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The Effect of Cooking on Chlorpyrifos and 3,5,6-Trichloro-2-Pyridinol Levels in Chlorpyrifos-Fortified Produce for Use in Refining Dietary Exposure

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Various types of produce were fortified with chlorpyrifos and then boiled, baked, canned, or concentrated as appropriate for the type of produce. Both uncooked and cooked samples were analyzed for chlorpyrifos and 3,5,6-trichloro-2-pyridinol, and then, chlorpyrifos cooking factors were calculated by comparing the postcooked concentration to the uncooked concentration. The cooking factors were dependent upon the commodity and cooking procedure: 0.320−1.19 for boiled samples, 0.022−1.18 for baked pulp, and 0.119−0.661 for canned samples. Concentrating chlorpyrifos-fortified orange juice 4-fold resulted in a concentration factor of only 2.6, indicating a loss of chlorpyrifos. Green bean and green pepper plants treated in the greenhouse yielded higher chlorpyrifos concentrations but similar cooking factors to lab-fortified samples. The cooking factors can be used with food consumption databases and modeling tools to refine the dietary exposure according to current product label uses.

KEYWORDS: Residues; cooking; chlorpyrifos; TCP; factor; exposure assessment

INTRODUCTION

To estimate potential pesticide exposures from food, it is important to estimate the level of exposure at the point of consumption in the home. Most produce is not consumed fresh, unwashed, or unprocessed, and in addition to commercial processing, produce is also consumed after commercial or home cooking. The effect that cooking has on the level of possible pesticide residues in food is required to refine the dietary exposure to a more realistic level. This study was conducted to provide cooking factors to allow for refinement of dietary exposure at the point of consumption in the home.

Chlorpyrifos [*O*,*O*-diethyl-*O*-(3,5,6-trichloro-2-pyridinyl)-phosphorothioate] has been widely used to control pests on plants and animals. Residues of chlorpyrifos in produce, as measured by residue trials conducted at the maximum label rate and minimum sampling interval after application, generally overstate the residue that will typically be found in produce in a grocery store or as consumed. Monitoring residue data from the U.S. Department of Agriculture (*1*, *2*) and the Dow AgroSciences market basket survey (*3*) indicate that most produce will have very low, or nondetectable, chlorpyrifos residues [generally below 0.01 mg/kg (10 ppb)].

To ensure the availability of produce with measurable levels of chlorpyrifos for the cooking experiments described in this study, produce was fortified or treated with chlorpyrifos at a level that allowed the measurement of chlorpyrifos before and after cooking. Fortification was required because the market basket levels of chlorpyrifos in produce are so low. Fortification of commercially obtained produce should closely mimic field-incurred residues because metabolism studies have shown that chlorpyrifos itself was not translocated to any appreciable extent in plants (*4*). Consequently, the presence of chlorpyrifos in fruits and vegetables was largely a result of a direct application to the developing fruit or vegetable during the pest control circumstance. In plants, chlorpyrifos is metabolized to 3,5,6-trichloro-2-pyridinol (TCP), which is then conjugated (*5*); therefore, samples were analyzed for both chlorpyrifos and TCP.

The effect of processing chlorpyrifos-treated apples has been studied (*6*). In that study, residues in whole apples were reduced by greater than 96% when processed into applesauce [seven of 11 samples below the limit of detection (LOD)]. Therefore, in the current study, fruits and vegetables were fortified at a level high enough to enable measurement of the chlorpyrifos levels after cooking, thereby enabling the calculation of a cooking factor. The projected level of fortification for this study was 1 mg/kg or 100 times the proposed limit of quantitation (LOQ). Thus, if postcooked levels of chlorpyrifos were below the LOQ, then there would have been at least a 100-fold dilution or loss from cooking that could be used in the refined exposure assessment. This study describes how to measure and calculate cooking factors and how the resulting data may be used in refining the exposure and therefore refining the risk assessment but does not go into detail on the refined risk assessment for chlorpyrifos in foods.

After the fruits and vegetables were fortified with chlorpyrifos, the produce was boiled, baked, canned, or concentrated.

