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Longitudinal Assessment of Blood Cholinesterase Activities over Two Consecutive Years among Latino Non-farmworkers and Pesticide-Exposed Farmworkers in North Carolina

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Abstract

Objective—This study (1) describes patterns of whole blood total cholinesterase, acetylcholinesterase, and butyrylcholinesterase activities across the agricultural season, comparing farmworkers and non-farmworkers; and (2) explores differences between farmworkers' and non-farmworkers' likelihood of cholinesterase depression.

Methods—Blood samples from 210 Latino male farmworkers and 163 Latino workers with no occupational pesticide exposure collected eight times across two agricultural seasons were analyzed. Mean cholinesterase activity levels and depressions 15% were compared by month.

Results—Farmworkers had significantly lower total cholinesterase and butyrylcholinesterase activities in July and August and lower acetylcholinesterase activity in August. Farmworkers had significantly greater likelihood of cholinesterase depression for each cholinesterase measure across the agricultural season.

Significance—A repeated-measures design across two years with a non-exposed control group demonstrated anticholinesterase effects in farmworkers. Current regulations designed to prevent pesticide exposure are not effective.

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COMPETING INTERESTS

The authors declare that they have no competing financial interests.

INTRODUCTION

Exposures to cholinesterase-inhibiting pesticides, including organophosphorus and carbamate pesticides, place farmworkers at risk for immediate neurotoxic effects and may be linked to delayed effects, including neurodegenerative diseases and effects on children exposed in utero.¹ While pesticide handlers are at the greatest risk for exposure and immediate health effects, field workers who do not routinely mix and apply pesticides are also at risk for exposure through drift and exposure to pesticide residues.²

Worker education as mandated by the U.S. Environmental Protection Agency Worker Protection Standard (WPS) is designed to reduce pesticide exposures. Increasing use of pyrethroid and other pesticides that do not inhibit cholinesterase activities has been promoted. Despite these measures, studies of pesticide metabolites in farmworkers in the US suggest that a significant number of workers are still exposed to cholinesterase-inhibiting pesticides.²⁻⁴ In addition to work-related exposure, most farmworkers live in substandard housing located near fields.⁵⁻⁷ Such housing has been found to contain residues of multiple pesticides that can further expose workers.⁸ These residues likely reflect drift or take-home pesticide pathways as workers bring pesticides into their residences, as well as the application of pesticides to try to control pest infestations.⁹

Monitoring cholinesterase activities of farmworkers can provide information on their exposure to organophosphorus and carbamate pesticides.^{10,11} Such pesticides can reduce the activity of the two cholinesterases, acetylcholinesterase (Enzyme Commission number 3.1.1.7) and butyrylcholinesterase (Enzyme Commission number 3.1.1.8) in tissues throughout the body. Blood provides a readily available tissue source of cholinesterases that can be used for monitoring exposures to anticholinesterases.¹² In red blood cells, acetylcholinesterase inhibited by organophosphorus pesticides is slowly reversible so reduced red blood cell activity can be quite prolonged, reflecting the cell's long circulation time and limited protein synthesis machinery. Binding to plasma butyrylcholinesterase by the same inhibitor generally leads to more rapid recovery due to synthesis of new butyrylcholinesterase molecules in the liver and secretion into the circulation. Similarly, carbamate pesticides covalently bind cholinesterases, but generally lead to more rapidly reversible inhibition than with the organophosphorus pesticides. These and other issues, such as the wide range of normal values in levels of cholinesterases,¹³ provide a rationale for repeated measures across time in assessing depression in cholinesterase activities.

Currently, monitoring of cholinesterase activities is widely recommended for workers who mix, load, and apply pesticides. It is mandated in only a few states, such as Washington and California.^{10,14} There is no requirement to monitor agricultural workers who are not applicators.

We previously analyzed total cholinesterase activities obtained from dried whole blood samples in non-applicator farmworkers in eastern North Carolina. We showed that cholinesterase activity was significantly lower later in the summer and that depressions in cholinesterase activity were related to the number of different organophosphorus and carbamate pesticide metabolites detected in urine.¹⁵ That study had several shortcomings,

including cholinesterase data from just a single year; lack of a comparison group; no information on potential residential pesticide exposure; and cholinesterase obtained from dried whole blood samples, preventing differentiation of acetylcholinesterase and butyrylcholinesterase activities. The current study was designed to remedy these shortcomings to more definitively assess evidence of work-related exposure to cholinesterase-inhibiting pesticides.

