

Variation Across the Agricultural Season in Organophosphorus Pesticide Urinary Metabolite Levels for Latino Farmworkers in Eastern North Carolina: Project Design and Descriptive Results

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Background *Community Participatory Approach to Measuring Farmworker Pesticide Exposure, PACE3, used a longitudinal design to document pesticide biomarkers among farmworkers. This article presents an overview of PACE3 and provides a descriptive analysis of participant characteristics and one set of pesticide biomarkers, the dialkylphosphate (DAP) urinary metabolites of organophosphorus (OP) pesticides.*

Methods *Two hundred eighty seven farmworkers were recruited during 2007 from 44 farmworker camps in 11 eastern North Carolina counties. Participants provided interviews, urine samples, blood samples, and saliva samples up to four times at monthly intervals beginning in May. A total of 939 data points were collected.*

Results *Farmworkers were largely men (91.3%) from Mexico (94.8%) with a mean age of 33.7 years (SE 0.82); 23.3% spoke an indigenous language. Across all data points, frequencies of detection and median urinary concentrations were 41.3% and 0.96 µg/L for dimethylphosphate (DMP), 78.3% and 3.61 µg/L for dimethylthiophosphate (DMTP), 33.3% and 0.04 µg/L for dimethyldithiophosphate (DMDTP), 40.5% and 0.87 µg/L for diethylphosphate (DEP), 32.3% and 0.17 µg/L for diethylthiophosphate (DETP), and 8.09% and 0.00 µg/L for diethyldithiophosphate (DEDTP). The frequencies of detection and urinary concentrations of the DAP metabolites increased during the season.*

Conclusions *More PACE3 participants were from Mexico, male, migrant workers, and spoke an indigenous language compared to national data. PACE3 participants had comparable frequencies of detection and urinary metabolite concentrations with participants in other studies. Variability in the frequencies of detection and urinary concentrations of the DAP metabolites indicates the importance of longitudinal studies of biomarkers of currently used pesticides in farmworker populations. Am. J. Ind. Med. 52:539–550, 2009. © 2009 Wiley-Liss, Inc.*

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INTRODUCTION

Pesticides are widely used in US agriculture, and everyone employed in agriculture is exposed to these pesticides. Although procedures to measure pesticide exposure continue to improve, data on the actual levels of pesticide exposure among agricultural workers are very limited. Migrant and seasonal farmworkers are largely minority workers who have little control of their occupational safety, including pesticide exposure [Carroll et al., 2005; Arcury and Quandt, 2007; Arcury et al., 2006]. Recent reviews indicate that little research has documented the levels of pesticide exposure among farmworkers or the health effects of this exposure, and they emphasize the need to collect biomarker data from large representative samples of farmworkers at several points across the agricultural season [Quandt et al., 2006; McCauley et al., 2006]. Longitudinal studies obtaining repeated assessments would better document the actual exposures and doses of pesticide experienced by farmworkers and provide the foundation for determining the health effects of these pesticide doses.

Although little research documents farmworker pesticide exposure, analysts suggest that farmworkers are exposed to high levels of pesticides [Fenske et al., 2003; Quandt et al., 2006; Calvert et al., 2008]. Few studies have collected biomarker indicators of pesticide exposure for farmworkers, and only one has collected biomarker indicators multiple times for each worker. The biomarkers typically measured in pesticide exposure research have included the six dialkylphosphate (DAP) urinary metabolites of organophosphorus (OP) insecticides: dimethylphosphate (DMP), dimethylthiophosphate (DMTP), dimethyldithiophosphate (DMDTP), diethylphosphate (DEP), diethylthiophosphate (DETP), and diethyldithiophosphate (DEDTP). The focus on the DAP urinary metabolites is appropriate, as they reflect exposure to a large number of the OP insecticides, and these remain the most widely used insecticides in US agriculture. The DAP urinary metabolites are indicators of current OP insecticide exposure, as these metabolites are excreted by adults within 72 hr [Bravo et al., 2004].

Most pesticide exposure research has been conducted with farmworkers employed in Washington State. Fenske and colleagues [2003] collected multiple urine samples from 20 apple thinners, and found high concentrations of the OP pesticide urinary metabolite DMTP. Thompson and colleagues [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 DMP, DMTP, and DMDTP were detected in 17.1, 93.7, and 54.6%, respectively, of the farmworker samples, with median concentrations less than the limit of detection (LOD), 9.7 and 1.2 $\mu\text{g/L}$, respectively. The frequency of detection and concentrations were greater for the 2003 compared to the 1999 survey participants. For 2003, the frequency of detection and median concentrations were 29.3% and less than the LOD for DMP, 92.6% and 65.2 $\mu\text{g/L}$ for DMTP, and 55.0% and 6.0 $\mu\text{g/L}$ for DMDTP. These data indicate substantial variability in these pesticide biomarkers from year to year.

Studies of farmworkers employed in states other than Washington are more limited. Arcury et al. [2005] examined DAP urinary metabolites in farmworkers who lived in eight family dwellings in western North Carolina. Data were collected in 2001–2002. They reported that for the 13 farmworkers living in these family dwellings from whom samples were collected, the number of detections for DMP was 12 (92%), for DMTP was 10 (77%), for DMDTP was 7 (54%), for DEP was 10 (77%), for DETP was 13 (100%), and for DEDTP was 5 (38%). Concentrations of the metabolites were generally higher than US population data from NHANES [Barr et al., 2004].

Eskenazi and colleagues [2007] conducted a study with a large sample of women in a California agricultural community. They found DAP metabolite concentrations in the women that were higher than those in the general US population. In addition, they observed significant associations of urinary concentrations with subsequent health outcomes in the children. While this is an important study, it does not address the occupational pesticide exposure of farmworkers.

This article provides an overview of a research project that recruited a representative sample of 287 farmworkers in eastern North Carolina during the 2007 agricultural season and collected data to measure biomarkers (urinary metabolites, cholinesterase) of pesticide exposure. This article also presents a descriptive analysis of study participant personal characteristics and urinary DAP metabolite concentrations.

