampling tubes were adjusted to a 21 L/min inhalation rate and multiplied by a 100% absorption factor. Total absorbed dose (TAD) was then calculated by summing the ADD and the AID.

Exposure Determined by Biomonitoring. Participants in the biomonitoring group wore short sleeve shirts, shorts, ankle socks, and golf shoes. Urinary biomonitoring was conducted for 3,5,6-trichloro-2-pyridinol (TCP), the major urinary metabolite of chlorpyrifos, using 3,5,6-trichloro-2-pyridinol$-C_{13}-N_{15}$ as an internal standard and GC/MS analysis (*22–24*). Total urine volume was collected for 27 h the day before exposure, for 27 h following the chlorpyrifos exposure, and analyzed for TCP. Volunteers were instructed to avoid exposure to any chlorpyrifos during the week prior to the golf-related exposure. Because the half-life of chlorpyrifos is approximately 27 h, the volunteers would have reached pseudosteady state TCP elimination on the day before the golf-related exposure (*23*). Prior to use in exposure estimates, golf exposure-related TCP clearance was adjusted by subtracting the 27 h baseline urinary clearance (concentration × urine volume collected over 27 h) on an individual golfer basis. Whole body dose of chlorpyrifos was calculated according to (*9, 23*)

$$\text{dose } \mu\text{g chlorpyrifos} = (\mu\text{g TCP excreted 27 h})/(0.7151) \times (198/350)/(0.5) \quad (1)$$

In the determination of the whole body dose of chlorpyrifos, the amount of TCP was divided by the urinary excretion factor of 0.4. This factor represents the ratio of molecular weights of TCP (198) and CHP (350.6) (i.e., 198/350.6 = 0.56) times the fraction of the absorbed dose expected to be excreted in urine (0.7151). The fraction expected to be excreted in urine is based on a human study in which an average of 72% of orally administered CHP was excreted in the urine as TCP. An additional correction factor of 0.5 is necessary because TCP was determined at the half-life excretion time.

Humans are not perfect continuous stirred-tank reactors and do not always display perfect first-order excretion kinetics so that the relatively short collection interval (27 h) will likely increase the uncertainty of the data collected. However, the drawback of longer collection intervals would be the increased chance of parent compound/metabolite contamination from other sources. In view of this conundrum, as well as to minimize the burden on the volunteers, we chose a short collection interval as the least problematic approach.

The total carbaryl dose was determined by biomonitoring of 1-naphthol in urine collected for 26 h pre- and postgolf exposure using a modification of a Center for Disease Control and Prevention method (*25*). 1-Naphthol is a common metabolite of both carbaryl and 1-naphthalene, and volunteers were instructed to avoid exposure to any carbaryl or naphthalene during the week prior to the golf-related exposure. Glucuronide and sulfate conjugates of 1-naphthol were enzymatically hydrolyzed using an acetate buffer solution containing β-glucuronidase. Briefly, triplicate 5 mL aliquots of urine were transferred to 20 mL screw top centrifuge tubes and amended with 10 μL of a 1-naphthol-d_7 internal standard solution. To each sample, 2.5 mL of buffer−enzyme solution and 2.5 mL of 0.1 M Na_2SO_4 was added, and the mixture was incubated for 7 h in a 37 °C water bath. An additional 2.5 mL of buffer−enzyme solution was added, and the mixture was incubated an additional 10 h. Following hydrolysis, 0.5 mL of 2 M H_2SO_4 and 5.0 mL of cholorobutane/ethyl ether (8:2) was added. The mixture was shaken on a wrist-action shaker for 1 h and centrifuged at 500*g* for 30 min. The organic layer was transferred to a 15 mL centrifuge tube and extracted with another 5.0 mL of cholorobutane/ethyl ether. The organic layers were combined and vortexed briefly with ~1 g of anhydrous Na_2SO_4. The solvent phase was transferred to a new tube with chlorobutane, reduced to 1 mL under N_2, transferred to a 2 mL autosampler vial, and derivatized for 1 h at 60 °C with 150 μL of *N*-methyl-*N*-[*tert*-butyldimethylsilyl] triflouroacetamide for GC/MS analysis, as described above.

