# **Repeated Pesticide Exposure Among North Carolina Migrant and Seasonal Farmworkers**

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**Background** Limited data document the multiple and repeated pesticide absorption experienced by farmworkers in an agricultural season or their risk factors. **Methods** Data were collected from 196 farmworkers four times at monthly intervals in 2007. Urine samples were tested for 12 pesticide urinary metabolites. Questionnaire data

provided measures of exposure risks. **Results** Farmworkers had at least one detection for many pesticide urinary metabolites; for example, 84.2% had at least one detection for acephate, 88.8% for 3,5,6-trichloro-2pyridinol. Most farmworkers had multiple detections for specific metabolites; for example, 64.8% had two or more detections for acephate, 64.8% for 3,5,6-trichloro-2-pyridinol,

79.1% for 3-phenoxybenzoic acid, and 86.7% for 2,4-dichlorophenoxyacetic acid. Housing type had a consistent significant association with metabolite detections.

**Conclusions** Farmworkers are exposed to multiple pesticides across an agricultural season, and they experience repeated exposures to the same pesticides. Reducing farmworker pesticide exposure and delineating the health outcomes of this exposure require more detailed data. Am. J. Ind. Med. 53:802–813, 2010. © 2010 Wiley-Liss, Inc.

KEY WORDS: exposure biomonitoring; pesticides; health disparities; agricultural health; occupational health; farmworker; minority; housing

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#### INTRODUCTION

Migrant and seasonal farmworker occupational exposure to pesticides is widely acknowledged [Quandt et al., 2006; Calvert et al., 2008; Arcury and Quandt, 2009; Arcury et al., 2009a], and this occupational pesticide exposure is considered an important health risk for farmworkers and their families [Villarejo, 2003; McCauley et al., 2006]. However, no published biomarker data document the number or percent of US farmworkers exposed to pesticides. Although Washington State now collects blood samples from pesticide applicators to measure cholinesterase depression resulting from pesticide exposure [Hofmann et al., 2009a,b], no other state or national surveillance system routinely collects urine or blood biomarkers to estimate the dose of pesticides farmworkers receive, the frequency with which they absorb pesticides, or their cumulative dose from multiple pesticides over extended time periods. Several factors have limited the collection of farmworker pesticide biomarker data. First,

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collecting biological samples from a large group of farmworkers is difficult, as they often live in dispersed and isolated locations, they migrate during the agricultural season, and their transportation and financial resources often limit their ability to appear at a central location for data location. Second, laboratory analysis for pesticide exposure biomarkers is expensive. Finally, the pesticides for which biomarkers are available are dependent on available laboratory techniques.

Existing studies of farmworker pesticide exposure that include biomarkers have been limited to a small number of metabolites, small numbers of participants, cross-sectional samples, or participants in intervention programs [Fenske et al., 2003; Salvatore et al., 2008; Thompson et al., 2008]. For example, Fenske et al. [2003] collected multiple urine samples from 20 apple thinners, and found high concentrations of the dialkylphosphate (DAP) organophosphorus (OP) pesticide urinary metabolite dimethyldithiophosphate (DMDTP). Other studies have documented the exposure of individuals living in farmworker communities [Quandt et al., 2004; Arcury et al., 2005, 2007; Bradman et al., 2005; Coronado et al., 2006; Eskenazi et al., 2007; Thompson et al., 2008]. For example, Thompson et al. [2008] report results based on their two cross-sectional surveys, 1999 and 2003, of approximately 210 Washington State farmworker adults who had a co-resident child. For the 1999 survey, they found that the DAP urinary metabolites dimethylphosphate (DMP), dimethylthiophosphate (DMTP), and DMDTP were detected in 17.1%, 93.7%, and 54.6%, respectively, of the farmworker samples. For 2003, the frequencies of detection were 29.3% for DMP, 92.6% for DMTP, and 55.0% for DMDTP. Arcury et al. [2009a,b] have shown that the proportion of farmworkers with pesticide exposure biomarkers is large. For example, of 939 urine samples collected from 283 farmworkers in 2007, 3,5,6-trichloro-2-pyridinol, a urinary metabolite for the insecticides chlorpyrifos and chlorpyrifos methyl, was detected in 46.2% of samples; 3-phenoxybenzoic acid, a general pyrethroid insecticide urinary metabolite, was detected in 56.4% of samples; and 2,4dichlorophenoxyacetic acid, the urinary metabolite for the herbicide 2,4-D, was detected in 68.1% of samples. These analyses have also shown that the percentage of farmworkers with pesticide exposure biomarkers varies across the agricultural season.

Analyses have not considered whether individual farmworkers have repeated exposures to specific pesticides in a single season based on biomarker data. Documenting the number of times individual farmworkers experience exposure to pesticides across an agricultural season is important. Repeated exposure increases risk for immediate and longterm health outcomes [Alavanja et al., 2004]. Knowing that farmworkers are exposed more than once will inform clinical care for farmworkers, factors needed to reduce exposure, and regulation to decrease exposure. The analysis has two aims. The first aim is to document the number of times (0-4) that the urinary metabolite biomarkers for 12 pesticides commonly used in North Carolina agriculture are detected in individual farmworkers. These are all non-persistent pesticides that are metabolized and excreted from the body within 72 hr of exposure. Therefore, biomarkers that indicate that a pesticide urinary metabolite is present repeatedly at 1 month or greater intervals offer strong evidence of repeated exposure to that pesticide. The second aim is to determine factors associated with the number of times individual farmworkers had urinary metabolite biomarkers for the 12 pesticides.