MATERIALS AND METHODS

Community Participatory Approach to Measuring Farmworker Pesticide Exposure: PACE3 (R01 ES08739) is an ongoing translational research program addressing the health of Latino farmworkers and their families in eastern North Carolina. The primary community partner for this

project is the North Carolina Farmworkers Project (NCFP) (Benson, NC); additional partners include Greene County Health Care, Inc. (Snow Hill, NC), and Columbus County Community Health Center, Inc. (Whiteville, NC). PACE3 used a longitudinal design in which data were collected from participants up to four times at monthly intervals in 2007. All sampling, recruitment, and data collection protocols, including signed informed consent, were approved by the Wake Forest University School of Medicine Institutional Review Board.

Conceptual Framework

The design of PACE3 is based on the conceptual framework proposed by Quandt and colleagues [2006]. Farmworker pesticide exposure can be understood as the result of proximal, distal, and moderating forces. Proximal determinants are workplace and household behaviors that bring workers into contact with pesticides. These behaviors are themselves the result of the corresponding environments. Thus, work environments that have regular, effective safety training and have an organization of work in which workers are able to exercise some degree of judgment in their work practices are more likely to have workers who exhibit good pesticide safety behaviors (e.g., regular hand washing). However, beliefs or psychosocial stressors may reduce the effects of a supportive environment on actual behaviors. Community factors may affect the work and home environments, as well. If pesticides are ubiquitous, the work and home environments may have levels of pesticides that are beyond the control of workers. While the conceptual framework recognizes multiple pathways for pesticide exposure, these multiple pathways are not addressed in this descriptive analysis.

Locale

Data collection was completed in 11 counties with large farmworker populations, including Brunswick, Columbus, Cumberland, Greene, Harnett, Johnston, Lenoir, Pitt, Sampson, Wayne, and Wilson Counties. For these counties in 2007, conservative estimates by the North Carolina Employment Security Commission put the number of migrant farmworkers without H2A visas at 13,675 (36.2% of those in North Carolina), the number of migrant farmworkers with H2A visas at 2,995 (34.3%), and the number of seasonal farmworkers at 5,800 (22.8%). The agricultural production in these counties varies, but the major hand-cultivated and hand-harvested crops include tobacco, sweet potatoes, and cucumbers.

Sample

PACE3 used a two-stage procedure to select farmworkers. First, the three partnering agencies prepared lists of

farmworker camps for the counties that they served. Camps were approached in order until each agency recruited a minimum number of camps and a specified number of participants. All camps that were approached agreed to participate. At the end of the first round of data collection NCFP had recruited 131 participants at 20 camps, Greene County Health Care, Inc., had recruited 64 participants at 11 camps, and Columbus County Community Health Center, Inc., had recruited 66 participants at 10 camps, for a total of 261 participants at 41 camps. NCFP encountered problems with three of the initially recruited camps, and these camps were replaced. Therefore, 44 farmworker camps participated in the study.

Second, participants 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, eight to ten farmworkers were recruited with interviewers recruiting participants as they became available. A total sample of 287 farmworkers was recruited: 261 at the first round of data collection, and 26 at the second round of data collection. Of all farmworkers approached by the interviewers, 13 chose not to participate, for a participation rate of 95.7%. At the second round of data collection, 41 participants were lost to follow-up, 20 were lost at the third round, and 12 were lost at the fourth round. Four rounds of data collection were completed with 197 farmworkers, three rounds with 27, two rounds with 14, and one round with 49.

Data Collection

Data collection included four components: a detailed interview, a finger stick blood sample to measure cholinesterase, a saliva sample for genetic analysis, and a first morning urine void to measure pesticide metabolites and metals. Participants were given an incentive valued at \$20 when they completed data collection for each round.

Data collection was completed from May through September 2007. Data collectors included eight fluent Spanish speakers, divided into three teams. One team was affiliated with the camps served by each of the three partnering agencies. A detailed interview was completed with the farmworker participants at each round of data collection. At every contact the questionnaire included items on living conditions and recent (in the 3 days before the interview) risk factors for pesticide exposure, including workplace activities and behaviors, household behaviors, psychosocial stressors, work environment, and household environment. At the first contact, the questionnaire also included items on participant personal characteristics (e.g., age, educational attainment) and current health status. 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.

Validated Spanish language versions of scales were used. 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.

Blood samples to measure cholinesterase were collected on blotter paper at each of the four interviews. Saliva samples for genetic analysis and first morning urine samples to measure metals were collected at the initial interview with each participant. Cholinesterase, metals, and genetic data are not discussed in this article.

For the measurement of pesticide urinary metabolites, 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 assured that the urine samples would be tested for agricultural chemicals and metals only, and not for the use of alcohol, drugs, or any health conditions. They were asked specifically to only provide their urine in the containers, not that of any other workers in the camp. 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, aliquoted the samples into labeled containers, and placed them in a laboratory freezer where they were stored at -20°C . The urine samples were shipped on dry ice to the Centers for Disease Control and Prevention in Atlanta, Georgia, using an overnight delivery service. These samples were analyzed for pesticide metabolites.

Laboratory Analysis

Six urinary DAP metabolites of OP pesticides (DMP, DMTP, DMDTP, DEP, DETP, and DEDTP) were measured in urine samples using the method of Bravo et al. [2004]. Urine samples were thawed to room temperature. A 2-mL aliquot of each sample was fortified with isotopically labeled internal standards, and then mixed. The urine samples were lyophilized overnight to remove all traces of water. The residue was dissolved in acetonitrile and diethyl ether and the DAP metabolites were chemically derivatized to their respective chloropropyl phosphate esters. The reaction mixture was concentrated, and the phosphate esters were measured using gas chromatography-positive chemical ionization-tandem mass spectrometry in the multiple reaction monitoring mode. Unknown analyte concentrations were quantified using isotope dilution calibration with calibration plots generated with each sample run. The reported LODs were 0.6 $\mu\text{g/L}$ for DMP, 0.2 $\mu\text{g/L}$ for DMTP, 0.1 $\mu\text{g/L}$ for DMDTP, 0.2 $\mu\text{g/L}$ for DEP, 0.1 $\mu\text{g/L}$ for DETP, and 0.1 $\mu\text{g/L}$ for DEDTP. To ensure quality data, additional

quality control materials, fortified samples, and blank samples were analyzed in parallel with all unknown samples.

Measures

The agricultural season is divided into four periods. Period 1 was May 1 to June 8, Period 2 was from June 9 to July 7, Period 3 was from July 8 to August 5, and Period 4 was from August 6 to September 4. These periods were selected as they roughly corresponded to the major periods of the eastern North Carolina agricultural season, with the major activities being tobacco and sweet potatoes being planted in Period 1; cucumbers being harvested, tobacco being topped, and sweet potatoes being planted in Period 2; tobacco being topped and harvested in Period 3; and tobacco being harvested and cured in Period 4.