Several pharmacokinetic studies with carbaryl have been conducted in pigs, rats, and humans (*26*). The fraction of administered carbaryl excreted in urine as 1-naphthol in 26 h was reported as 0.22 (*17*). As was done for chlorpyrifos, prior to use in carbaryl exposure estimates, baseline urinary 1-naphthol levels were subtracted from the golf exposure-related 1-naphthol levels. Whole body dose of carbaryl was calculated as follows:

$$\text{dose } \mu\text{g carbaryl} = (\mu\text{g 1-naphthol excreted 26 h})/(0.22) \times (201.2/144.2) \quad (2)$$

Transferable Foliar Residues (TFRs). TFRs were determined using either a "California roller" devise (CA roller) or a water-dampened 9 cm × 23 cm piece of cheesecloth (*27, 28*) in triplicate at 0.25, 1, 2, and 5 h postapplication. Pesticide residues were extracted from the cloth with 350 mL of hexane on a rotary shaker for 1 h. A 200 mL hexane aliquot was removed from the sample jar, collected in an evaporating flask through Whatman # 1 filter paper containing 20 g of anhydrous sodium sulfate, and reduced to 10 mL under vacuum. These extracts were analyzed for the parent insecticides using GC/NPD.

Quality Control. Matrix spikes (fortified with insecticides or metabolites) and matrix blanks were analyzed with every sample set for each matrix. Matrices were fortified and analyzed over a range covering the concentrations detected in the actual exposure samples. Recoveries for chlorpyrifos and carbaryl were 89.9% ± 10.7% and 79.7% ± 7.4% from dosimeters and CA rollers and 94.7% ± 4.7% and 92.6% ± 7.8% from personal air samplers, respectively. Recoveries of TCP and 1-naphthol from fortified urine and distilled deionized water (ddw) samples were 101.7% ± 11.2% and 94.5% ± 18.4%, respectively.

The limit of quantitation (LOQ) was established as the amount of analyte that produced a signal 3 times greater than the background signal of the matrix blank (signal-to-noise ratio of >3 to 1). Additionally, all analytes were amended into blank media at twice the established LOQ level and were reliably detected. Both TCP and 1-naphthol produced a signal-to-noise ratio of >3 to 1 at the 1.0 μg/L concentration level. Isotope dilution quantitation was used for the analysis of TCP and 1-naphthol. Labeled internal standard ($^{13}C_2{}^{15}N$-3,5,6-TCP or 1-naphthol-d_7, respectively) was added to every sample and calibration sample. The relative response of the quantitation ion from the target metabolite divided by the peak area of the quantitation ion from the internal standard was used to generate calibration curves. Calibration curves, which included at least 7 points at varying concentrations, were linear ($R^2 > 0.99$) for each analyte and ranged from 1 to 250 ng/mL. Qualitative identification of metabolites was based on retention time and by comparison of the sample's mass spectrum, after background correction, with the characteristic ions in the spectrums of the analytical standards. The relative intensities of the target ions agreed within 30% of the relative intensities of these ions in the standard spectrum for positive identifications.

Hazard Assessment. Hazard to golfers was assessed independently using the U.S. EPA hazard quotient calculation (*24, 29*) utilizing exposure estimates from both dosimetry and biomonitoring data. The estimated absorbed dose (AD) was divided by the U.S. EPA acute dietary reference dose (Rfd) to give a hazard quotient (AD/Rfd = HQ). The current U.S. EPA acute dietary Rfds are 5 μg/(kg d) for chlorpyrifos (*13*) and 10 μg/(kg d) for carbaryl (*30*). A HQ value less than or equal to 1.0 indicates that the residues present are at concentrations below those that are expected to cause adverse effects to humans. A HQ value greater than 1.0 does not necessary infer that adverse effects will occur but rather that the absence of adverse effects is less certain.