## **METHODS**

This analysis was part of an ongoing translational research program addressing the health of Latino farmworkers and their families in eastern North Carolina [Arcury et al., 2009a]. This program, Community Participatory Approach to Measuring Farmworker Pesticide Exposure: PACE3, used a community-based participatory research design to document farmworker pesticide exposure, disseminate information on pesticide exposure to farmworkers, and inform pesticide policy. Community partners for this project were the North Carolina Farmworkers Project (NCFP, Benson, NC), and Student Action with Farmworkers (Durham, NC); additional partners include Greene County Health Care, Inc. (Snow Hill, NC), and Columbus County Community Health Center, Inc. (Whiteville, NC). In 2007, the PACE3 collaborators collected urine samples from farmworkers up to four times at monthly intervals to measure the presence of pesticide biomarkers. All sampling, recruitment, data collection, and dissemination protocols, including signed informed consent, were approved by the Wake Forest University School of Medicine Institutional Review Board.

#### Study Sample

The study sample was recruited in 11 eastern North Carolina Counties with large farmworker populations, including Brunswick, Columbus, Cumberland, Greene, Harnett, Johnston, Lenoir, Pitt, Sampson, Wayne, and Wilson. Participant selection and recruitment included two stages. First, farmworker camps were randomly selected from lists of those served by our community partners. Neither a complete list of farmworker camps nor the total number of farmworker camps was available for these Counties; however, our community partners endeavored to maintain complete lists of camps. All 44 camps approached agreed to participate. Second, farmworkers in each of the camps were recruited. In camps with seven or fewer residents, all farmworkers were invited to participate. In camps with more than seven residents, 8-10 farmworkers were recruited. All participants gave signed informed consent. Thirteen farmworkers approached by interviewers chose not to participate, for a participation rate of 95.7%. A total of 287 farmworkers were recruited; however, this analysis was limited to the 196 farmworkers who completed an interview and provided a urine sample at all four observations.

# **Data Collection**

Data included a detailed interview and a first morning void urine sample collected four times at 1-month intervals. Data collection was completed from May to September 2007. Trained data collectors included eight fluent Spanish speakers, divided into three teams. The detailed interview completed with the farmworker participants for each wave of data collection included items on living conditions, pesticide exposure risk factors, health characteristics, work environment, and household environment. At the first wave, the questionnaire also included items on participant personal characteristics (e.g., age, educational attainment). The questionnaire used in these interviews was developed in English and translated by an experienced translator who was a native Spanish speaker familiar with Mexican Spanish. The translated questionnaire was reviewed by four fluent Spanish speakers familiar with farm work, and then pre-tested with 16 Spanish-speaking farmworkers and revised as needed. At the end of each interview, the interviewer gave the participants urine collection containers with labels attached. Participants were instructed to fill the containers with their first void upon rising the next morning. They were asked specifically to only provide their urine in the containers, not that of any other workers in the camp, and they were asked not to put any other fluid or chemicals in the urine containers. Participants placed their urine containers in a cooler with blue ice that was provided to them. Each morning a project interviewer stopped by the camp and retrieved the containers, transported them to the nearest of the three collaborating community partners, aliquotted the samples into labeled containers, and placed them in a laboratory freezer where they were stored at  $-20^{\circ}$ C. These samples were analyzed for pesticide metabolites. Participants were given an incentive valued at \$20 when they completed data collection for each round.

## Laboratory Analysis

The frozen urine samples were shipped overnight on dry ice to a laboratory of the Centers for Disease Control and Prevention in Atlanta, Georgia, for analysis. Samples were analyzed using methods described elsewhere [Olsson et al., 2004; Barr et al., 2007; Montesano et al., 2007]. Briefly, 2 ml urine samples were hydrolyzed by enzymes to liberate the glucuronide- or sulfate-bound conjugated metabolites. Hydrolysates were extracted using a mixed mode solidphase extraction cartridge. Concentrated extracts were analyzed using high performance liquid chromatographytandem mass spectrometry. Two precursor/product ion pairs were analyzed per analyte, one for quantification and one for confirmation. Analyte concentrations were quantified using isotope dilution calibration. The acronyms, analytes, parent chemicals, and limits of detection for the 12 metabolites are reported in Table I. Approximately 10% of the samples tested were positive and negative quality control samples.