Two outcome measures are the detection and concentration for the six DAP urinary metabolites of OP pesticides: DMP, DMTP, DMDTP, DEP, DETP, and DEDTP. Detection indicates if any of the metabolites was found in a urine sample. Concentration is the amount of the metabolite measured in $\mu\text{g/L}$.

Three sets of measures are used to describe the participants and their environments. The first set includes the individual characteristics sex; age in the categories 18–24 years, 25–29 years, 30–39 years, and 40 years and older; educational attainment in the categories 0–6 years, 7–9 years, and 10 or more years; country of birth with the values United States, Puerto Rico, Mexico, and other; country of residence with the values United States, Puerto Rico, Mexico, and other; the three dichotomous measures of language speaks English, speaks Spanish, and speaks indigenous (American Indian) language; seasons in US agriculture in the categories, 1 year or less, 2–3 years, 4–7 years, and 8 or more years; worker type in the categories migrant worker or seasonal worker; and H2A visa status. The values for these measures did not change during the agricultural season.

The second set of measures describes crops in which participants worked for at least 1 of the 3 days before the data collection for each of the four periods. Farmworkers are coded as to whether they worked at least 1 of the 3 days preceding the data collection, and whether or not they worked in tobacco, sweet potatoes, cucumbers, blueberries, strawberries, cabbage, sod, other vegetables (beans, green beans, chillies, corn, squash, tomatoes, potatoes, watermelon, melons), or other crops (cotton, peanuts, flowers, hay, soybeans, wheat). The 3 day look-back period corresponds with the period in which most OP pesticides are metabolized.

The third set of measures focuses on pesticide safety. Participants indicated if they had never received pesticide safety training, if they had not received training in the current year, and if they received training in the current year, whether they understood the pesticide safety training (some or none, most, all). Participants indicated if pesticides had

been applied in their room or in the camp in the week before the interview. They indicated the number of days in the previous 3 days (0, 1, 2, 3) that they had mixed, loaded or applied pesticides, worked in fields in which pesticides had been applied in last 7 days, or worked next to fields in which pesticides were being applied.

Statistical Analysis

Descriptive statistics, counts and percentages, were used to describe farmworker characteristics for the season as a whole (287 farmworkers in the sample) and for each time period (per the number of farmworkers interviewed in the respective period). Pesticide metabolites were described with counts and frequencies for detection and percentiles for concentrations. Concentrations were not adjusted for creatinine. A large number of values were below the LOD for each metabolite. To determine percentile values below the LOD, we used a robust regression on order statistics (ROS) approach to obtain percentile estimates for each metabolite [Helsel, 2005]. The ROS assumes that values below the LOD follow a lognormal distribution. A regression equation based on the normal scores of the values above the LOD was used to predict the censored observations. Finally, the estimated non-detected values were combined with the detected values to compute summary statistics as if no censoring had occurred. The robust ROS approach is different from a simple substitution method in the sense that only the corporate collection of estimates below the LOD is used to compute the percentiles. The 95% confidence intervals for the medians of the metabolites at each time period (except for DEDTP due to the lack of sufficient number of detects) were obtained using a bootstrapping method [Efron and Tibshirani, 1993].

RESULTS

The majority (91.3%) of farmworkers in this study are men (Table I). They have a mean age of 33.7 years (SE 0.82); 41.2% are under age 30, 32.1% are in their 30s, and 26.8% are 40 years of age or older. Most have little education, with 51.9% having 6 or fewer years, and 33.8% having seven to nine years. Most were born in Mexico (94.8%) and Mexico is their country of residence (85.0%). All farmworkers speak Spanish, with 11.5% speaking English. It is noteworthy that 23.3% of farmworkers speak an indigenous language.

Although 16.7% of farmworkers have worked in US agriculture for one season or less, 37.3% have worked in US agriculture for eight or more seasons, and 28.6% have worked for four to seven seasons. Most consider themselves to be migrant workers (88.8%), with 52.3% having H2A visas. The great majority (90.6%) live in grower provided housing. About equal percentages (40%) live in houses or trailers, and 20% live in a barrack.

TABLE I. Personal Characteristics of Farmworkers, Eastern North Carolina, 2007 (N = 287)

| Personal characteristics | n | % |
|----------------------------|-----|------|
| Sex | | |
| Male | 262 | 91.3 |
| Female | 25 | 8.7 |
| Age | | |
| 18–24 years | 63 | 22.0 |
| 25–29 years | 55 | 19.2 |
| 30–39 years | 92 | 32.1 |
| 40 or more years | 77 | 26.8 |
| Educational attainment | | |
| 0–6 years | 149 | 51.9 |
| 7–9 years | 97 | 33.8 |
| 10 or more years | 41 | 14.3 |
| Country of birth | | |
| Mexico | 272 | 94.8 |
| US | 3 | 1.0 |
| Other | 12 | 4.2 |
| Country of residence | | |
| Mexico | 244 | 85.0 |
| US | 32 | 11.1 |
| Other | 11 | 3.8 |
| Language | | |
| Speaks English | 33 | 11.5 |
| Speaks Spanish | 287 | 100 |
| Speaks indigenous language | 67 | 23.3 |
| Seasons in US agriculture | | |
| 1 year or less | 48 | 16.7 |
| 2–3 years | 49 | 17.1 |
| 4–7 years | 82 | 28.6 |
| 8 or more years | 107 | 37.3 |
| Worker Type | | |
| Migrant worker | 255 | 88.8 |
| Seasonal worker | 32 | 11.1 |
| H2A visa | | |
| No | 137 | 47.7 |
| Yes | 150 | 52.3 |
| Housing | | |
| Grower provided | 260 | 90.6 |
| Other | 27 | 9.4 |
| Housing type | | |
| House | 117 | 40.8 |
| Barracks | 58 | 20.2 |
| Trailer | 112 | 39.0 |

Farmworkers generally worked at least 1 of the 3 days before the data collection (Table II). Most had worked in tobacco, with 32.3% of those interviewed in the first period working in tobacco at least 1 of the 3 days before the data collection, 48.1% in the second period, 74.3% in the third period, and 83.5% in the fourth period. A substantial percent