In this article we focus on data collected from Latino farmworkers in eastern North Carolina and a comparison group of Latino non-farmworkers in occupations unlikely to expose them to pesticides. Blood samples were obtained through venipuncture across summers of 2012 and 2013, and self-reports of residential exposure sources were also obtained. Our objectives in this study were to (1) describe patterns of whole blood total cholinesterase, acetylcholinesterase, and butyrylcholinesterase activities across the agricultural season, comparing farmworkers and non-farmworkers; and (2) explore the differences between farmworkers' and non-farmworkers' likelihood of cholinesterase depression across the agricultural season, taking into account self-reported residential pesticide exposure.

METHODS

Analyses reported here are based on data collected as part of an ongoing community-based participatory research program, "CBPR on Pesticide Exposure & Neurological Outcomes for Latinos: PACE4". PACE4 used a prospective two-group design in which data were collected at a baseline interview and 8 subsequent contacts with interview and blood sample collection across summers 2012 and 2013. The two groups were farmworkers, assumed to be exposed to pesticides in the workplace, and non-farmworkers, restricted to those in occupations with no major occupational pesticide exposure. All study procedures were approved by the Wake Forest Health Sciences Institutional Review Board. Study participants gave signed, informed consent before taking part in the study.

Locale

Farmworkers were recruited in east central North Carolina, the location one of the project's community partners, the North Carolina Farmworkers Project. Non-farmworkers were recruited from Forsyth County in west central North Carolina. El Buen Pastor Latino Community Services was the Forsyth County community partner. Agriculture is practiced extensively in the east central region.

Sample

PACE4 participants were men aged 30 years and older. All self-identified as Latino or Hispanic; most spoke Spanish as their primary language. Farmworkers were currently employed in agriculture and had worked in agriculture for at least three years. Non-farmworkers could not have been employed for the past three years in jobs that expose workers to pesticides, including agriculture, forestry, landscaping, grounds keeping, lawn maintenance, and pest control.

Community partners assisted with recruitment. NC Farmworkers Project staff approached farmworker camps they served, and explained the project to camp residents, including the

inclusion and exclusion criteria, time commitments, and incentives. Volunteers were screened to ensure that they met the inclusion criteria and signed informed consent was obtained. Winston-Salem staff worked with El Buen Pastor Latino Community Services and other community organizations to identify potential participants. Potential participants were then contacted by project staff who explained the project; volunteers were screened to ensure that they met the inclusion criteria and signed informed consent was obtained.

A total of 235 farmworkers and 212 non-farmworkers completed the baseline interviews (beginning of 2012), with 210 farmworkers and 163 non-farmworkers completing Contact 1 at which the first blood samples were collected; 138 farmworkers and 117 non-farmworkers completed Contact 4 (end of 2012). Among farmworkers, 108 completed Contact 5 (beginning of 2013), and 98 completed Contact 8 (end of 2013). Among non-farmworkers, 68 completed Contact 5, and 72 completed Contact 8. The number of participants who provided blood samples for cholinesterase analyses was somewhat smaller than the number who completed each contact.

Groups of farmworkers were asked to volunteer, only the number who agreed to volunteer is available (the denominator is unknown); generally, all of the farmworkers in a camp who met the inclusion criteria volunteered. However, individual farmworkers who did not want to participate could have avoided contact with the project staff or indicated that they did not meet the inclusion criteria to avoid refusal. Among the non-farmworkers, 101 individuals were contacted who did not meet the inclusion criteria. Of those contacted and meeting the inclusion criteria, 87 individuals refused to participate for a participation rate of 70.9% ($212/(87+212)$). Reasons given for refusing included the time commitment and length of the study (51), blood draws (27), need to come to a clinic for data collection (31), and providing contact information (30) (individuals could give more than one reason for refusing).

Data collection

Data collection relevant to these analyses included interviews, all conducted face-to-face, and venous blood draws. Participants received a \$30 cash incentive for answering the baseline questionnaire and after each of the clinic visits at Contacts 2, 5, 6 and 9; they received a \$20 cash incentive after each of the intermediary visits at Contacts 3, 4, 7 and 8. Participants who completed all nine contacts received a total of \$230.