TABLE I. Pesticide Urinary Metabolites Included in the Analysis With Analyte, Parent Chemical, and Limit of Detection

Pesticide urinary metabolites	Analyte	Parent chemical	Limit of detection (ng/ml)	
Organophosphorus insecticides				
APE	Acephate	Same	0.28	
TCPy	3,5,6-trichloro-2-pyridinol	Chlorpyrifos; chlorpyrifos methyl	0.2	
Dmet	Dimethoate	Dimethoate	0.03	
MDA	Malathion dicarboxylic acid	Malathion	0.025	
MMP	Methamidaphos	Methamidaphos, acephate	0.64	
Carbamate fungicide				
ETU	Ethylene thiourea	Mancozeb; bis-dithiocarbamates fungicide	0.11	
Pyrethroid insecticides				
3PBA	3-Phenoxybenzoic acid	General pyrethroid metabolite	0.1	
TCC	trans-2,2-(dichloro)-2-Dimethylvinylcyclopropane carboxylic acid	Permethrin; cypermethrin; cyfluthrin	0.4	
CCC	cis-2,2-(dichloro)-2-Dimethylvinylcyclopropane carboxylic acid	Permethrin; cypermethrin; cyfluthrin	0.2	
Herbicides				
2,4-D	2,4-Dichlorophenoxyacetic acid	Same	0.2	
ACE	Acetochlor mercapturate	Acetochlor	0.1	
MET	Metolachlor mercapturate	Metolachlor	0.2	

#### Measures

#### Pesticide metabolites

This analysis was limited to the 12 pesticide urinary metabolites listed in Table I. The OP insecticide urinary metabolites associated with acephate, chlorpyrifos, dimethoate, and malathion were selected as indicators of direct occupational exposure as these pesticides had no residential uses. Each of these insecticides is applied to a crop that farmworkers plant, cultivate, or harvest in North Carolina. Acephate and chlorpyrifos are both applied to tobacco. Dimethoate and malathion are used for numerous vegetable and fruit crops (e.g., asparagus, beans, broccoli, cauliflower, cucumbers, peppers, tomatoes) that are hand planted, cultivated, or harvested in North Carolina. Dimethoate and malathion are also applied to row crops (e.g., cotton and soybeans) that are grown near to where many farmworkers live. Methamidaphos is used for cotton (it has no residential uses), which is often grown in fields near to farmworker camps. Therefore, farmworker contact with dimethoate and malathion, as well as with methamidaphos, may also be through environmental exposure (e.g., drift).

Farmworkers could have had occupational exposure to the carbamate fungicide mancozeb. Mancozeb is used for a wide variety of fruits and vegetables that farmworkers cultivate and harvest. Farmworkers could also experience occupational exposure with several pyrethroid insecticides. Pyrethroid insecticides are replacing OP insecticides in several situations. 3PBA is a general pyrethroid metabolite that cannot be associated with a single pyrethroid insecticide. However, the metabolite 3PBA is not associated with all pyrethroid insecticides; for example, it is not a metabolite of cyfluthrin. The metabolites TCC and CCC are associated with the pyrethroid insecticides permethrin and cyfluthrin. Permethrin is used on a variety of vegetables and fruits that farmworkers cultivate or harvest. Cyfluthrin is applied to cotton in North Carolina, and, although farmworkers do not cultivate or harvest cotton, it is often planted in fields close to where farmworkers live and work.

Farmworkers could have had occupational exposure with several herbicides. 2,4-D and metolachlor are general use herbicides widely used in North Carolina. The herbicide acetochlor is generally used for field corn, but corn fields treated with acetochlor may be rotated to tobacco.

Urine samples to measure the 12 pesticide metabolites were collected across four periods of the agricultural season. Period 1 was May 1–June 8, period 2 was June 9–July 7, period 3 was July 8–August 5, and period 4 was August 6–September 4. These periods roughly correspond to the major phases of eastern North Carolina agriculture; the major activities were: planting tobacco and sweet potatoes in period 1; harvesting cucumbers, topping tobacco, and planting sweet potatoes in period 2; topping and harvesting tobacco in

period 3; and harvesting and curing tobacco in period 4. Two measures of detection were constructed for each of the 12 pesticide urinary metabolites. "At least one detection" indicated whether a farmworker had detections for a pesticide in any one of the four periods of the agricultural season; this measure had the values no detection or at least one detection. "Number of detections" across the agricultural season indicated the number of detections for a urinary pesticide metabolite across the four periods of the agricultural season; this measure had the values 0-4.

Information on the timing of pesticide application relative to the collection of urine samples would be valuable for this study. However, data on the timing of pesticide application were not available. Pesticide applicators in North Carolina are not required to report this information; in the US, only pesticide applicators in California and Florida have reporting requirements. This project involves farmworkers in the research project. If farmers and licensed pesticide applicators were involved in the research, they do not always maintain information on when pesticides are applied. The 1994 US-EPA Worker Protection Standard requires that farmers maintain a record of the pesticides they apply; in 2008 a new North Carolina regulation required that these farmers maintain these records for 2 years.

## Covariates

Farmworker characteristics included in this analysis were limited to characteristics that were stable across the agricultural season and which were expected to affect pesticide exposure. These characteristics included age (18–24, 25–29, 30–39, and 40 or more years), educational attainment (0–6 and 7 or more years), speaks English (yes/no), speaks indigenous language (yes/no), time in US agriculture (1 or less, 2–7, and 8 or more years), worker type (migrant worker without H2A visa, migrant worker with H2A visa, seasonal worker; the H2A visa is a temporary work visa that allows an individual work legally in agriculture for a specified period of months), and housing type (house, barracks, trailer).