TABLE II. Farmworkers Who Worked In Specific Crops At Least 1 of the 3 Days Prior to Interview, Eastern North Carolina, 2007 (N = 287)

| Crops | Period | | | | | | | |
|-------------------------------|---------|-------|---------|-------|---------|-------|---------|-------|
| | 5/1–6/8 | | 6/9–7/7 | | 7/8–8/5 | | 8/6–9/4 | |
| | n | % | n | % | n | % | n | % |
| Total farmworkers | 254 | 100.0 | 233 | 100.0 | 230 | 100.0 | 230 | 100.0 |
| Worked 1 or more days | 246 | 96.9 | 226 | 97.0 | 217 | 94.3 | 221 | 96.1 |
| Crops | | | | | | | | |
| Tobacco | 82 | 32.3 | 112 | 48.1 | 171 | 74.3 | 192 | 83.5 |
| Sweet potatoes | 76 | 29.9 | 55 | 23.6 | 17 | 7.4 | 16 | 7.0 |
| Cucumbers | 9 | 3.5 | 29 | 12.4 | 15 | 6.5 | 0 | 0 |
| Blueberries | 30 | 11.8 | 8 | 3.4 | 0 | 0 | 0 | 0 |
| Strawberries | 11 | 4.3 | 5 | 2.1 | 0 | 0 | 0 | 0 |
| Cabbage | 12 | 4.7 | 1 | 0.4 | 0 | 0 | 0 | 0 |
| Sod | 9 | 3.5 | 12 | 5.2 | 9 | 3.9 | 10 | 4.3 |
| Other vegetables ^a | 26 | 10.2 | 10 | 4.3 | 11 | 4.8 | 7 | 3.0 |
| Other ^b | 10 | 3.9 | 5 | 2.1 | 1 | 0.4 | 1 | 0.4 |

^aBeans, green beans, chillies, corn, squash, tomatoes, potatoes, watermelon, melons.

^bCotton, peanuts, flower, hay, soybeans, wheat.

of farmworkers had worked planting sweet potatoes in the first (29.9%) and second (23.6%) periods. In the first period a substantial percentage of farmworkers had worked in blueberries (11.8%) and other vegetables (10.2%), while a substantial percentage had worked in cucumbers in the second period (12.4%).

About one in five farmworkers have never received pesticide safety training (Table III). Approximately 80% of the farmworkers across the four periods stated that they had received pesticide safety training this year. However, about one-quarter of all participants indicated they understood some or none of the training, fewer than one-in-four indicated they understood most of their pesticide safety training, and only about one-quarter stated that they understood all of the pesticide safety training they received. From 10 to 20% across the periods indicated that pesticides had been applied in the room in which they slept in the week before the interview. About one-quarter indicated that pesticides had been applied in their camp in the week before the interview during the first period, 14.6% in the second, and 5.2% and 6.1% in the third and fourth, respectively. In the first period, 10.2% indicated that they had mixed, loaded or applied pesticides in 1 or more of the 3 days before data collection; this percentage declined to 4.3% and 5.7% in the second and third, and to 0.9% in the fourth, respectively. The percentage of farmworkers who reported having worked in fields to which pesticides had been applied in the previous 7 days for 1 or more of the 3 days before data collection decreased from 33.9% in the first period, to 24.9% in the second period, 20.9% in the third period, and 11.7% in the final period.

Of the 939 urine samples collected from the 287 farmworkers, DMP was detected in 388 (41.3%), DMTP was detected in 735 (78.3%), DMDTP was detected in 313 (33.3%), DEP was detected in 380 (40.5%), DETP was detected in 303 (32.3%), and DEDTP was detected in 76 (8.1%) (Table IV). The 50th percentile concentrations of these metabolites were 0.96 µg/L for DMP, 3.61 µg/L for DMTP, 0.04 µg/L for DMDTP, 0.87 µg/L for DEP, 0.17 µg/L for DETP, and 0.00 µg/L for DEDTP.

The frequency of detection and median concentrations for each of the DAP urinary metabolites tended to increase across the agricultural season, with some decrease in frequency of detection for the last period (Table V, Fig. 1 and Fig. 2). For example, the frequency of detection for DMTP increased from 53.6% in Period 1, to 83.3% in Period 2, and 93.4% in Period 3, and returned to 85.0% in Period 4. The median concentrations for DMTP increased from 1.84 µg/L in Period 1, to 3.44 µg/L for Period 2, to 4.37 µg/L for Period 3, and to 4.98 µg/L for Period 4. The frequency of detection for DEP declined from Period 1 to Period 2, with values of 38.8% and 32.6%, but increased to 38.0% in Period 3, and 52.9% in Period 4. The median concentrations for DEP decreased from 0.96 µg/L in Period 1, to 0.60 µg/L for Period 2, but increased to 0.77 µg/L for Period 3, and to 1.51 µg/L for Period 4.

DISCUSSION

The goals of this article have been to provide an overview of the PACE3 research project, and to present a

TABLE III. Pesticide Exposure Characteristics of Farmworkers, Eastern North Carolina, 2007 (N = 287)

| Exposure characteristic | Period | | | | | | | |
|------------------------------------------------------------------------------------------------|---------|------|---------|------|---------|------|---------|------|
| | 5/1–6/8 | | 6/9–7/7 | | 7/8–8/5 | | 8/6–9/4 | |
| | n | % | n | % | n | % | n | % |
| Pesticide safety training | | | | | | | | |
| Never received training | 65 | 25.6 | 54 | 23.2 | 48 | 20.9 | 41 | 17.8 |
| Did not receive training in current year | 12 | 4.7 | 10 | 4.3 | 9 | 3.9 | 6 | 2.6 |
| Received training in current year and understood some or none | 56 | 22.0 | 65 | 27.9 | 62 | 27.0 | 64 | 27.8 |
| Received training in current year and understood most | 58 | 22.8 | 47 | 20.2 | 50 | 21.7 | 51 | 22.2 |
| Received training in current year and understood all | 63 | 24.8 | 57 | 24.5 | 61 | 26.5 | 68 | 29.6 |
| Pesticides applied in room | | | | | | | | |
| No | 218 | 85.8 | 185 | 79.4 | 206 | 89.6 | 205 | 89.1 |
| Yes | 36 | 14.2 | 48 | 20.6 | 24 | 10.4 | 25 | 10.9 |
| Pesticides applied in camp | | | | | | | | |
| No | 191 | 75.2 | 199 | 85.4 | 218 | 94.8 | 216 | 93.9 |
| Yes | 63 | 24.8 | 34 | 14.6 | 12 | 5.2 | 14 | 6.1 |
| Number of previous 3 days mixed, loaded, or applied pesticides | | | | | | | | |
| 0 day | 228 | 89.8 | 223 | 95.7 | 217 | 94.3 | 228 | 99.1 |
| 1 to 3 days | 26 | 10.2 | 10 | 4.3 | 13 | 5.7 | 2 | 0.9 |
| Number of previous 3 days worked in fields in which pesticides had been applied in last 7 days | | | | | | | | |
| 0 day | 168 | 66.1 | 175 | 75.1 | 182 | 79.1 | 203 | 88.3 |
| 1 day | 16 | 6.3 | 7 | 3.0 | 4 | 1.7 | 3 | 1.3 |
| 2 days | 25 | 9.8 | 22 | 9.4 | 23 | 10.0 | 11 | 4.8 |
| 3 days | 44 | 17.3 | 29 | 12.4 | 21 | 9.1 | 13 | 5.7 |