RESULTS AND DISCUSSION

Exposure Determined by Dosimetry. Residues collected onto WBD represent the amount of insecticide that actually transferred to individuals participating in a simulated 18-hole round of golf from treated turfgrass. A "worst case" scenario application of chlorpyrifos, full course applications followed by a 1 h reentry,

Table 2. Chlorpyrifos (CHP) and Carbaryl (CARB) Residues and Estimated Absorbed Dose from Whole Body Dosimeters (WBD) and Personal Air Samplers (XAD-2)

	CHP full course (20)[a]	CHP half course (8)[a]	CHP full course at night (4)[a]	CARB full course (8)[a]
WBD section				
veil[b]	1.4 (1.2)	3.2 (2.8)	1.6 (0.9)	1.8 (2.0)
hands/lower arm[b]	12.6 (1.8)	10.1 (2.2)	23.7 (2.3)	35.4 (7.5)
upper arm[b]	3.9 (0.9)	3.0 (0.7)	9.8 (2.0)	4.8 (1.5)
torso[b]	11.8 (2.2)	8.9 (5.3)	25.3 (6.6)	6.2 (1.5)
upper leg/pants[b]	18.8 (5.5)	13.2 (4.7)	32.9 (5.5)	26.6 (3.8)
lower leg/sock[b]	25.9 (3.8)	21.0 (2.6)	35.3 (3.5)	47.2 (7.0)
total[b]	74.4 (10.5)	59.4 (8.0)	128.6 (21.2)	122.0 (17.0)
ADD[c]	0.102 μg/kg bw	0.081 μg/kg bw	0.176 μg/kg bw	0.145 μg/kg bw
personal air sampler				
XAD-2[d]	5.03 μg (1.5)	3.9 μg (0.7)	6.1 μg (0.8)	N.D.[g]
AID[e]	0.071 μg/kg bw	0.056 μg/kg bw	0.087 μg/kg bw	
TAD[f]	0.173 μg/kg bw	0.137 μg/kg bw	0.263 μg/kg bw	0.145 μg/kg bw

[a] (N) = number of volunteer golfers. [b] Mean micrograms of insecticide ($\pm$SD, standard deviation). [c] Absorbed dermal dose (ADD): assumed a 9.6% absorption rate for CHP and a 8.3% absorption rate for CARB and a 70 kg body weight (bw). [d] Mean micrograms of insecticide residue collected on XAD-2 resin by air samplers ($\pm$SD) at 2 L/min. [e] Absorbed inhalation dose (AID): assumed a 21 L/min breathing rate, 100% absorption rate, and a 70 kg body weight. [f] TAD = Total absorbed dose. [g] N.D. = Not determined.

Table 3. Summary of Urinary Concentration and Total Clearance of TCP and 1-Naphthol in Pre- and Postgolf Exposure

		pregolf exposure		postgolf exposure	
application	N	conc mean (μg/L)	total clearance (μg[a] $\pm$ SD)[b]	conc mean (μg/L)	total clearance (μg $\pm$ SD)
			TCP		
full course, 1 h re-entry	16	5.23	6.66 (2.6)	10.56	9.51 (1.9)
full course, 12 h re-entry[c]	4	5.85	6.43 (2.6)	9.25	10.55 (2.6)
half course, 1 h re-entry	8	4.71	5.27 (2.4)	7.41	6.75 (2.0)
			1-napthol		
full course, 1 h re-entry	8	5.70	5.7 (3.9)	10.4	13.58 (4.8)

[a] Total metabolite clearance (urinary metabolite concentration $\times$ 27 h [TCP] or 26 h [1-naphthol] volume). [b] SD = standard deviation. [c] Application occurred at 8 p.m. All golf rounds started at 8 a.m. the morning following night application.

resulted in an average of 74.4 μg $\pm$ 10.5 μg chlorpyrifos collected onto WBD (**Table 2**). Adjusting for a 70 kg body weight and a 9.6% dermal penetration rate, the estimated ADD is 0.102 μg/kg. This accounts for ~60% of the TAD, with the remaining chlorpyrifos dose coming via inhalation exposure (AID = 0.071 μg/kg), for a TAD of 0.173 μg/kg (**Table 2**). Comparing this TAD to the acute dietary Rfd dose of 5 μg/(kg d) results in a HQ of 0.0346. Half course chlorpyrifos applications (first 9-holes with a 1 h reentry interval) resulted in an ~30% reduction in corresponding TAD (0.137 μg/Kg) and HQ (0.0274) values compared to full course applications.