#### **Statistical Analysis**

Descriptive statistics were examined for farmworkers in our sample who had a urine sample and interview in all four data collection periods. Frequencies and percents were reported for farmworker characteristics of interest (age, educational attainment, English and indigenous language speaking, seasons in US agriculture, worker type with visa status, and housing type). For each metabolite of interest, the number and percentage of participants were reported that fell into each mutually exclusive detection category (0, 1, 2, 3, or 4 detections). The number and percentage of participants with at least one detection were also summarized for each metabolite. We also examined the distribution of the number of metabolites with at least one detection after combining all 12 metabolites of interest, as well as for groupings of OPs, carbamates, pyrethroids, and herbicides.

We examined the associations of farmworker characteristics with having at least one detection and with having multiple detections for each pesticide urinary metabolite. We did not include the pyrethroid urinary metabolites TCC and CCC for these analyses due to their small numbers of detections. For analyses of multiple detections of a specific metabolite, we grouped the number of detections into categories to reduce the effects of small cell size: APE, TCPy, ETU, 3PBA, 2,4-D, and MET were put in the categories 0 detections, 1 or 2 detections, and 3 or 4 detections; Dmet was put in the categories 0 detections, and 1 or 2 detections; MMP was put in the categories 0 detections, and 1-3 detections; finally, MDA and ACE were put in the categories 0 detections, 1 detection, and 2 or 3 detections. Associations between multiple detections across the specific pesticide urinary metabolites and housing type were limited to all 12 metabolites, the OP metabolites, and the herbicide metabolites, with all 12 metabolites in the categories of 4-6metabolites detected, 7 metabolites detected, and 8-10 metabolites detected; OP metabolites in the categories of 1 or 2 metabolites detected, 3 metabolites detected, and 4 or 5 metabolites detected; and herbicide metabolites in the categories of 1 or 2 metabolites detected, and 3 metabolites detected.

A sensitivity analyses was conducted by performing the same analysis on the larger group of 283 participants in the study. The results are similar compared to the analyses based on the sub-sample of 196. Chi-squared tests (or Fisher's exact tests, when appropriate) were used to examine the associations and the corresponding *P*-values are listed. These data analyses were performed with SAS 9.2 (SAS Institute, Cary, NC) and *P*-values <0.05 were considered statistically significant.

## RESULTS

# **Farmworker Characteristics**

About one-third of the 196 farmworkers included in this analysis was younger than 30 years, about one-third was in their 30s, and about one-third was aged 40 years or older (Table II). About half had 6 or fewer years of education. Fewer than 1 in 10 spoke English, and 14.8% spoke an indigenous language. Many (44.0%) had 8 or more years of experience in US agriculture, while 10.9% were in their first year. Almost 70% of the participants had an H2A visa. About 40% lived in houses and 38.2% lived in trailers, while 17.4% lived in barracks.

**TABLE II.** Personal Characteristics of Farmworkers, Eastern North Carolina,

 2007, 196
 Participants With Four Data Points, and 283
 Participants in the Total

 Sample
 Participants
 Participants
 Participants

	-	ticipants data points	283 participants for the total sample		
<b>Personal characteristics</b>	n	%	n	%	
Age (years)					
18-24	27	13.8	60	21.2	
25-29	42	21.4	55	19.4	
30-39	66	33.7	91	32.2	
40 or more	61	31.1	77	27.2	
Educational attainment					
0–6 years	97	49.5	146	51.6	
7 or more years	99	50.5	137	48.4	
Speaks English	16	8.2	31	11.0	
Speaks indigenous language	29	14.8	64	22.6	
Seasons in US agriculture (year	s) <sup>a</sup>				
1 or less	21	10.9	46	16.4	
2-7	87	45.1	129	46.1	
8 or more	85	44.0	105	37.5	
Worker type/visa status					
Migrant without H2A visa	35	17.9	105	37.1	
Migrant with H2A visa	137	69.9	146	51.6	
Seasonal	24	12.2	32	11.3	
Housing type					
House	83	42.4	117	41.3	
Barracks	34	17.4	58	20.5	
Trailer	79	40.3	108	38.2	

<sup>a</sup>3 missing observations.

The 196 farmworkers included in this analysis are similar in most regards to the larger sample of 283 farmworkers that participated in the study. Differences include the lower percentage in this analytical sample of farmworkers who spoke an indigenous language (14.8% vs. 22.6%), and different percentages of migrants without and with an H2A visa. The percentage without an H2A visa changed from 37.1% to 17.9%, while the percent with an H2A visa changed from 51.6% to 69.9%.