descriptive analysis of study participant personal characteristics and of one set of pesticide biomarkers, the urinary DAP metabolites, for the study participants across the agricultural season. PACE3 is the first study of pesticide exposure across an agricultural season that includes a large sample of farmworkers. The characteristics of farmworkers vary in different regions of the country; for example, there is a greater percentage of recent immigrants in the Southeast compared to the West Coast. Farmworker populations

experience change in characteristics over time; for example, the number of indigenous farmworkers is increasing in many parts of the county. Descriptions of the characteristics of farmworkers who participate in different studies are necessary to understand comparability of study results. The personal characteristics of the 287 farmworkers who participated in this study can be compared to the personal characteristics documented for the national farmworker population [Carroll et al., 2005], and to those for farm-

TABLE IV. Limit of Detection ($\mu\text{g/L}$), Frequency of Detection and Urinary DAP Concentrations ($\mu\text{g/L}$) for Farmworkers, Eastern North Carolina, for all Samples, 2007 (N = 939)

| Metabolite | Limit of detection | Concentrations | | | | | | |
|------------|--------------------|------------------------|------|---------|--------------------|------|-------|--------|
| | | Frequency of detection | | Maximum | Concentrations (%) | | | |
| | | n | % | | 25th | 50th | 75th | 95th |
| DMP | 0.6 | 388 | 41.3 | 395.09 | 0.29 | 0.96 | 3.80 | 21.11 |
| DMTP | 0.2 | 735 | 78.3 | 2577.78 | 1.18 | 3.61 | 10.36 | 102.30 |
| DMDTP | 0.1 | 313 | 33.3 | 577.62 | 0.00 | 0.04 | 1.01 | 22.18 |
| DEP | 0.2 | 380 | 40.5 | 127.42 | 0.29 | 0.87 | 2.96 | 11.89 |
| DETP | 0.1 | 303 | 32.3 | 141.97 | 0.02 | 0.17 | 1.25 | 7.00 |
| DEDTP | 0.1 | 76 | 8.1 | 96.25 | 0.00 | 0.00 | 0.00 | 0.81 |

TABLE V. Frequency of Detection and Urinary DAP Concentrations ($\mu\text{g/L}$) for Farmworkers, Eastern North Carolina, for the Four Data Collection Periods, 2007

| Metabolite | Period 1: 5/1–6/8 (n = 250) | | | | | | | | Period 2: 6/9–7/7 (n = 233) | | | | | |
|------------|-----------------------------|------|----------------|------|--------|------|-------|------------------------|-----------------------------|----------------|------|--------|-------|--------|
| | Frequency of detection | | Concentrations | | | | | Frequency of detection | | Concentrations | | | | |
| | | | Maximum | | | | | | | Maximum | | | | |
| | n | % | Maximum | 25th | median | 75th | 95th | n | % | Maximum | 25th | median | 75th | 95th |
| DMP | 92 | 36.8 | 395.09 | 0.18 | 0.69 | 3.20 | 14.37 | 80 | 34.3 | 67.1 | 0.18 | 0.71 | 2.76 | 20.56 |
| DMTP | 134 | 53.6 | 1557.23 | 0.43 | 1.84 | 6.64 | 49.16 | 194 | 83.3 | 836.8 | 1.58 | 3.44 | 10.59 | 108.64 |
| DMDTP | 35 | 14.0 | 286.02 | 0.01 | 0.08 | 0.59 | 12.75 | 66 | 28.3 | 182.9 | 0.02 | 0.17 | 1.10 | 17.82 |
| DEP | 97 | 38.8 | 127.42 | 0.40 | 0.96 | 2.68 | 7.15 | 76 | 32.6 | 37.9 | 0.19 | 0.60 | 2.04 | 10.53 |
| DETP | 42 | 16.8 | 141.97 | 0.03 | 0.13 | 0.65 | 7.45 | 61 | 28.2 | 52.1 | 0.10 | 0.34 | 1.01 | 5.58 |
| DEDTP | 6 | 2.4 | 96.25 | 0.00 | 0.00 | 0.00 | 0.42 | 23 | 9.9 | 11.7 | 0.00 | 0.01 | 0.05 | 1.03 |

| Metabolite | Period 3: 7/8–8/5 (n = 229) | | | | | | | | Period 4: 8/6–9/4 (n = 227) | | | | | |
|------------|-----------------------------|------|----------------|------|--------|-------|--------|------------------------|-----------------------------|----------------|------|--------|-------|--------|
| | Frequency of detection | | Concentrations | | | | | Frequency of detection | | Concentrations | | | | |
| | | | Maximum | | | | | | | Maximum | | | | |
| | n | % | Maximum | 25th | median | 75th | 95th | n | % | Maximum | 25th | median | 75th | 95th |
| DMP | 99 | 43.3 | 134.98 | 0.28 | 0.89 | 3.75 | 18.73 | 117 | 51.5 | 139.79 | 0.53 | 1.44 | 5.44 | 27.72 |
| DMTP | 214 | 93.4 | 2577.78 | 1.72 | 4.37 | 10.27 | 138.21 | 193 | 85.0 | 944.87 | 2.12 | 4.98 | 14.34 | 171.76 |
| DMDTP | 106 | 46.3 | 577.62 | 0.04 | 0.25 | 1.54 | 27.61 | 106 | 46.7 | 192.24 | 0.03 | 0.22 | 1.46 | 33.50 |
| DEP | 87 | 38.0 | 45.44 | 0.27 | 0.77 | 3.28 | 12.24 | 120 | 52.9 | 36.01 | 0.65 | 1.51 | 5.00 | 16.28 |
| DETP | 112 | 48.9 | 52.43 | 0.19 | 0.53 | 1.69 | 10.16 | 88 | 38.8 | 101.38 | 0.16 | 0.52 | 1.80 | 7.00 |
| DEDTP | 24 | 10.5 | 48.18 | 0.00 | 0.00 | 0.02 | 0.81 | 23 | 10.1 | 22.82 | 0.00 | 0.01 | 0.05 | 1.43 |

workers who have participated in other recent studies in the southeastern US [Arcury et al., 2007].