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Table 1. Specifics of the Cooking Procedures Used for Each Fortified Commodity[a]

commodity	preparation prior to fortification (same as uncooked sample)	boiling: water added (g)	boiling: time (min)	baking: temp (° C)	baking: time (min)	canning[b]: pressure (psi)	canning[b]: time (min)
apples	peeled, cored, sliced, sample ≈ 500 g	65[c]	17	177	32	6	9
broccoli	washed, cut into spears, sample ≈ 500 g	1500	8	NA	NA	NA	NA
cabbage	washed, outer leaves removed, cut into wedges, sample ≈ 500 g	1500	15	NA	NA	NA	NA
cherries	washed, pitted, sample ≈ 500 g	65	15–16	177	30	6	10
green beans	washed, stems and blemishes removed, sample ≈ 350 g	1500	18[d]	NA	NA	11[d]	25
peaches	peeled, halved, pits removed, sample ≈ 500 g	1500	15–17	NA	NA	6	10
peppers	washed, halved, stems and seeds/membranes removed, sample = 8 halves (outside treated)	1500	15	177	30	NA	NA
sweet potato	peeled and cubed for boiling and canning; washed and left whole for baking, sample ≈ 500 g (2 whole potatoes)	1500	20–22	204	55[e]	10[f]	90
winter squash	seeds removed, boil: peeled and quartered, bake, quartered (peel left on); can, peeled and cubed, sample ≈ 500 g	1500	19–21	177	55[e]	10	90

[a] NA = not applicable. [b] Cold-packed commodities (apples, cherries, beans, and peaches) were added to a jar, and then, enough boiling water was added to fill the jar. The hot packing procedure used for sweet potato and squash is described in the text. [c] All or most of the water evaporated during boiling. [d] Fortified green bean data are presented in the table. Greenhouse-treated green beans were boiled for 15 min and canned at 10 psi (control canned treated sample did not form a vacuum). [e] The pulp and peel were analyzed separately. [f] Some of the water from hot packing would not fit in the jars (control, 102 mL; fort-1, ~85 mL; and fort-2, ~5 mL).

The level of chlorpyrifos in the commodity, and in added or accumulated water from the cooking method, was measured before and after cooking to determine a cooking method factor (postcooking concentration/uncooked concentration). In addition to monitoring the fate of chlorpyrifos during cooking, the amount of TCP was also quantified because TCP is the primary degradation product of chlorpyrifos in crop tissues (*5*, *7*) and a major degradate of aqueous hydrolysis at neutral and acidic pH (*5*). Monitoring for the presence or absence of TCP after cooking provided insight into the mechanism of loss of chlorpyrifos (e.g., degradation or volatilization). At a near neutral pH of 6.5−7.5, chlorpyrifos is stable to aqueous hydrolysis, with degradation half-lives ranging from three to 99 days (*8*). The degradation rate of chlorpyrifos reportedly increases with increasing temperature (*8*).

MATERIALS AND METHODS

Produce. Apples (variety red Delicious), broccoli (variety unknown), cabbage (variety Green), cherries (variety Bing), green beans (variety unknown), peaches (variety unknown), peppers (used peppers grown in the greenhouse, see below), sweet potatoes (variety uncertain, possibly Beauregard), squash (variety Acorn), and orange juice were used. The commodities were obtained from grocery stores or wholesale fruit vendors near Indianapolis, Indiana. After purchase, the commodities were stored refrigerated (approximately 6 °C) until used.

Fortification and Treatment. The fortification procedure was specific to the type of produce, but the general procedure is described here. First, the produce was prepared (washed, peeled, seeds removed, etc. as described in **Table 1**). The prepared produce was spread evenly onto a tray, and the weight of the produce was recorded. The fortification solution was prepared by diluting Lorsban 4E (45.4% active substance, 0.105 mL) to 100 mL with tap water to achieve an approximate 500 μg/mL solution {[(0.105 mL × 0.454 g/mL)/100 mL] × 1 000 000 μg/g = 477 μg/mL}. The produce was fortified with an appropriate volume of fortification solution (2 mL per kg sample) to achieve a residue level of 1 mg/kg in the produce. The diluted fortification solution was applied as a fine mist onto the surface of the produce (as it lay on a tray) using a CAMAG TLC sprayer; the tray was rotated during application, but the produce was not stirred or turned over. After the spray solution was allowed to dry on the produce, the produce was covered with plastic film and refrigerated. Orange juice was fortified similarly, except that the required amount of formulated, diluted fortification solution was pipetted directly into aliquots of the juice.

Green beans (types Kentucky Wonder and Purple Teepee) and green bell peppers (type Big Bertha PS Hybrid) were planted in the greenhouse in individual pots. The plants were watered and fertilized to maintain strong vegetative growth, and nonorganophosphate insecticides were applied as needed. The plants were subsequently treated with chlorpyrifos using a procedure that would provide residues that should closely approximate the type of residues produced as a result of the labeled uses of chlorpyrifos. The target treatment rate was 2.5 kg/ha, approximately equivalent to the maximum allowable chlorpyrifos application to vegetables. The formulated treatment solution was prepared by diluting Lorsban 4E (0.50 mL) to 100 mL with tap water for a diluted concentration of 2270 μg/mL. Each plant was sprayed with an aliquot of the formulated, diluted treatment solution (5.5 mL) that was applied as a fine mist, aiming the application solution toward the edible produce, although the foliage also received treatment. Afterward, the spray solution was allowed to dry on the plants.