## At Least One Detection for Each Pesticide Urinary Metabolite

The percentage of farmworkers with at least one detection was substantial for many of the pesticide urinary metabolites (Table III). Among the urinary metabolites for the OP insecticides, 84.2% of the individual farmworkers had a least one detection for APE, 88.8% had at least one detection for TCPy, and 73.5% had at least one detection for MDA. Over one-third (34.2%) had at least one detection for

	At least one detection (N $=$ 196)		Number of detections (n $=$ 196)									
	n	n %	Zero		o One		Two		Three		Four	
			n	%	n	%	n	%	n	%	n	%
Organophosphorus insecticides												
APE (acephate)	165	84.2	31	15.8	38	19.4	76	38.8	36	18.4	15	7.7
TCPy (chlorpyrifos)	174	88.8	22	11.2	47	24.0	65	33.2	42	21.4	20	10.2
Dmet (dimethoate)	37	18.9	159	81.1	32	16.3	5	2.6	0		0	
MDA (malathion)	144	73.5	52	26.5	90	45.9	46	23.5	8	4.1	0	
MMP (methamidaphos; acephate)	67	34.2	129	65.8	60	30.6	5	2.6	2	1.0	0	
Carbamate fungicide												
ETU (mancozeb)	116	59.2	80	40.8	69	35.2	33	16.8	13	6.6	1	0.5
Pyrethroid insecticides												
3PBA (pyrethroid)	191	97.4	5	2.6	36	18.4	76	38.8	58	29.6	21	10.7
TCC (permethrin)	16	8.2	180	91.8	15	7.7	1	0.5	0		0	
CCC (permethrin)	2	1.0	194	99.0	2	1.0	0		0		0	
Herbicides												
2,4-D	192	98.0	4	2.0	22	11.2	40	20.4	71	36.2	59	30.1
ACE (acetochlor)	184	93.9	12	6.1	120	61.2	59	30.1	5	2.6	0	
MET (metolachor)	88	44.9	108	55.1	49	25.0	23	11.7	14	7.1	2	1.0

TABLE III. Number of Participants by Number of Detections for Pesticide Urinary Metabolites, Eastern North Carolina, 2007 (n = 196)

MMP, and almost one in five (18.9%) had at least one detection of Dmet. Almost three in five (59.2%) farmworkers had at least one detection for ETU, the carbamate urinary metabolite. Almost all (97.4%) farmworkers had at least one detection for the general pyrethroid urinary metabolite 3PBA. Only a few farmworkers had detections for the pyrethroid urinary metabolites TCC (8.2%) and CCC (1.0%). Almost all of the farmworkers had at least one detection for the herbicide urinary metabolites 2,4-D (98.0%) and ACE (93.9%). Over two in five (44.9%) farmworkers had at least one detection for the herbicide urinary metabolites TCC (8.2%) and MET.

## Multiple Detections for Pesticide Urinary Metabolites

A large number of farmworkers had multiple detections for several of the specific pesticide urinary metabolites (Table III). Considering the OP insecticides, almost two in five (38.8%) had two detections, almost one in five (18.4%) had three detections, and 7.7% had four detections of APE. Similarly, for TCPy, 33.2% had two detections, 21.4% had three detections, and 10.2% had four detections. Although none of the farmworkers had four detections for MDA, 23.5% had two detections, and 4.1% had three detections. Few farmworkers had multiple detections for Dmet and MMP. Over one in five (23.9%) participants had multiple detections for ETU. For the pyrethroid insecticide metabolite 3PBA, almost two in five (38.8%) had two detections, 29.6% had three detections, and 10.7% had four detections.

Multiple detections were common for each of the herbicide urinary metabolites. Thirty percent of the farm-workers had four detections of 2,4-D, 36.2% had three detections, and 20.4% had two detections. Although no farmworkers had four detections of ACE, 30.1% had two detections, and 2.6% had three detections. One percent of the farmworkers had four detections for MET, with 7.1% having three detections, and 11.7% having two detections.

A diverse array of specific pesticide urinary metabolites was detected in most farmworkers (Table IV). Almost twothirds of the farmworkers had at least one detection for seven or more of the 12 specific pesticide urinary metabolites. Over two-thirds of the farmworkers had at least one detection for three or more of the five specific OP pesticide urinary metabolites. Over 9 in 10 farmworkers had at least one detection for two or three of the three herbicide pesticide urinary metabolites.

### **Predictors of Detection**

Housing type is the only farmworker characteristic that had a consistent significant association with at least one detection and with multiple detections across most of the metabolites. Fewer farmworkers living in a barracks, compared to those living in a house or those living in a trailer, had a least one detection of APE (P = 0.0004), Dmet **TABLE IV.** Number of Metabolites for Which Farmworkers had At Least One Detection, Eastern North Carolina, 2007 (n = 196)

Pesticide class	Number of metabolites for which farmworkers had at least one detection	n	%
All 12 metabolites	4	4	2.0
	5	23	11.7
	6	41	20.9
	7	60	30.6
	8	41	20.9
	9	19	9.7
	10	8	4.1
Organophosphorus insecticides	1	5	2.5
	2	56	28.6
	3	79	40.3
	4	47	24.0
	5	9	4.6
Carbamate fungicide	0	80	40.8
	1	116	59.2
Pyrethroid insecticides	0	5	2.6
	1	175	89.3
	2	14	7.1
	3	2	1.0
Herbicides	1	12	6.1
	2	100	51.0
	3	84	42.9

(P = 0.0062), MMP (P = 0.0047), 3PBA (P = 0.0203), and MET (P < 0.0001; Table V). Fewer farmworkers in a house had at least one detection of ACE than those living in a trailer or a barracks (P = 0.0099). Fewer farmworkers living in a barracks, compared to those living in a house or those living in a trailer, had multiple detections of APE (P = 0.0012), MMP (P = 0.0047), 3PBA (P = 0.0079), ACE (P < 0.001), and MET (P < 0.001; Table V). Equivalent percentages of farmworkers living in barracks and houses had multiple detections of 2,4-D, and these percentages were lower than for farmworkers living in trailers (P = 0.048).