Although applied at a 7-fold higher rate than chlorpyrifos, carbaryl residues collected on the dosimeters did not increase proportionately. Overall, 122 μg of carbaryl was collected onto WBD following full course applications (**Table 2**). Using an 8.3% dermal absorption rate and the acute dietary Rfd of 10 μg/(kg d), the estimated ADD for carbaryl of 0.145 μg/kg results in a calculated HQ of 0.0145. Carbaryl residues were not detected in the personal air samplers (calculated LOD = 2 μg of total exposure assuming a 21 L/min breathing rate), indicating exposure to this insecticide is almost entirely dermal derived.

Exposure Determined by Biomonitoring. The total metabolite clearance and metabolite concentration in urine collected pre- and postgolfing exposure are summarized in **Table 3**. The volume of urine provided by the individual golfers ranged from 0.55 to 3.7 L. In most instances, urinary volume was less following golfing activities than before, presumably due to mild dehydration. Volunteers were always asked to confirm complete 27 h (TCP) or 26 h (1-naphthol) urinary collection. In all cases, postexposure metabolite concentrations were higher than preexposure concentrations. For all volunteer golfers, the preapplication exposure TCP levels (average of 5.1 μg/L TCP, N = 28) were consistent with reference concentration levels for the general population as reported by Hill et al. (*31*), and Barr et al. (*32*) and background levels reported by Byrne et al. (*23*). The pre-exposure urinary 1-naphthol levels for all volunteer golfers (average 5.7 μg/L 1-naphthol, N = 8) were consistent with reference concentration levels for the general population as reported by Hill et al. (*31*),.

Estimates of whole body dose as judged by urinary biomonitoring are summarized in **Table 4**. Metabolites detected in the preexposure urine were subtracted from postexposure metabolite clearance on an individual basis, so that the postgolf exposure estimates represent only the absorbed dose that occurred during the simulated play of golf. Dividing the mean chlorpyrifos AD (adjusted for prestudy clearance) following the full course application scenarios (0.159 μg/(kg d) $\pm$ 0.032, N = 16) by the acute oral Rfd (5 μg/(kg d)) yields a HQ value of 0.0318. Although TCP is a common metabolite for both chlorpyrifos and chlorpyrifos-methyl, the difference in TCP levels before and after golfing are likely due to chlorpyrifos exposure alone that occurred during golf, because the exposures were highly choreographed and the resulting exposures were uniformly well above background exposure levels. Nevertheless, some of the measured TCP may have been derived from environmental exposure to TCP itself, which would overestimate the resulting chlorpyrifos dose (*33*). TCP was not analyzed in the environmental samples; however, direct exposure to TCP in these scenarios is likely minimal because the entire exposure occurred within 5 h following chlorpyrifos application, and very little formation of TCP would be expected to occur on foliage surfaces in that time frame (*34*). The amount of carbaryl absorbed during a round of golf under our worst case scenario (adjusted for prestudy clearance) was 0.544 μg/(kg d) $\pm$ 0.183. Dividing this dose by the carbaryl Rfd (10 μg/(kg d)) yields a HQ of 0.054.

Table 4. Summary of Absorbed Chlorpyrifos and Carbaryl Dose as Measured by Urinary Biomonitoring Following Golf Exposure

application	N	mean volunteer body weight (kg)	Absorbed Dose (μg/Kg)[a] mean (SD)[b]	range
		chlorpyrifos		
full course, 1 h re-entry	16	89.4	0.159 (0.032)	0.121−0.220
full course, 12 h re-entry[c]	4	87.2	0.238 (0.098)	0.128−0.349
half course, 1 h re-entry	8	81.8	0.090 (0.027)	0.062−0.126
		carbaryl		
full course, 1 h re-entry	8	91.3	0.544 (0.183)	0.311−0.807

[a] Chlorpyrifos and carbaryl equivalents calculated from TCP and 1-naphthol concentrations detected in pre-exposure urine (μg/(kg d)) were subtracted from the postgolf exposure equivalents (μg/(kg d)), so resulting absorbed dose estimates represent the absorbed dose resulting from exposure during the simulated play of golf. [b] SD = standard deviation. [c] Application occurred at 8 p.m. All golf rounds started at 8 a.m. the morning following night application.