Water (1500 ± 0.3 g) was measured and heated to boiling. A 5 mL aqueous aliquot of either chlorpyrifos or TCP (approximately 0.3 mg/mL) was added into the heated water and boiled for 15 min. After it was cooled, the water was weighed and then aliquoted for analysis.

Sample Collection and Cooking. Trays containing the fortified produce samples were removed from the refrigerator 22−45 h after spraying. To obtain unbiased samples, opposite one-eighth portions of the produce on a given tray were combined into a single sample for either cooking (duplicates) or analyzing directly (duplicates) as detailed in **Table 2**. All of the edible greenhouse-treated produce was harvested, irrespective of produce size, at 5−7 DAT (days after treatment) vs a typical preharvest interval of 21 days for most vegetables, thereby maximizing chlorpyrifos residue levels. For analysis, the appropriate amount of greenhouse-treated produce was selected at random and composited, producing representative samples. Duplicate greenhouse-treated samples were cooked by each procedure specific to the commodity (beans were boiled and canned, and peppers were boiled and baked). After they were cooled, the commodity samples were homogenized and analyzed as described below. Duplicate uncooked greenhouse-treated samples were also homogenized and analyzed. Control samples (cooked and uncooked) of each commodity were also analyzed.

The samples for cooking were boiled, baked, canned, or concentrated as described in **Table 1**, **Table 2**, and below. The procedures that were used simulated commercial practices or common household practices.

Table 2. Number and Types of Samples that Were Analyzed for Each Commodity and Cooking Procedure

commodity	uncooked—control	uncooked (fortified unless noted)	boiled	boiling liquid	baked	canned	concentration	total
apples	3	6	2 fort 1 cntl	0	2 fort 1 cntl	2 fort 1 cntl	0	18
broccoli	1	2	2 fort 1 cntl	2 fort 1 cntl	0	0	0	9
cabbage	2	2	2 fort 1 cntl	2 fort 1 cntl	0	0	0	10
cherries	3	6	2 fort 1 cntl	0	2 fort 1 cntl	2 fort 1 cntl	0	18
green beans[b]	4	4 fort[c] 4 treated[d]	2 fort 2 cntl 2 trtd	2 fort 2 cntl 2 trtd	0	2 fort 2 cntl 2 trtd	0	30
peaches	2	4	2 fort 1 cntl[a]	2 fort 1 cntl	0	2 fort 1 cntl	0	15
peppers[b]	2	4 fort[c] 4 treated[d]	2 fort 2 cntl 2 trtd	2 fort 2 cntl 2 trtd[d]	2 fort 2 cntl 2 trtd	0	0	28
sweet potatoes	3	6	2 fort 1 cntl	2 fort 1 cntl	2 fort[e] 1 cntl[e]	2 fort 1 cntl	0	21
winter squash	3	6	2 fort 1 cntl	2 fort 1 cntl	2 fort[e] 1 cntl[e]	2 fort 1 cntl	0	21
orange juice	1	2	0	0	0	0	2 fort 1 cntl	6
water—chlorpyrifos[f]	0	2	0	2 fort	0	0	0	4
water–TCP[f]	0	2	0	2 fort	0	0	0	4
total	24	54	33	31	18	21	3	184

[a] "Cntl" indicates a control sample. [b] Fortified and incurred residues (greenhouse treated) were evaluated separately. [c] "Fort" indicates a fortified sample. [d] "Treated" and "trtd." indicate a greenhouse-grown plant treated, and then, samples harvested. [e] Peels were analyzed separately. [f] Water was fortified with chlorpyrifos or TCP.

In general, samples that were cooked (not canned) were stored frozen at approximately −20 °C. Canned samples were stored at room temperature until they were prepared for analysis.

The boiling procedure used for each commodity is described in more detail in **Table 1**. Tap water was brought to boiling over high heat in a saucepan; less water was used for apples and cherries, preparing a sauce (not boiled until after cherries added). The sample of produce was added and boiled 8−20 min depending upon the type of produce. The mixture was allowed to cool, and the liquid was separated from the solids (not separated for apples and cherries, since the samples were sauces, therefore, no "boiling liquid" was analyzed). Each phase was weighed, large pieces were chopped, and the samples were frozen pending homogenization. The liquid was aliquoted for analysis. The chlorpyrifos- and TCP-fortified water samples were boiled for 15 min.

The baking procedure used for each commodity is described in more detail in **Table 1**. The sample was placed in a tared glass crystallization dish, and the weight of the produce was recorded. The sample was baked at 177 or 204 °C (350 or 400 °F) until cooked throughout (easily pierced with a fork). After the samples were baked, the peel was separated from the pulp of sweet potatoes and winter squash. The peel and remaining pulp were weighed. Large pieces were cut into smaller ones, and the samples were frozen pending homogenization.