Questionnaires included demographic characteristics (at baseline only) such as age, educational attainment, language preference, work and occupation characteristics. At every contact after baseline, participants were asked if, during the past week: (1) pesticides had been applied in their residence; (2) pesticides had been applied in their yard; and (3) pesticides had been applied to agricultural fields within one-half mile of their residence.

Blood samples were drawn into 4ml green top heparin-containing tubes by a trained phlebotomist using venipuncture. Tubes were inverted 8–10 times to ensure the anticoagulant was thoroughly mixed into the sample. At farmworker camps, tubes were temporarily stored in a cooler on ice packs for transportation to the field office where they were frozen at -20°C . Non-farmworkers had their blood drawn in a clinic setting where

samples were refrigerated up to six hours during the clinic lab collection before being frozen -20°C . At the end of each summer, all blood samples were moved to storage at -80°C .

Measures

We defined a depression of cholinesterase as a change $\geq 15\%$ from an individual's yearly maximum value, assuming that a participant's maximum value was the best indicator of cholinesterase recovery in the absence of baseline cholinesterase values. We used 15% reduction previously,¹⁵ after exploring a range of possible cut points and considering Washington State's use of 20% reduction to trigger inquiries to address possible pesticide exposure and Lefkowitz et al.,¹³ who observed that 12.1% differences in red blood cell cholinesterase activity have a 95% probability of being significant departures from baseline.

Data collection time was divided into four periods roughly corresponding to months. June included data collected from 1 June to 7 July; July, from 8 July to 7 August; August, from 8 August to 7 September; and September, from 8 September to 21 October.

Recent residential pesticide exposure was calculated from three questions regarding recent home, yard, and neighborhood pesticide application in the previous week. Responses were summed, and values could range from 0 to 3 at each time point. These were collapsed into three categories of 0, 1, and 2 or more exposure sources.

Laboratory analysis

Blood samples were shipped on dry ice to Oklahoma State University. Total cholinesterase, acetylcholinesterase and butyrylcholinesterase activities were assayed with a modification of the radiometric method of Johnson and Russell using differential inhibition conditions.¹⁶ To avoid possible reactivation of cholinesterases by "reversible" inhibitors, whole blood samples were thawed on ice and assayed individually as rapidly as possible after thawing, using a minimal tissue dilution (1:5 in the final reaction).^{17,18} Each sample was assayed under two conditions, with duplicate reactions for each. Immediately after thawing, 20 μl aliquots of whole blood were added to four separate 7-ml glass scintillation vials. In two of the vials, 60 μl of 50 mM potassium phosphate buffer, pH 7 (phosphate buffer) was added while in the other two, 60 μl of the specific acetylcholinesterase inhibitor BW 284C51¹⁹ in phosphate buffer was added (final concentration 0.5 μM ; this concentration completely inhibited acetylcholinesterase activity under the assay conditions). Samples were vortexed, and then placed into a water bath at 26°C for 60 seconds before adding 20 μl of substrate ($[^3\text{H}]$ acetylcholine iodide, 1 mM final concentration) in phosphate buffer. The reactions were allowed to incubate a further 30 seconds after substrate addition and then terminated by addition of 100 μl of acidified buffer (pH 2.5). Five ml of an organic scintillation cocktail was then added. Vials were vortexed and counted the following day to allow equilibration of the aqueous and organic phases. A time-course of hydrolysis was conducted to measure total hydrolysis of the substrate, using the nonlinear fit, one phase association program in GraphPad Prism version 5 to estimate Y_{max} . Activity in unknown samples was then referenced to Y_{max} (with cpm values of Y_{max} representing the 100 nmol acetylcholine in each reaction). Total cholinesterase activity was defined as the amount of activity in the samples without BW 284C51, butyrylcholinesterase activity was defined as the amount of

activity in the samples incubated with BW 284C51, and acetylcholinesterase activity was defined as the difference in activity between those two conditions.