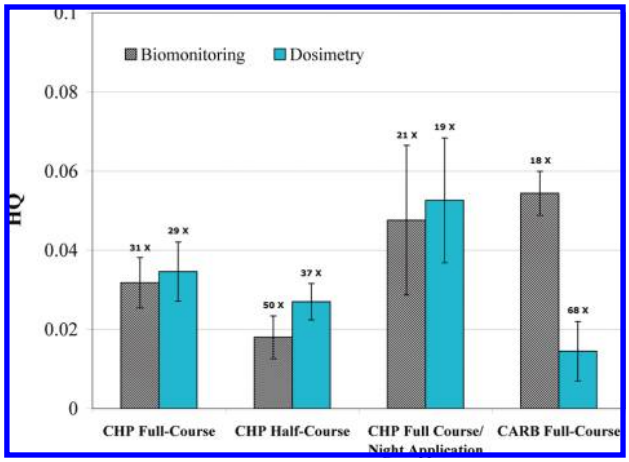

Figure 1. Hazard quotients (HQs) determined for a single 4 h round of golf following applications of chlorpyrifos (CHP) or carbaryl (CARB). Values above the standard deviation bars indicate the magnitude below a HQ = 1.

A rotating group of volunteer golfers were used for all exposure scenarios, and individual volunteers were randomly placed in either the dosimetry or biomonitoring group the week before the actual exposure event. The dosimetry group participants were mostly covered from head to toe so that it was not possible for them to receive an absorbed pesticide dose representing that of an actual golfer. Therefore, in our experimental design, dosimetry was used primarily to determine routes of exposure, and biomonitoring was used to estimate the total dose received by golfers. This precluded pairing the biomonitoring and dosimetry groups. Without exposure route information, it is difficult to relate biomonitoring results to sources and routes of exposure and to develop effective mitigation practices.

The above exposure estimates, which are based on a 1 h reentry interval following full course and full rate applications of chlorpyrifos and carbaryl, are substantially below (19−68 times) current U.S. EPA acute Rfd values (HQ < 1, **Figure 1**), indicating safe exposures under these criteria. Acute Rfds were selected over chronic Rfds as a toxicological end point in the HQ calculations because: (1) it is unlikely that golfers will encounter these worst case exposures on every round of golf over a period of many years, and exposure associated with turfgrass applications decreases rapidly over the first several days (*24, 27–29*); (2) the insecticides tested are rapidly metabolized and excreted, and cumulative effects from subsequent golfing exposures are not likely; and (3) the U.S. EPA uses a similar approach for postapplication/residential risk assessments (*14*).

Dosimetry and biomonitoring measurements of AD starting 1 h after postapplication irrigation following full course applications of chlorpyrifos were 0.173 μg/kg ± 0.027 (HQ = 0.034) and 0.159 μg/kg ± 0.032 (HQ = 0.032), respectively. These independent estimates of AD are not statistically different (unpaired *t*-test; $p > 0.05$). Field trials were replicated for each exposure scenario over the course of several summers, and exposure values were averaged together. Through careful planning, each simulated round of golf began at 8 a.m., and weather conditions were standardized by having high temperatures of at least 75 °F (24 °C), no precipitation, and wind speeds of less than 10 miles per hour. By this process, climactic variations that could influence insecticide transferability (e.g., solar radiation, temperature, and humidity) were minimized.