The canning procedure used for each commodity is described in **Table 1**. Sweet potatoes and winter squash were "hot-packed" in which the sample was added to boiling tap water (500 g for sweet potatoes, 100 g for squash) and the mixture was heated just to boiling. The hot mixture was immediately poured into tared canning jars, the weight was obtained, and the jar was sealed. Apples, cherries, green beans, and peaches were "cold-packed" in which the sample was placed in the tared canning jar, the weight was obtained, the jar was filled with boiling tap water, the weight of the mixture was obtained, and finally, the jar was sealed.

Whether hot- or cold-packed, the sealed canning jars were placed in pressure cookers containing approximately 5 cm of hot or boiling water. The pressure cookers were closed, and heat was applied until the pressure was high enough to maintain the air vent/cover lock in the up (closed) position. The vent pipe was then covered with the pressure regulator while heat was added to increase the pressure. The desired pressure was achieved and maintained by adjusting the heat. After they were heated for the appropriate time at the appropriate pressure, the pressure cookers were removed from the heat and allowed to cool. Once the pressure returned to normal and the air vent/cover lock dropped, the jars were removed and allowed to cool. The samples remained sealed in the canning jars at room temperature until homogenized.

Duplicate orange juice samples (400 mL) were transferred to 1000 mL round-bottomed flasks, weighed, and then concentrated on a rotary vacuum evaporator (80 °C water bath, dry ice trap) to approximately one-quarter of the original volume. The concentration took approximately 45−60 min per sample. The concentrated liquid was aliquoted for analysis.

Samples were cryogenically milled in the presence of liquid nitrogen and/or dry ice to a fine powder using an Agvise hammer mill.

Analytical Methods and Calculations. Briefly, the produce analysis method consisted of a single extraction of a 1.5 g aliquot with acetone/water (80/20, v/v, 30 mL, 30 min extraction), followed by acidification of an aliquot of the extract (1.0 mL) and concentration with a C_{18} solid phase extraction (SPE) column eluted with acetonitrile/0.1 N HCl (90/10, v/v) (omitted SPE for the water samples). The SPE eluent or water extract was further acidified and saturated with sodium chloride, and the residues were partitioned into 1-chlorobutane. After the residues were dried and an internal standard (^{13}C ^{15}N-chlorpyrifos stable isotope) was added, chlorpyrifos was analyzed without derivatization while TCP was derivatized with N-methyl-N-(*tert*-butyl dimethylsilyl)trifluoroacetamide to form the silyl-dimethyl-tertbutyl derivative ($C_{11}H_{15}Cl_3NOSi$). Determination was by capillary gas chromatography (DB-5MS capillary column) with mass selective detection, with the mass selective detector using negative chemical ionization and operating in the selective ion mode. The ions that were monitored during the analysis of the crop and water samples were as follows: *m*/*z* 313 for chlorpyrifos, *m*/*z* 161 for TCP, *m*/*z* 318 for ^{13}C ^{15}N-chlorpyrifos (internal standard), *m*/*z* 217 for the chlorpyrifos-methyl internal standard for the peach samples, and *m*/*z* 161 for 2,3,6-TCP (internal standard).

The chlorpyrifos analytical method performance during the study was determined by analysis of freshly fortified control samples (n = 48) over the range of 15−7500 ng/sample. The mean recovery was 87% (s = 8.2%). The TCP analytical method performance during the study was determined by analysis of freshly fortified control samples (n = 43) over the range of 15−1500 ng/sample. The mean recovery was 89% (s = 11.5%). Both the chlorpyrifos and the TCP LODs and

Table 3. Average Chlorpyrifos and TCP Concentrations and Calculated Chlorpyrifos Cooking Factor for Each Commodity and Cooking Procedure[a]