Compared to the characteristics of the farmworkers who participated in the 2001-2002 National Agricultural Workers Survey (NAWS) [Carroll et al., 2005], a greater percentage of the PACE3 participants are male (91.3% for PACE vs. 75% for NAWS), of Mexican heritage (94.8% vs. 72%), and migrant (88.8% vs. 42%). Fewer PACE3 participants (11.5%) speak English than do NAWS participants (18%). Only 5% of NAWS participants are indigenous, compared to 23.3% of the PACE3 participants who speak an indigenous language. The NAWS does not include workers with H2A visas, and 52.3% of the PACE3 participants have H2A visas.

The PACE3 participants are similar to the farmworkers in a 2004 survey completed in the same areas of North Carolina [Arcury et al., 2007]. More PACE3 participants are female (8.7%) and older (26.8% aged 40 years and older) than was true of the 2004 survey (1.3% female, 17.1% aged 41 years and older). The PACE3 sample also includes fewer workers with H2A visas (52.3%) than for the 2004 survey (62.8%).

About 75 to 80% of the farmworkers who participated in PACE3 indicated that they had received pesticide safety training. About one-third of those who had received pesticide safety training stated that they did not understand the information that was presented to them. These results are consistent with other studies completed in North Carolina and elsewhere, in which from one-quarter to one-half of farmworkers have not received any pesticide safety training [U.S. GAO, 2000; Arcury et al., 2001; Shipp et al., 2005].

Other studies have not reported information on residential pesticide application (pesticides applied in room, pesticides applied in camps), on whether farmworkers mixed, loaded or applied pesticides, or if they worked in fields to which pesticides had recently been applied. Therefore, our results cannot be compared to other studies. However, these results indicate that the PACE3 participants are exposed to pesticides used residentially, with about 14% having pesticides applied in their sleeping room and about 13% having pesticides applied in their camp in the previous week. Although no more than 10% of participants reported mixing, loading or applying pesticides in any one period, from 12 to 34% report working one or more days

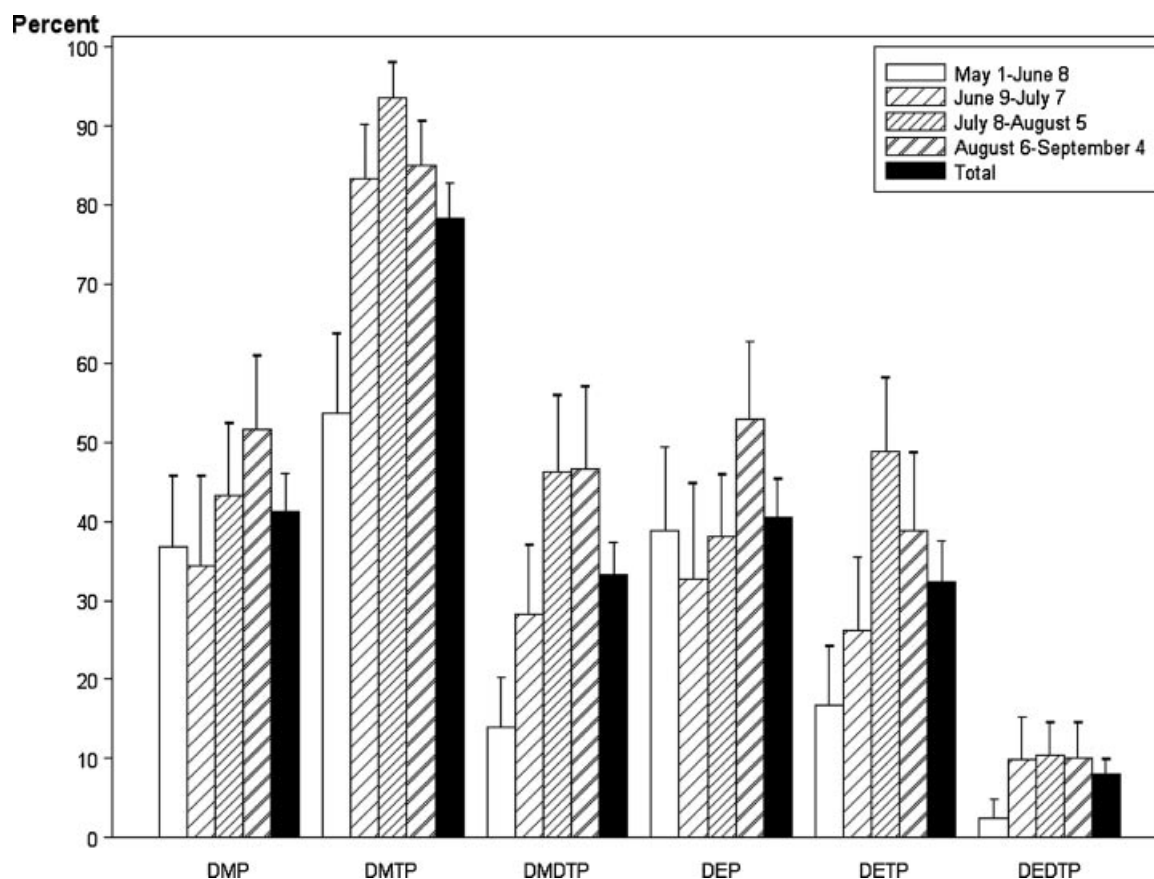


FIGURE 1. Frequency of Detection for DAP Metabolites across the Agricultural Season, Eastern North Carolina Farmworkers, 2007.

in fields to which pesticides had been applied in the previous week.