Geer at al. (*33*) reviewed data from five studies evaluating chlorpyrifos exposure to pesticide handlers submitted to the EPA by registrants. Exposure estimates from dosimetry, assuming dermal absorption factors of 1, 3, and 10%, were compared to the exposure estimates based on biomonitoring. The resulting median ratios of 0.45, 0.71, and 1.28, respectively, suggest the chlorpyrifos absorption range is between 3 and 10%. We used two conservative assumptions to derive an ADD from the WBD in the current study: (1) Although clothing is known to substantially reduce dermal exposure, dermal exposure estimates were calculated by combining the residue collected on the entire WBD. The body regions left exposed in the biomonitoring group (and typically left exposed by golfers), namely the head, lower legs, hands, and lower arms, accounted for 48−85% of the total dermal residues transferred to the golfers (**Table 2**). (2) The summed WBD residues were then multiplied by the dermal absorption rate of 9.6% (*12*), the high end of the penetration values estimated for chlorpyrifos. Both assumptions should tend to overestimate the ADD.

Dermal absorption, however, is known to be increased due to occlusion (*36*), use of sunscreens (*36*), skin moisture (*37*), and degree of skin hydration (*38*). Additionally, the extent of absorption though skin is influenced by body location (*39*), as well as the loading concentration (mass/area) of the material, presence of carrier or formulation, and temperature, making estimates of dermal absorption during real-life scenarios problematic (*40*). There are also concerns regarding the adequacy of the cotton dosimeter to serve as a skin surrogate (*41*). Despite these limitations, dosimetry is still considered the best external means for assessing dermal exposure (*40*). Thus, the good agreement between the actual absorbed dose determined by biomonitoring and that estimated from dosimetry techniques provides validation of the dosimetry techniques and the assumptions used to calculate dose from the dosimetry media.

Assessments performed following full course and full rate carbaryl applications resulted in exposure estimates of 0.145 μg/(kg d) ± 0.02 (HQ = 0.015) from dosimetry (assuming a 8.3% dermal penetration rate, **Table 2**) and 0.544 μg/(kg d) ± 0.183 (HQ = 0.054) from biomonitoring (**Table 4**). These assessments are statistically different, (unpaired *t*-test; $p > 0.05$), highlighting the limitations of the assumptions in both the

Table 5. Mean 4 h Transferable Foliar Residues (TFRs) and Calculated Transfer Factors (TFs) following Postapplication Irrigation

	mean 4 h TFR (μg/cm^2)[a]	dermal exposure (μg)[b]	transfer factor (cm^2/h)
	chlorpyrifos		
CA roller	0.003	74.4	6100
	carbaryl		
CA roller	0.019	122.0	2000
cloth wipe	0.112	122.0	340

[a] 1−5 h postirrigation. [b] Values taken from **Table 2**.

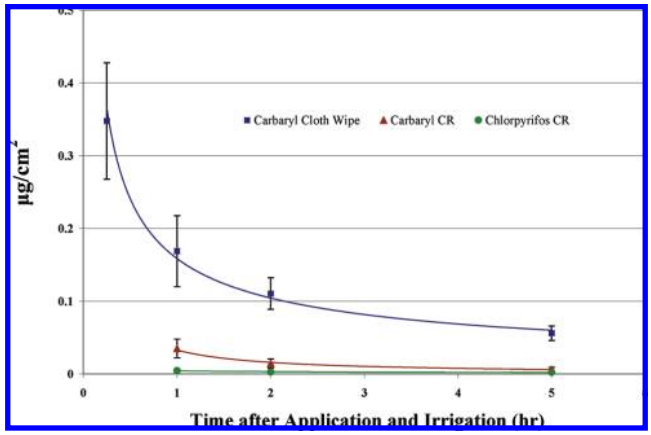

Figure 2. Transferable foliar residues (TFRs) of chlorpyrifos and carbaryl over the first 5 h following postapplication irrigation as measured by the CA roller (CR) and cloth wipe techniques.

dosimetry and biomonitoring methods. However, both result in HQs significantly less than 1, again indicating a wide margin of safety for golf-related carbaryl exposure.