	total chlorpyrifos			total TCP	
commodity	average precooking (ng/g) (RSD)	average postcooking (ng/g) (RSD)	cooking factor	average precooking (ng/g) (RSD)	average postcooking (ng/g) (RSD)
apples, boiled	1258 (11%)	700 (23%)	0.556	14 (16%)	15 (14%)
apples, baked	1359 (2%)	828 (9%)	0.609	15 (3%)	24 (19%)
apples, canned[b]	878 (14%)	580 (53%)	0.661	15	31 (36%)
broccoli, boiled	550 (32%)	514 (14%)	0.935	12	<LOQ
cabbage, boiled fortified	577 (9%)	480 (38%)	0.832	<LOQ	<LOQ
cherries, boiled	539 (14%)	620 (1%)	1.150	<LOQ	11
cherries, baked	560 (19%)	649 (16%)	1.159	<LOQ	11
cherries, canned[b]	664 (12%)	394 (15%)	0.594	<LOQ	15 (12%)
green beans, boiled fortified	727 (20%)	456 (16%)	0.627	<LOQ	<LOQ
green beans, boiled greenhouse-treated	4076 (11%)	2374 (4%)	0.582	208 (13%)	68 (1%)
green beans, canned fortified[b]	662 (7%)	191 (21%)	0.288	<LOQ	42 (12%)
green beans, canned greenhouse-treated[b]	2328 (16%)	832 (15%)	0.358	56 (11%)	159 (13%)
OJ, concentrated	1267 (1%)	3352 (1%)	2.645	<LOQ	19 (10%)
peaches, boiled	581 (2%)	290 (31%)	0.498	ND	<LOQ
peaches, canned[b]	461 (2%)	237 (7%)	0.515	ND	14 (10%)
peppers, boiled fortified	676 (43%)	579 (10%)	0.856	<LOQ	<LOQ
peppers, boiled greenhouse-treated	3403 (4%)	4061 (8%)	1.193	22 (3%)	50 (11%)
peppers, baked fortified	640 (17%)	747 (22%)	1.166	<LOQ	13 (6%)
peppers, baked greenhouse-treated	4536 (23%)	3710 (33%)	0.818	25 (14%)	63 (2%)
sweet potato, boiled	526 (10%)	352 (29%)	0.668	18 (24%)	<LOQ
sweet potato, baked pulp[c,d]	451 (17%)	<LOQ	0.022	<LOQ	<LOQ
sweet potato, baked peel[d]	451 (17%)	1014 (17%)	2.249	<LOQ	107 (15%)
sweet potato, baked pulp and peel[e]	451	107	0.224	<LOQ	19
sweet potato, canned[b]	596 (10%)	154 (12%)	0.258	25 (8%)	67 (7%)
winter squash, boiled	818 (7%)	262 (11%)	0.320	17	16 (1%)
winter squash, baked pulp[d]	619 (20%)	728 (6%)	1.176	<LOQ	17 (28%)
winter squash, baked peel[d]	619 (20%)	486 (25%)	NA	<LOQ	10
winter squash, canned[b]	685 (4%)	81 (6%)	0.119	18 (4%)	43 (19%)
water–chlorpyrifos, boiled[f]	253 (2%)	ND	0.040	477 (5%)	606 (1%)
water–TCP, boiled	NA	NA	NA	1330 (0.4%)	1773 (2%)

[a] RSD = relative standard deviation of both samples, if both results were greater than the LOQ; if only one value was greater than the LOQ, that value is reported and no RSD is provided. [b] Entire canned sample (including liquid) analyzed. [c] Cooking factor calculation assumes pulp concentration at LOQ (10 ng/g). [d] Uncooked sample analyzed whole (not divided into pulp and peel). [e] Calculation of weight averaged pulp and peel, assumes pulp concentration at LOQ (10 ng/g). [f] Cooking factor calculation assumes the postcooked water concentration at LOQ (10 ng/g).

LOQs reported for the study were 4.5 and 15 ng/sample, respectively (equivalent to 3 and 10 ng/g). The recovery results, as well as the study LOD and LOQ, indicated that the analytical method was adequate for the quantitation of chlorpyrifos and TCP in the commodities. The residue values obtained for the cooked and uncooked samples were corrected by adjusting the measured values for the daily average recovery of fortified control samples that were analyzed concurrently with the cooked and uncooked produce.

The chlorpyrifos cooking factor was calculated by dividing the concentration measured in the postcooked samples by the concentration measured in the uncooked samples.

$$\text{cooking factor} = \frac{\text{postcooked concentration}}{\text{uncooked concentration}}$$

Whenever possible, the cooking factor compared the commodity as consumed, cooked vs uncooked (apple slices, cabbage wedges, etc.). However, for the baked sweet potatoes and baked winter squash, the peel and the pulp were analyzed separately after cooking but together in the uncooked sample.

RESULTS

Fortification and Treatment. The average uncooked samples contained approximately 720 ng chlorpyrifos/g commodity (**Table 3**). There are several possible reasons for the lower-than-predicted concentrations (target 1000 ng/g). These include the potential for the spray solution to not reach the commodity (sprayed off edge of tray or missed commodity and reached tray), degradation during the storage time, adherence to the plastic wrap, removal upon handling in preparation for storage, etc. Any cause would apply to both the uncooked and the cooked samples therefore not affecting the calculated cooking factors.