The frequency of detection and concentrations of the DAP metabolites for the PACE3 participants outline the pattern of exposure across an agriculture season. Almost all participants have at least one metabolite in every urine sample. Frequency of detection and concentrations tended to increase during the season. The crops in which the farmworkers participating in PACE3 worked changed across the season, with workers becoming more involved in cultivating and harvesting tobacco. In the last period of the season, August 6 to September 4, 83.5% of participants had worked in tobacco in the three days before their urine samples were collected, with 7.0% having worked in sweet potatoes, 4.3% in sod, 3.0% in other vegetables, and 0.4% in other crops. The most widely used insecticide for tobacco is the OP pesticide acephate [Southern and Sorenson, 2008]. However, acephate does not metabolize to the DAPs measured in this study, thus exposure to acephate will not be reflected in our measures. The only other OP insecticide that is applied to tobacco is chlorpyrifos, but its use is very limited.

DMTP is the metabolite with the greatest number of detectable concentrations (78.3% of all urine samples) and

the highest concentrations (median of 3.61 $\mu\text{g/L}$) for the PACE3 participants. DMP and DMDTP are also prevalent, with 41.3 and 33.3% of all samples, respectively, containing these metabolites. OP insecticides used in North Carolina that would metabolize to DMTP and DMP are malathion and phosmet [Bravo et al., 2004]. Malathion and phosmet are not recommended for use with tobacco [Southern and Sorenson, 2008], so the source of these exposures is not clear. DEP (40.7%) and DETP (32.3%) are also prevalent among the urine samples collected from the PACE3 participants. The OP insecticide used in North Carolina that would metabolize to DEP and DETP is chlorpyrifos [Bravo et al., 2004], but the use of chlorpyrifos on tobacco is limited, raising questions about the source of exposure.

The frequency of detection and concentrations of DAP metabolites for the PACE3 participants can be compared to those reported in urine samples collected from agricultural populations in Washington State [Curl et al., 2002; Thompson et al., 2008] and California [Bradman et al., 2005] (Table VI). The participants from Washington State were farmworkers, with about one-third being women, and all of the California participants were women living in an agricultural community. Only 8.7% of PACE3 participants were women. Therefore, any differences between men and

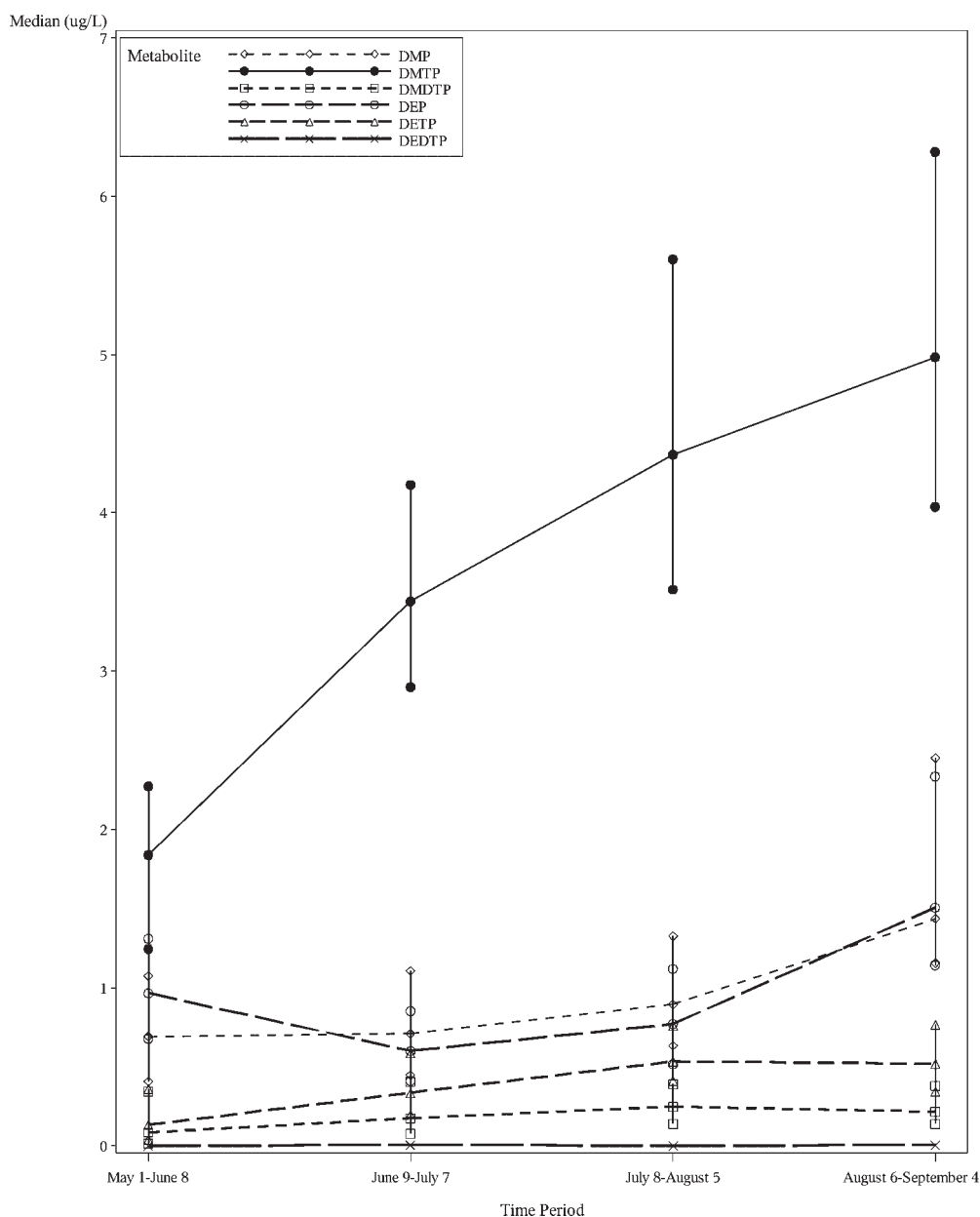


FIGURE 2. Median Concentrations ($\mu\text{g/L}$) for DAP Metabolites across the Agricultural Season, Eastern North Carolina Farmworkers, 2007.

women in pesticide exposure and metabolism affect these comparisons. Variation in frequency of detection and concentrations across studies reflects the pesticides used in the different agricultural systems.