Housing type is also the only farmworker characteristic with a consistent significant association with at least one detection for multiple specific pesticide metabolites (Table VI). Of farmworkers living in barracks, 17.7% had at least one detection for seven of the 12 specific urinary metabolites and 17.7% had at least one detection for 8-10 of the 12 specific urinary metabolites; the comparable percentages for farmworkers living in houses were 30.1% and 43.4%, respectively, and for farmworkers living trailers were 36.7% and 32.9%, respectively. For detections of OP pesticide urinary metabolites, of those living in barracks 44.1% had at least one detection for three of the five and 5.9% had at last one detection for four or five of the five during the

agricultural season. The comparable percentages for farmworkers living in houses were 33.7% and 37.4%, respectively, and for farmworkers living trailers were 45.6% and 29.1%, respectively. For detections of the three herbicide urinary metabolites, of those living in barracks 14.7% had at least one detection for all three; the comparable percentage for farmworkers living in houses was 57.8%, and for farmworkers living trailers was 39.2%.

Sensitivity analysis resulted in few changes in associations of housing type and pesticide urinary metabolite detections. When all 283 farmworkers were included in the analysis, housing type no longer had a significant association with the number of Dmet detections or with at least one detection for multiple of OP pesticide urinary metabolites.

One pesticide urinary metabolite, APE, had a consistent set of factors associated with multiple detections. In addition to the association of multiple detections of APE with housing type, multiple detections of APE were associated with educational attainment and seasons in US agriculture. Of those with 0–6 years of education, 34.0% (33) had 3 or 4 detections of APE, while among those with 7 or more years of education, 18.2% (18) had 3 or 4 detections (P = 0.0227). Of those with one or less season in US agriculture, 14.3% (3) had 3 or 4 detections of APE, of those with 2–7 seasons, 18.4% (16) had 3 or 4 detections, and among those with 8 or more seasons, 37.7% (32) had 3 or 4 detections (P = 0.0239).

#### DISCUSSION

This analysis shows that most farmworkers are exposed to an array of pesticides across an agricultural season and that many farmworkers are repeatedly exposed to the same pesticides across an agricultural season. At least one urine sample collected from the great majority of farmworkers contained the pesticide metabolites associated with the OP insecticides acephate (APE), chlorpyrifos (TCPy), malathion (MDA), pyrethroid insecticides (3PBA), and the herbicides 2,4-D and acetochlor. At least one urine sample collected from a substantial majority of farmworkers contained ETU, the metabolite for the carbamate fungicide mancozeb. At least one urine sample collected from substantial numbers of farmworkers provided urine samples in which the metabolites for methamidaphos (MMP), an OP insecticide, and metolachor (MET), an herbicide, were detected.

The majority of farmworkers had more than one detection and, therefore, more than one exposure, for the OP pesticide urinary metabolites APE (acephate) and TCPy (chlorpyrifos), the pyrethroid insecticide urinary metabolite 3PBA, and the herbicide urinary metabolite 2,4-D. Between about one-fifth and one-third of the farmworkers had more than one detection for the pesticide urinary metabolites MDA (malathion), ETU (mancozeb), ACE (acetochlor), and MET (metolachlor). Similarly, the majority of farmworkers had detections for numerous pesticide urinary metabolites at least

**TABLE V.** Association of Housing Type With At Least on Detection and With Grouped Numbers of Detections for Pesticide Urinary Metabolites, Eastern North Carolina, 2007 (n = 196)

	Ho	ouse	Bar	racks	Tra	ailer	Detec	tections ( <i>P</i> )	
Metabolites and number of							None versus at	Grouped number	
detections	n	%	n	%	n	%	least one <sup>a</sup>	of detections <sup>a</sup>	
Organophosphorus insecticides									
APE							0.0004	0.0012	
0	10	12.1	13	38.2	8	10.1			
1-2	48	57.8	18	52.9	48	60.8			
3-4	25	30.1	3	8.8	23	29.1			
ТСРу							ns	ns	
0	13	15.7	2	5.9	7	8.9			
1-2	45	54.2	23	67.7	44	55.7			
3-4	25	30.1	9	26.5	28	35.4			
Dmet							0.0062	0.0062	
0	60	72.3	33	97.1	66	83.5			
1-2	23	27.7	1	2.9	13	16.4			
MDA							ns	ns	
0	22	26.5	7	20.6	23	29.1			
1	40	48.2	13	38.2	37	46.8			
2–3	21	25.3	14	41.2	19	24.1			
MMP							0.0047	0.0047	
0	47	56.6	30	88.2	52	65.8			
1–3	36	43.4	4	11.8	27	34.2			
Carbamate fungicide	00	10.1		11.0	<u> </u>	0 1.2			
ETU							ns	ns	
0	33	39.8	11	32.4	36	45.6	110	no	
1-2	43	51.8	21	61.8	38	48.1			
3-4	7	8.4	2	5.9	5	6.3			
Pyrethroid insecticides	1	0.4	2	0.0	5	0.0			
3PBA							0.0203	0.0079	
0	2	2.4	3	8.8	0		0.0203	0.0079	
1–2	48	57.8	24	70.6	40	50.6			
3-4	40 33	39.8	24 7	20.6	40 39	50.6 49.4			
Herbicides	33	39.0	1	20.0	39	49.4			
							20	0.049	
2,4-D	0	0.4	0	FO	0		ns	0.048	
0	2	2.4	2	5.9	0	044			
1-2	32	38.6	11	32.4	19	24.1			
3-4	49	59.0	21	61.8	60	76.0	0.0000	.0.001	
ACE	10	10.1				10	0.0099	< 0.001	
0	10	12.1	1	2.9	1	1.3			
1	41	49.4	32	94.1	47	59.5			
2-3	32	38.6	1	2.9	31	39.2			
MET		_					< 0.0001	< 0.001	
0	31	37.4	29	85.3	48	60.8			
1-2	39	47.0	5	14.7	28	35.4			
3-4	13	15.7	0		3	3.8			