The greenhouse-treated green beans used for boiling contained an average residue of 4076 ng chlorpyrifos/g, while the beans used for canning had an average residue of 2328 ng chlorpyrifos/g. The beans used for canning had lower initial residues than those that were boiled because the plants sprayed for canning were fuller and had more beans than the plants sprayed for boiling. The greenhouse-treated peppers contained an average residue of 3969 ng chlorpyrifos/g. The variability of the residues in the uncooked, greenhouse-treated samples was the same as the variability seen in the uncooked, fortified samples (average RSD = 13%).

Fate of Chlorpyrifos and TCP in Boiling Water. The degradation of chlorpyrifos and TCP in boiling water was examined. As shown in **Table 3**, the chlorpyrifos in the fortified, precooked, water samples partially degraded to TCP. The samples may have been inadvertently heated, and the pH of the tap water was not measured, so hydrolysis may have occurred. The samples were fortified at approximately 1000 ng/g, but the measured chlorpyrifos concentration averaged 253 ng/g. TCP was measured in these same samples at an average concentration of 477 ng/g. This was equivalent to 843 ng chlorpyrifos/g (based on molecular weight conversion factor of 1.77, 350.59/198.44) to give a total fortification level of 1096 ng chlorpyrifos/g water. After boiling, no chlorpyrifos was detected, but TCP was measured at an average concentration of 606 ng/g, equivalent to 1071 ng chlorpyrifos/g water. Therefore, chlorpyrifos at elevated temperatures in aqueous

solutions appears to be readily converted to TCP, with virtually no losses due to volatilization. There was no loss of TCP from the boiled TCP-fortified water samples, indicating the stability of TCP during boiling; the slightly higher TCP concentration after boiling resulted from concentration due to loss of water during boiling.

Chlorpyrifos and TCP Concentrations in the Cooked Samples. Sample weights of most produce after boiling remained essentially the same as the precooked weights (average 101%, 68−120%). As expected however, the baked samples generally concentrated during cooking, resulting in an average 74% of the original weight (55−89%), not including the sweet potato and squash, which were divided into pulp and peel after baking. The canned samples were diluted an average of 168% (117−217%); the canned samples were analyzed with the liquid.

Average chlorpyrifos and TCP levels (from duplicates) for the commodities before and after cooking are reported in **Table 3**. Chlorpyrifos concentrations after cooking were dependent upon the commodity and cooking procedure, ranging from 81 to 828 ng/g (fortified whole samples only). All chlorpyrifos levels were well above the LOQ except for sweet potato pulp, thereby enabling calculation of cooking factors. The variability of the chlorpyrifos concentration in the cooked fortified samples (produce and any water when >LOQ) was slightly higher than for the uncooked samples (average RSD = 17 vs 13% for uncooked samples). The variability in the cooked greenhouse-treated samples (produce and any water) was predictably lower, with an average RSD of 12%, due to the composited nature of the samples.

As shown in **Table 3**, generally, the TCP concentrations in the commodities after cooking were similar to the TCP concentrations in the samples that were not cooked. The individual TCP concentrations in the uncooked chlorpyrifos-fortified produce ranged from nondetectable (ND) to 25 ng/g (does not include the greenhouse-treated green beans and greenhouse-treated peppers, which are discussed later). The individual TCP concentrations in the cooked commodities ranged from <LOQ (detectable but not quantifiable) to 67 ng/g. The only commodities and cooking procedure that resulted in a significant increase in TCP concentration (change of >20 ng/g) were the canned sweet potatoes, in which the concentration changed from an average of 25 ng/g to an average of 67 ng/g, and canned winter squash, in which the concentration changed from an average of 18 to 43 ng/g.

In the water remaining after boiling commodities, TCP levels were very low; <LOQ in the broccoli water, cabbage water, fortified green bean water, fortified pepper water, and winter squash water and 9−15 ng/g in the treated green bean water, peach water, treated pepper water, and sweet potato water.

In the greenhouse-treated green bean samples, the chlorpyrifos and TCP concentrations decreased in the commodity as a result of boiling. The TCP concentrations in the uncooked beans averaged 208 ng/g, with this higher level presumably being present as a result of the metabolism of chlorpyrifos to TCP in the plant (*4*, *5*). This concentration decreased to an average of 68 ng/g in the boiled beans (see **Table 3**). Chlorpyrifos and TCP were measured in the water after boiling the beans, at 13−15 and 62−85 ng/g, respectively. On the basis of the total water remaining after boiling (1100 g), the total TCP accounted for would indicate that either chlorpyrifos or TCP was extracted from the beans into the water during boiling, with the majority of any extracted chlorpyrifos being degraded to TCP. In the greenhouse-treated green bean samples that were canned, the chlorpyrifos concentrations also decreased from an average of 2328 ng/g to an average of 832 ng/g, while the TCP concentrations increased from an average of 56 ng/g to an average of 159 ng/g. The chlorpyrifos was degraded to TCP as determined mathematically—the combined TCP and chlorpyrifos residues after canning, expressed as chlorpyrifos equivalents, were approximately equal to the chlorpyrifos residue level of the uncooked sample. The water used to fill the canning jars was not analyzed separately from the beans, so it is possible that the TCP was present in the water phase of the canned sample.