Data analysis

We summarized personal characteristics for farmworkers and non-farmworkers using frequency counts and percentages for categorical variables and means and standard deviations for age. To evaluate the differences in total cholinesterase, acetylcholinesterase, and butyrylcholinesterase activities between farmworkers and non-farmworkers, we used a linear mixed effects model approach to account for the repeated measures across the four different months in a two year period. Because the primary interest was the monthly difference by farmworker status, the basic models included year (2012 and 2013), month, farmworker status, and month x farmworker interaction. We further expanded the basic models and added recent residential pesticide exposure as a time-varying covariate to reflect contemporaneous residential exposure. Least square means and 95% confidence intervals were presented in figures. Similar models were fit for depression in cholinesterase activities. We used a generalized estimating equation (GEE) approach to account for the repeated measures within the same participant. Observations from participants with only one measure within a given year were excluded from this portion of the analyses. Odds ratios and 95% confidence intervals were presented for each month as well as the overall duration of the study. All analyses were performed using SAS 9.4 (Cary, NC). A p-value of less than 0.05 was considered statistically significant.

RESULTS

These analyses include 372 of 373 individuals who participated in Contact 1. The remaining individual was not included because he refused to give a blood sample at any of the contacts.

Workers ranged in age from 30 to 70 years (Table 1). The mean ($\pm$ SD) age for farmworkers was 39.4 ($\pm$ 7.8) years, compared to 41.3 ($\pm$ 8.9) years for non-farmworkers. Farmworkers had lower educational attainment than non-farmworkers; only 8.6% of farmworkers had 12 or more years of education, compared to 32.3% of non-farmworkers. All farmworkers were born in Mexico, while over a third of non-farmworkers were born elsewhere. Most of both worker groups spoke Spanish as their preferred language. Almost all farmworkers (95.2%) were guest workers in the U.S. on H2-A visas, compared to none of the nonfarmworkers. Due to the study design, all farmworkers were employed in agriculture. Nonfarmworkers were distributed across a variety of industries with the largest proportion employed in construction (44.8%) and production (18.5%). Across all months of data collection, farmworkers reported more recent residential pesticide exposure than did non-farmworkers (Table 2).

Mean ($\pm$ SE) levels of total, acetylcholinesterase, and butyrylcholinesterase activities for farmworkers and non-farmworkers by month are shown in Figure 1a–c; estimates are adjusted for year (2012 and 2013). Compared to non-farmworkers, farmworkers had significantly lower total cholinesterase and butyrylcholinesterase activities in July and August, and significantly lower acetylcholinesterase activity in August. Within farmworkers, all months were significantly different from each other for

butyrylcholinesterase activity. June was different from each other month for total cholinesterase activity, and August and September were different from each other. For acetylcholinesterase activity, June was different from each other month. Within non farmworkers, significant differences in total and acetylcholinesterase activities were observed between July and August and between August and September. No significant between-month differences were observed within non-farmworkers for butyrylcholinesterase activity. Models adjusted for recent residential pesticide exposure had similar results (data not shown).

Table 3 presents comparisons between farmworkers and non-farmworkers of likelihood of depressions of 15% or more from an individual's maximum cholinesterase activity, by month. For total cholinesterase, farmworkers had almost four-fold greater odds of depressed cholinesterase activity in August and one and a half times greater odds overall, compared to nonfarmworkers. The association was attenuated, but still significant for August when the model was adjusted for recent residential pesticide exposure. For acetylcholinesterase, the pattern is the same, although differences between farmworkers and non-farmworkers reached significance for September in the unadjusted model. For butyrylcholinesterase, farmworkers had two-fold and three-fold greater odds of depressed cholinesterase in July and August, respectively, and more than one and a half times greater odds overall. These differences remained significant when adjusted for recent residential pesticide exposure. Odds ratios were highest (3.13–3.8) in the month of August for each cholinesterase activity without considering recent residential exposures, and remained highest in August (2.8–3.33) when residential exposures were included in the model.

DISCUSSION

We showed previously that non-applicator farmworkers in eastern North Carolina are exposed to a wide range of pesticides, including organophosphorus and carbamate pesticides, and that many workers show seasonal patterns of repeated exposure across the summer.^{2,20} The present study found mean cholinesterase activities in farmworkers that appear to reflect pesticide usage and exposure, with the lowest activities during the months of most intensive agricultural activity. The pattern differs somewhat from our previous findings,¹⁵ where lowest mean cholinesterase activity was earlier in the summer. This difference may be attributable to differences in cholinesterase activity measurement and differences in study participants. We previously obtained a single measure of cholinesterase activity from dried whole blood spots. Blood spot sample processing required considerable dilution and high temperatures that may have facilitated spontaneous reactivation of carbamylated cholinesterase prior to analyses,¹⁵ resulting in measurement only of organophosphorus pesticide-inhibited cholinesterase activity. The current study is based on the collection of venous whole blood samples. Current study farmworkers were almost exclusively H-2A visa holders who arrived as guest workers from Mexico in late spring. All were men; they worked most of the summer in tobacco. The prior sample was more heterogeneous, including a small group of women and farmworkers residing in North Carolina year round; they likely worked in a broader range of crops to which a different set of pesticides may have been applied.