ns, not significant.

<sup>a</sup>Chi-squared statistic used to test differences in detection across housing type.

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FABLE VI. Association of Housing Type With Number of Multiple Detections Across the Specific Pesticide Urinary Metabolites, Eastern North Carolina, 2007
(n = 196)

No Pesticide class	Number of metabolites for which farmworkers had at least one detection	House		Barrack		Trailer		
		n	%	n	%	n	%	Р
All 12 metabolites	4-6	22	26.5	22	64.7	24	30.4	0.001
	7	25	30.1	6	17.7	29	36.7	
	8-10	36	43.4	6	17.7	26	32.9	
Organophosphorus	1-2	24	28.9	17	50.0	20	25.3	0.005
insecticides	3	28	33.7	15	44.1	36	45.6	
	4-5	31	37.4	2	5.9	23	29.1	
Herbicides	1–2	35	42.2	29	85.3	48	60.8	< 0.0001
	3	48	57.8	5	14.7	31	39.2	

once across the agricultural season. All of the farmworkers had detections for at least four different urinary metabolites, and most of the farmworkers had detections for at least seven different urinary metabolites. The majority of the farmworkers had detections for at least three different OP pesticide urinary metabolites and for at least two different herbicide urinary metabolites.

The type of housing in which the farmworkers lived was consistently associated with the detection of pesticide metabolites in their urine, the number of different pesticide metabolites detected in their urine, and the number of times most specific pesticide metabolites were detected in their urine. Fewer farmworkers who lived in barracks as opposed to houses or trailers had detections of the pesticide urinary metabolites. The houses in which farmworkers in North Carolina live tend to be old farm houses and outbuildings (e.g., old tobacco barns and pig farrowing barns) converted to dormitory use [Vallejos et al., 2009]. The trailers used for farmworker housing in North Carolina tend be older, used trailers [Vallejos et al., 2009]. Many houses and trailers are in poor repair with old carpeting, holes in the floors, walls, and ceilings, and inadequate ventilation. Therefore, houses and trailers may be more difficult to clean than barracks, and may offer more opportunities for pesticides to enter the dwelling. Barracks are built especially for farmworker housing, are newer and in better repair, and have concrete floors. They have had less opportunity to accumulate pesticides and they are often easier to clean (i.e., they can be hosed out each year). Of course, housing in itself cannot cause pesticide exposure. Housing may be an indicator of other unmeasured factors that affect pesticide exposure.

The particular association of several farmworker personal characteristics with APE (acephate) is also important. Acephate is widely used to treat tobacco [Southern and Sorenson, 2008], and almost all of the farmworkers participating in this study worked extensively in tobacco [Arcury et al., 2009a,b]. Farmworkers with greater experience in US agriculture, as measured by numbers of seasons worked, had more detections of APE; this may reflect less concern with exposure among those with the greatest experience. Farmworkers with more education had fewer multiple detections for APE; this may again reflect greater ability to read pesticide safety materials. Unfortunately, no other analyses of the prevalence of farmworker acephate exposure based on biomarker are available for comparison with our results. Lonsway et al. [1997] report the amount of acephate deposited on the clothes of agricultural workers involved in mixing and spraying pesticides on tobacco. Curwin et al. [2003] show that hand washing is effective in removing acephate from the hands of tobacco workers.

These results add to the epidemiological knowledge of the risks for pesticide exposure among farm laborers. Other analyses of these data have shown that the proportion of farmworkers with detections for pesticide metabolites is large and that the proportion of farmworkers with detections varies across the agricultural season [Arcury et al., 2009a,b]. These results show that individual farmworkers have multiple detections and, therefore, multiple exposures for numerous pesticides across the agricultural season and for multiple exposures to the same pesticide across the agricultural season. All of the pesticide metabolites considered in this analysis are non-persistent and excreted within 72 hr of exposure. Urine samples were collected at monthly intervals, with the first samples collected after the participants had been in North Carolina for several weeks. Therefore, each detection must be considered a different exposure that occurred while the farmworker was in North Carolina.