In the treated green pepper samples, the chlorpyrifos and TCP concentrations increased in the commodity as a result of boiling. In addition, both chlorpyrifos and TCP were detected in the water. These results are conflicting because if the chlorpyrifos was extracted into the water, then the concentration in the commodity should have been lower. These data may be reflective of the variability of the commodity residues due to treatment and sampling. In the baked, treated green pepper samples, the chlorpyrifos concentrations decreased while the TCP concentration increased, indicating degradation of chlorpyrifos during baking.

The boiled, baked, concentrated, and uncooked samples were frozen on the day of cooking. The canned samples were stored at room temperature 1−19 days prior to freezing and homogenization. All frozen samples were stored for 9−83 days pending analysis. The storage stability of chlorpyrifos and TCP in various crops has been demonstrated in frozen storage up to 1716 days (*9*). The water samples were stored frozen for 12−92 days. The stability of chlorpyrifos in water stored frozen has been documented (*10*), but because chlorpyrifos degradation was noted in water prior to boiling, it is possible that the degradation in the water occurred before storage due to heating (not boiling), pH, and/or the presence of chlorine.

Cooking Factors. In all but two cases the chlorpyrifos concentration after cooking was quantifiable, so a cooking factor could be calculated. The average cooking factors are reported in **Table 3**, along with the average chlorpyrifos and TCP concentrations before and after cooking.

In the apples, there was a reduction in the chlorpyrifos concentration irrespective of cooking procedure and weight change; the cooking factors were 0.556, 0.609, and 0.661 for boiling, baking, and canning, respectively.

In the broccoli, there was essentially no change in commodity weight and correspondingly little change in the chlorpyrifos concentration, resulting in a cooking factor of 0.935 for boiling.

In the cabbage, there was also very little change in commodity weight, but a slight decrease in chlorpyrifos concentration, resulting in a cooking factor of 0.832 for boiled cabbage.

There was little weight change in cherries that were boiled or baked, but a dilution of approximately 160% occurred during canning. Both the chlorpyrifos and the TCP concentrations increased during boiling and baking, resulting in a cooking factor of 1.15 and 1.16 for boiling and baking, respectively. In canning cherries, there was a significant reduction in the chlorpyrifos concentration, and a slight increase in the TCP concentration, indicating that degradation to TCP occurs to a limited extent. The chlorpyrifos cooking factor was 0.594 for canning cherries. As with the greenhouse-treated canned green beans, the chlorpyrifos was degraded to TCP as determined mathematically—the combined TCP and chlorpyrifos residues after canning, expressed as chlorpyrifos equivalents, were approximately equal to the chlorpyrifos residue level of the uncooked sample.

There was a minimal weight change when boiling green beans, but a significant weight increase (dilution of approximately 210%) occurred during canning. There was a

significant decrease in the chlorpyrifos concentration irrespective of the cooking procedure. The cooking factors for boiling fortified and greenhouse-treated green beans were similar at 0.627 and 0.582, respectively. The cooking factors for canning fortified and greenhouse-treated green beans were also similar, at 0.288 and 0.358, respectively. Note that the cooking factors for canning are approximately half those for boiling, most likely due to dilution of the samples. Also note that there was an increase in the TCP concentration for both fortified and greenhouse-treated canned green beans, which was previously discussed for the greenhouse-treated bean samples.

The orange juice was concentrated approximately 4-fold; however, the average chlorpyrifos cooking factor is 2.6, indicating a loss of chlorpyrifos during concentrating.

There was a minimal weight change for boiled peaches, but canned peaches were approximately 140% of the uncooked weight. There was a significant reduction in the chlorpyrifos residues after both cooking procedures, resulting in cooking factors of 0.498 and 0.515 for boiling and canning, respectively.

The weights of the pepper samples did not change during boiling but were reduced during baking (concentrated to 55−75% of the original weight). Overall, the chlorpyrifos concentrations did not change significantly for cooked peppers. The cooking factors for boiling fortified and greenhouse-treated peppers were similar at 0.856 and 1.19, respectively. As described above, the greenhouse-treated data are puzzling, so the fortified cooking factor of 0.856 may be more realistic. The cooking factors for baking fortified and treated peppers were somewhat diverse, at 1.166 and 0.818, respectively. Degradation to TCP was noted for both the fortified and the treated pepper samples, but a cooking factor greater than one is still possible when combined with a weight decrease.