Differences are apparent among the participants in these three studies. Analyses of the Washington State samples provided limited information about the diethyl metabolites. For the dimethyl metabolites, the frequency of detection is greater for DMP, about the same for DMTP, and lower for DMDTP for farmworkers in the North Carolina study compared to those in the Washington State study. Concentrations are greater for DMP and lower for DMTP and

DMDTP for farmworkers in the North Carolina study compared to those in the Washington State study. The frequencies of detection are roughly the same for DMTP, lower for DMP, DMDTP, DEP, DETP, and substantially lower for DEDTP for farmworkers in the North Carolina study compared to women in the California agricultural community study. Concentrations for all the metabolites are lower for farmworkers in the North Carolina study compared to women in the California agricultural community study.

The main strength of PACE3 is that it documents pesticide exposure across an agricultural season. Results

TABLE VI. Comparison of Frequencies of Detection and Median Concentrations for DAP Urinary Metabolites: Washington State 1999 and 2003, California 1999-2000, and North Carolina 2007

| | Washington State ^a | | California 1999-2000 ^c | | | North Carolina 2007 | | | | |
|------------------------|-------------------------------|-----------------|-----------------------------------|------------------------|------------------------|---------------------|---------------------|---------------------|---------------------|---------------------|
| | 1999 N = 205 | 2003 N = 202 | Pre-natal 1 N = 590 | Pre-natal 2 N = 498 | Post-partum N = 489 | Total n = 969 | Period 1 n = 250 | Period 2 n = 233 | Period 3 n = 229 | Period 4 n = 227 |
| Frequency of Detection | | | | | | | | | | |
| DMP | 17.1 | 29.3 | 50.3 | 71.5 | 67.1 | 41.3 | 36.8 | 34.3 | 43.3 | 51.5 |
| DMTP | 93.7 | 92.6 | 65.6 | 97.6 | 85.9 | 78.3 | 53.6 | 83.3 | 93.4 | 85.0 |
| DMDTP | 54.6 | 55.0 | 48.6 | 57.6 | 56.9 | 33.3 | 14.0 | 28.3 | 46.3 | 46.7 |
| DEP | 0.0 ^b | — | 60.4 | 39.8 | 81.4 | 40.5 | 38.8 | 32.6 | 38.0 | 52.9 |
| DETP | 48.0 ^b | — | 49.1 | 98.6 | 65.9 | 32.3 | 16.8 | 28.2 | 48.9 | 38.8 |
| DEDTP | — | — | 45.6 | 12.3 | 29.2 | 8.1 | 2.4 | 9.9 | 10.5 | 10.1 |
| Median Concentration | | | | | | | | | | |
| | µg/L | | µg/L ^d | | | µg/L | | | | |
| DMP | <LOD | <LOD | 0.84 | 1.51 | 3.69 | 0.96 | 0.69 | 0.71 | 0.89 | 1.44 |
| DMTP | 9.7 | 56.2 | 4.10 | 6.02 | 10.03 | 3.61 | 1.84 | 3.44 | 4.37 | 4.98 |
| DMDTP | 1.2 | 6.0 | 0.71 | 1.04 | 0.63 | 0.04 | 0.08 | 0.17 | 0.25 | 0.22 |
| DEP | | | 0.82 | 0.14 | 2.70 | 0.87 | 0.96 | 0.60 | 0.77 | 1.51 |
| DETP | | | 0.42 | 2.13 | 0.59 | 0.17 | 0.13 | 0.34 | 0.53 | 0.52 |
| DEDTP | | | 0.20 | 0.74 | 0.74 | 0.00 | 0.00 | 0.01 | 0.00 | 0.01 |

^a[Thompson et al. 2008].^b[Curl et al. 2002].^c[Bradman et al. 2005].^dConverted from nmol/L.

show considerable variation in frequency of detection and concentrations across the season and this variation is consistent with other studies [Bradman et al., 2005; Thompson et al., 2008]. OP pesticides are generally metabolized and the metabolites are excreted within a few days. Farmworker exposure to OP pesticides depends on task performed and crop on any given day. Pesticide application on a crop varies across the season. Most of the farmworkers in this study cultivated and harvested tobacco; OP insecticides are more likely to be applied to tobacco early in the season, while growth regulators, which are not OP insecticides, are more likely to be applied late in the season. Any single measure of urinary metabolites cannot be considered a dependable indicator of exposure for an individual. Looking across measures for multiple individuals provides an indication of the level of pesticide exposure in the population of farmworkers. Urinary metabolite exposure measures collected at a single time in an agricultural season are also not a good indicator of population pesticide exposure. Seasonal variability indicates that samples need to be collected systematically across the agricultural season to get an indication of pesticide exposure in the population of farmworkers.

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 PACE3 sample was limited to the camps known to each community partner organization, and participants were limited to those living in the camps at the time of recruitment. PACE3 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, the number of detects, and their concentrations were limited to the capabilities of the existing laboratory procedures; for example, DAP metabolites do not include acephate, which is a widely used insecticide in North Carolina. In addition, DAPs can also be derived from exposure to the preformed metabolites in the environment or from OP pesticide exposures in non-occupational or non-residential pathways.

Nevertheless, PACE has several strengths. PACE3 used a longitudinal design to collect pesticide exposure and pesticide biomarker data at 1 month intervals for a large sample of Latino farmworkers in North Carolina. The characteristics of these farmworkers are comparable to those workers participating in the NAWS, with some notable differences. These differences include a greater percentage of PACE3 participants compared to NAWS participants

being from Mexico, being male, being migrant workers, speaking an indigenous language, and having an H2A visa. The characteristics of the PACE3 participants are similar to the characteristics of participants of earlier studies conducted in eastern North Carolina.

Frequency of detection and concentrations of DAP urinary metabolites of OP insecticides indicate that almost all PACE3 participants had at least one metabolite in every urine sample, and that the frequency of detection and concentrations tended to increase during the season. Direct comparisons of the frequency of detection and concentrations for the PACE3 participants to other farmworker studies are difficult due to the few studies that have been completed, and differences in the compositions of the study populations. However, PACE3 participants had comparable frequency of detection and concentrations with the participants in these other studies. The variability in the frequency of detection and concentrations of the DAP urinary metabolites across the agricultural season indicates the importance of a longitudinal design for studies of pesticide biomarkers in farmworker populations.

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