These results differ from the results of other studies in that we have repeated measures of pesticide exposure from a large, broadly selected sample of farmworkers with a high participation rate. Studies with a single cross-sectional or pre-post design (e.g., strawberry workers before and after pesticide application in strawberry fields) are important for understanding the exposure of farmworkers to specific pesticides [Fenske et al., 2003; Coronado et al., 2006; Salvatore et al., 2008]. However, these studies cannot document the multiple exposures to pesticides that farm-workers experience. Yet data on the extent of chronic pesticide exposure, as that which is documented by this study, is needed to delineate the health effects of such chronic, if low level, exposure [Alavanja et al., 2004].

Although the design for the PACE3 study has several strengths, its limitations should be considered in evaluating its results. PACE3 was conducted in a limited area of one state; other areas and states may differ in their patterns of pesticide use and exposure. The sample was limited to the camps known to the community partners, and participants were limited to those living in the camps at the time of recruitment. We collected pesticide biomarker data for an extended part of the agricultural season; however, the period covered did not include workers harvesting cucumbers and sweet potatoes in September and October. The specific pesticide metabolites and the number of detections were limited to the capabilities of the existing laboratory procedures. In addition, metabolites can also be derived from exposure to the preformed metabolites in the environment or from pesticide exposures in non-occupational pathways including in housing and food [Zhang et al., 2008].

Several of the pesticide metabolites detected in the urine samples of the farmworkers who participated in this study are also commonly detected in the general US population. For example, Barr et al. [2005] use 1999-2000 National Health and Examination Survey data to show that 91.0% of persons aged 6-59 years had a detection of the metabolite TCPy, and 52.0% had a detection of the metabolite MDA. Chlorpyrifos, the parent pesticide for the urinary metabolite TCPy, was widely used in residential applications before it was banned for residential use at the end of 2001. Julien et al. [2008] report on increased pyrethroid insecticide loadings in general residential settings as pyrethroids replace organophosphate pesticides for residential use. Therefore, it could be argued farmworker exposure to these pesticides resulted from sources other than occupational exposure. However, several of metabolites detected in the urine samples from large numbers of farmworker participants, such as the metabolites APE, ETU, ACE, and MET are limited to agricultural use. This demonstrates an agricultural basis for exposure to these pesticides.

Farmworkers, and probably all agricultural workers, are often exposed to the same pesticides multiple times across an agricultural season. It is possible that some farmworkers are continuously exposed to some pesticides year round. Therefore, farmworkers are exposed multiple times to the same pesticides across their work lives. It is also apparent that farmworkers are exposed to multiple pesticides, multiple times across an agricultural season. Research to understand the potential effects of pesticide exposure on farmworker health, particularly long-term health outcomes from repeated or continuous low-level pesticide exposure, must be more realistic in measuring exposure. A single, cross-section assessment of pesticide exposure does not reflect either individual or population exposure.

Healthcare providers, advocates, and investigators have expressed concern for the health effects of pesticide exposure for farmworkers in the US as well as in other nations [e.g., London and Ballie, 2001; London, 2003; Kunstadter, 2007; Arcury and Quandt, 2009]. Establishing the effects pesticides have on farmworker health beyond immediate intoxication is difficult [Zahm and Blair, 1997, 2001; Zahm et al., 2001]. The lack of documentation for the number of different pesticides to which farmworkers are exposed, and the number of times they are exposed to these pesticides has limited understanding the potential health effects. For example, all of the OP, carbamate, and pyrethroid pesticides are neurotoxins. These results indicate that farmworkers are not exposed to a single insecticide in a season, but often are exposed to multiple insecticides, multiple times across a season. The combination of pesticides, the number of exposures in a season, and the size of the dose received with each exposure must be considered in estimating the effect of exposure on neurological and other health outcomes. Understanding that there is a cumulative dose from multiple pesticides over extended periods is important for improved risk assessment and risk mitigation.

These results are important for policy. Too often individuals opposed to greater pesticide safety regulation have argued that farmworkers suffer little exposure to pesticides and, therefore, more regulation is not needed. These results show that this argument is false. Other analyses of PACE3 data document that about one-quarter of farmworkers do not get the training required by current regulations, and, of those who receive training, about onequarter do not understand the training they receive [Whalley et al., 2009]. These results also document that many farmworkers are not provided the hygiene facilities in the fields (e.g., water to wash, soap, towels) or laundry facilities to meet current regulation. The results from these different analyses argue for greater enforcement for existing pesticide safety regulations and for stronger pesticide safety regulations in agriculture. Finally, this analysis indicates that housing is an important correlate of pesticide exposure for farmworkers. Efforts to enforce and improve existing housing standards for migrant farmworkers may reduce pesticide exposure as well as improve the general quality of life for farmworkers [Vallejos et al., 2009].

Continuing research on farmworker pesticide exposure is needed. This research has not been able to fully explicate the number of times farmworkers are exposed to pesticides across an agricultural season and the factors causing these exposures with only four data points. Research is needed that will expand the collection of farmworker pesticide exposure epidemiological data beyond North Carolina. Research is also needed to collect farmworker exposure data daily for an extended period across the agricultural season. This research should include measures of immediate health status. Finally, research is needed in which farmworker pesticide exposure and health outcomes are examined longitudinally.

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