In the sweet potato samples, there was a minimal weight change during boiling and baking and significant dilution during canning. During boiling and canning, there was a significant decrease in the chlorpyrifos concentration, resulting in cooking factors of 0.668 and 0.258, respectively. In addition, degradation to TCP was evident in the canned samples. After baking, the whole sweet potato was separated into pulp and peel; the total weight change was minimal (approximately 82% of the original weight). As expected, the majority of the chlorpyrifos remained on the peel, with less than quantifiable amounts in the pulp. Taking a worst-case approach and assuming a chlorpyrifos concentration equal to the LOQ (10 ng/g), the cooking factor in the pulp would be 0.022. The cooking factor using the weight percentage and concentration in each fraction, again assuming chlorpyrifos at the LOD in the pulp, was calculated to be 0.224 for a whole baked sweet potato. In-grown chlorpyrifos residues in sweet potatoes are probably present in the pulp rather than on the peel, because only the above-ground portions of the plant are treated, and any residue in the below-ground portion is due to translocation; therefore, a cooking factor of 0.224 may be more realistic.

In the winter squash samples, there was minimal weight change during boiling, significant concentration during baking, and significant dilution during canning. As a result of boiling and canning, there was a significant decrease in the chlorpyrifos concentration, with cooking factors of 0.320 and 0.119. The decline appears to be related to boiling, with the lower cooking factor from canning resulting from the sample weight dilution. The baked squash was treated after being sliced open and with the peel on, so both the peel and the pulp were treated (unlike the sweet potato where only the outside of the potato was treated). Even when comparing whole weight (peel plus pulp) after cooking to the weight before cooking, there was a decrease. The cooking factor was 1.18 for baked pulp. The peel is inedible so a cooking factor was not calculated.

DISCUSSION

Chronic dietary exposure can be calculated using a simple algorithm:

$$E_i = (R_i \times C_i \times P_i)/1000 \quad (1)$$

$$E_t = \sum E_i \quad (2)$$

where E_i is the exposure from the pesticide on food i (mg/kg body weight/day), R_i is the chemical residue on food i (μg residue/g food, μg/g), C_i is the daily consumption of food i (g food/kg body weight/day), P_i is the probability of consuming a certain residue on food i on a single day, and 1000 converts micrograms of residues to milligrams.

If the food in the above equation is considered as a particular type of produce that may be consumed cooked, the equation can be modified as follows:

$$E_i = \sum (R_i \times F_{\text{cooking}} \times C_i \times P_c)/1000 \quad (3)$$

where F_{cooking} is the appropriate cooking factor or one (1) if consumed raw, P_c is the probability of consuming a certain residue on food i cooked by method c, and the total exposure from a particular type of produce i is the sum of all of the ways that the produce can be consumed, whether raw or cooked. For example, the exposure to chlorpyrifos from consumption of cherries could be determined by summing the exposure from fresh, boiled, baked, and canned consumption, each exposure relating the chemical residue on the fresh cherries to the predicted residue on the cooked cherries by use of the appropriate cooking factor. Food consumption data are available from the U.S. Department of Agriculture Continuing Survey of Food Intakes by Individuals (CSFII), conducted from 1989 through 1992 (*11*−*13*). Residue levels of fresh produce can be obtained from field trial data and market basket survey data. The use of cooking factors allows refinement of dietary exposure for those foods in which there are detectable residues in fresh produce, as determined by market basket surveys. In cases where the residue levels are less than quantifiable (<LOQ), the cooking factor may still be used to refine the residue levels from the LOQ. For example, canned squash could use 10% of the LOQ.

It is worth noting that the cooking factors determined for this study may be used for alternate cooking procedures with similar physical characteristics. Baking is heating without moisture, while boiling is heating with excessive moisture. Steaming or sauteeing, which are heating with a small amount of moisture, may be considered to have a cooking factor that is intermediate between boiling and baking.

The cooking factors are directly applicable to the dietary exposure evaluation model (DEEM, version 7.075), a commercially available software package. DEEM can be used to estimate exposure to a pesticide resulting from food consumption by the general U.S. population and certain subpopulations. The model combines the consumption data and residue data for a given pesticide to evaluate dietary risk (*14*). DEEM allows for two adjustment factors, one of which could be the cooking factor, while the other could be for the percent of a crop that might be treated with the particular pesticide.

DEEM contains recipe translation files that convert the food as consumed (e.g., apple pie) into the proper proportions of raw agricultural commodities (e.g., apples, sugar, wheat flour, etc.). By including the cooking factors for the appropriate produce, it would be possible to refine the dietary exposure for the produce as-consumed, whether raw, simply heated, or baked into a pie.

ABBREVIATIONS USED

DAT, days after treatment; LOQ, limit of quantification; LOD, limit of detection; ND, not detected (below the LOD).

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