Transferable Foliar Residues (TFRs) and Dermal Transfer Factors (TFs). A limited amount of insecticide residue is available for transfer by 1 h postapplication, as judged by TFRs levels. The mean chlorpyrifos TFRs collected over the 4 h golfing period was 0.003 μg/cm^2 (**Table 5**). This amount equates to a transfer rate of 0.027% (chlorpyrifos was applied at 11.2 μg/cm^2). Similarly, carbaryl was applied at 78.5 μg/cm^2, and the mean TFRs collected over the 4 h golfing period were 0.019 μg/cm^2, which represents a transfer rate of 0.024%. Carbaryl TFRs were also collected using the dampened cheesecloth starting 0.25 h postapplication. As previously reported for chlorpyrifos (*24*), a rapid decrease in carbaryl TFRs occurred between 0.25 and 1 h as the irrigation water dried and the insecticide began to associate with the waxy layer of the turfgrass itself (*26, 27*). Carbaryl TFRs declined from 0.348 μg/cm^2 at 0.25 h to 0.135 μg/cm^2 by 1 h, a reduction of approximately 50% (**Figure 2**). This finding suggests that initial dermal exposure can be reduced by as much as 50% if golfers observe a 1 h reentry interval. Subsequent TFRs decline after the first 1 h drying time was much slower (**Figure 2**), and a longer reentry interval would not yield proportional reductions in exposure.

Reducing the exposure period to 2 h by functionally treating only the first 9 holes (half course applications) reduced total chlorpyrifos AD as measured by biomonitoring (0.090 μg/kg $\pm$ 0.027) by ~30% compared to full course applications. Total AD was not reduced by the expected 50% following these half course applications, indicating most exposure occurs during the first 2 h postapplication. This finding is supported by a gradual decline of TFRs over the 4 h exposure period (**Figure 2**), indicating insecticide transfer would be higher during the first two hours of play (i.e., the first 9 holes). As a best management practice (BMP), half- or partial-course applications spaced over a day or two to allow TFRs to dissipate could significantly reduce potential exposure to golfers. The BMP of extending the reentry interval by applying at night did not reduce (unpaired *t*-test; $p = 0.33$) AD (0.238 μg/kg $\pm$ 0.098) compared to 1 h reentry interval exposures (0.159 μg/kg $\pm$ 0.032) and was not an effective mitigation strategy. Evening applications with its cool stagnant nights, no solar radiation, and the formation of morning dew prevented chlorpyrifos from dissipating as expected. This highlights the continued need to evaluate BMPs to reduce postapplication pesticide exposure.

Dermal transfer factors (**Table 5**) relevant to golfer activity were calculated using the method of Zweig et al. (*42*):

$$\text{TF (cm}^2\text{/h)} = \text{dermal exposure } (\mu\text{g})/\text{TFR } (\mu\text{g/cm}^2)/4 \text{ h} \quad (3)$$

where TF = transfer factor, dermal exposure (μg) was derived from dosimeters, and TFR was the mean 4 h TFR value over the period concurrent to simulated golfing activities (1−5 h postapplication). The good agreement among the biomonitoring and dosimetry techniques, combined with the measurement of environmental residues (TFRs), provides a thorough picture of transferable pesticide residues and golfer exposure and forms the basis for predicting absorbed dermal dose (ADD) by golfers using the standardized CA roller technique:

$$\text{ADD} = \text{TFR } (\mu\text{g/cm}^2) \times \text{TF (cm}^2\text{/h)} \times \text{DA} \times 4 \text{ h}/70 \text{ kg} \quad (4)$$

where DA is the appropriate dermal absorption fraction of the pesticides and 4 h represents a single, 4 h, 18-hole round of golf.

The CA roller technique was developed to give a more realistic and standardized estimate of the amount of TFRs that are available for transfer to recreational users of chemically treated turf (*40*). The CA roller-derived TF for carbaryl is notably lower than the comparable TF for chlorpyrifos. This result is likely due to the increased water solubility of carbaryl compared to chlorpyrifos. Because all applications were followed by postapplication irrigation, it is likely that carbaryl, due to its higher water solubility, initially remains associated with irrigation water and is easily transferred to the cloth by absorption under the weight of the CA roller, whereas chlorpyrifos will likely associate with the waxy layer of the turfgrass more readily. Although the carbaryl-contaminated water suspended in the turfgrass and thatch layer is efficiently transferred to the weighted roller, the same transfer phenomenon is not occurring to the cotton dosimeters, which are not generally in direct contact with the turfgrass.

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