Data from a comparison group helps to support the conclusion that the patterns of cholinesterase activities among farmworkers found in the present study reflect occupational exposures. Farmworkers' pattern of decreasing mean cholinesterase activities through the summer reached significance for August. Between-group comparisons of cholinesterase depressions found significantly greater likelihood of depressed cholinesterase activities among farmworkers than non-farmworkers. These differences were attenuated somewhat when models were adjusted for self-reported recent residential pesticide exposure, but August differences in each cholinesterase activity remained significant as did July and overall activity for butyrylcholinesterase. The depression of butyrylcholinesterase activity earlier in the season than acetylcholinesterase activity is similar to the pattern seen by Crane et al.²¹ in a study of adolescent pesticide applicators in Egypt where blood sample collection was timed to known organophosphorus pesticide application. This suggests there could be some butyrylcholinesterase-preferential inhibitors used early in the agricultural season. Older human literature and more recent modeling studies suggest that specific organophosphorus pesticides inhibit blood butyrylcholinesterase more effectively than acetylcholinesterase,^{22,23} and that inhibition of butyrylcholinesterase precedes that of acetylcholinesterase with repeated low level exposures.^{24,25} Thus, earlier application of organophosphorus pesticides more potent towards butyrylcholinesterase as well as an earlier onset of butyrylcholinesterase inhibition with repeated exposures could account for earlier significant changes in butyrylcholinesterase activity noted between farmworkers and non-farmworkers.

Only a few studies of cholinesterase activities in agricultural workers have included a control group; these studies are mostly conducted outside the US and contain limited description of the inclusion criteria for controls.^{21,26} Choosing a suitable comparison group for Latino farmworkers in the US is difficult. We attempted to find a comparison group that was unlikely to experience exposure by excluding workers in such industries as landscaping and by recruiting controls in a more urban area. Data were collected from these study participants on lifetime and current exposure using a published instrument.²⁷ The results establish that our intended differences in pesticide exposure between the groups are reflected in their reports of work and living environments.²⁸ Genetic variability may affect cholinesterase activities,²⁹ as well as pesticide metabolism.³⁰ While measuring such variability was beyond the scope of this study, we included only individuals of Hispanic heritage from North, Central, or South America to try to reduce this unmeasured variability. The non-farmworker group was more diverse in terms of country of origin. Recruiting controls from an urban non-agricultural area was an important consideration, as other research has shown that non-farmworker controls recruited from the same rural villages as workers experience longitudinal patterns of cholinesterase activities similar to farmworkers, likely due to endemic pesticide exposure in the rural environment.²¹

Previous research by ourselves and others has shown that pesticide usage to control residential pests is high in the poor quality housing inhabited by farmworkers, both grower-provided housing and that obtained in local housing markets.^{8,31,32} Although we did not measure pesticides in this housing, we used items that tapped both household and neighborhood exposure possibilities in the week prior to each blood sample collection. As expected, farmworkers, many of whom live adjacent to pesticide-treated fields, reported

more potential exposure sources. Taking this into account attenuated some between-group differences in likelihood of cholinesterase depression, but some remained significant.

Farmworkers' risk of pesticide exposure is widely recognized, and the current WPS was published in 1992 to put in place measures to protect farmworkers and pesticide applicators. The WPS mandates training of farmworkers so they understand pesticides they may encounter at work, the health risks pesticides present, and how to protect themselves from pesticide exposure through personal hygiene and use of personal protective equipment which, for most field workers, consists of clean work clothes that cover extremities. The WPS also obligates growers to train workers, post information when pesticides are used, provide workers with access to field sanitation supplies (water, soap, and towels), and take steps to keep workers out of areas being treated with pesticides. The results of this study indicate that, despite the measures mandated by the WPS and some transition to non-anticholinesterase pesticides like pyrethroids, non-applicator farmworkers are still being exposed to pesticides. This is corroborated by studies in farmworker populations throughout the US documenting pesticide exposure through biomarkers,^{3,9,20} as well as studies that show that WPS training is not always provided to workers, that growers are often not in compliance with laundry and bathing facilities and provision of personal protective equipment for workers to be able to practice WPS-recommended protective measures, and that farmworkers' need to work leads them to accept such circumstances.³³⁻³⁷

Study results should be interpreted in light of several limitations. First, these findings come from farmworkers in one area of the US. The pesticides used may be different from those in other parts of the country where farmworkers work in different crops. Second, the lack of baseline values meant that cholinesterase activity reductions had to be calculated from the highest available measure, so some misclassification of cholinesterase activity reductions may have occurred. Third, the use of 15% as the threshold for classifying cholinesterase activity reduction may have resulted in some false positives, though 15% is a generally accepted value.^{38,39} Fourth, no data, including pesticide application records or biomarker assessment, were available to confirm pesticide exposure.

Nonetheless, this study has a number of strengths. First, quantification of cholinesterase activities used a method more likely than the Ellman method⁴⁰ to minimize reactivation of carbamate-inhibited cholinesterases. The Ellman method is, by far, the most common approach used to evaluate cholinesterase activity and inhibition. It is suitable for higher throughput approaches, and uses a simple photometric detector without use of a radiochemical and associated wastes. For the present study, however, we used a radiometric method to measure cholinesterase activities for several reasons. First, radiometric methods in general are highly sensitive. Second, the Ellman method typically performs poorly when samples (e.g., whole blood) contain high levels of hemoglobin or other interfering chromophores. Third, the Ellman method typically requires extensive dilution of the tissues for optimal measurements. This could be important with some cholinesterase-inhibiting pesticides (carbamates) that favor reactivation with extensive dilution. Both the low dilution of the blood samples in the reaction (1:5) and the immediate assay of the samples after thawing minimized the potential for reactivation of carbamate-inhibited cholinesterases.

Beyond laboratory analyses, the primary strength of this study is its design, which included repeated measures of cholinesterase activities over two growing seasons and inclusion of a control group residing in a non-agricultural area. Other studies have used repeated measures. However, measures are sometimes taken at long time intervals and many study participants have only a single measurement,¹¹ or the control group is likely also exposed to endemic agricultural pesticides.²¹

Neurotoxic cholinesterase-inhibiting pesticides continue to be used in agriculture, and farmworkers are exposed. Even low level exposure may place individuals at risk for negative future health consequences. Farmworkers constitute a vulnerable population who, because of language barriers and economic pressure, may not understand their health risks or take steps to protect themselves. This study indicates that steps are needed to ensure farmworkers' workplace safety.

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Figure 1a–c.

Mean ($\pm$ SE) levels of (a) total, (b) acetylcholinesterase, and (c) butyrylcholinesterase activities for farmworkers and non-farmworkers by month. Bars for farmworkers and non-farmworkers are offset slightly for clearer presentation. Estimates are adjusted for year (2012 and 2013).

Table 1

Personal characteristics of non-farmworkers ($n=162$) and farmworkers ($n=210$) in North Carolina, 2012.

	Non-farmworkers n (%)	Farmworkers n (%)
Age (years)		
30–34	44 (27.2)	75 (35.7)
35–44	60 (37.0)	81 (38.6)
45	58 (35.8)	54 (25.7)
Educational attainment (years)		
0–6	56 (35.8)	92 (44.0)
7–11	53 (32.9)	99 (47.4)
12	52 (32.3)	18 (8.6)
Place of birth		
Mexico	106 (65.4)	210 (100)
Central America	40 (24.7)	0
United States, including Puerto Rico	4 (2.5)	0
Other	12 (7.4)	0
Language spoken		
Spanish	161 (99.4)	207 (98.6)
English	0	1 (0.5)
Indigenous language	1 (0.6)	2 (1.0)
H-2A visa		
No	162 (100)	10 (4.8)
Yes	0	200 (95.2)
Work categories		
Agriculture	0	210 (100.0)
Construction	71 (43.8)	0
Customer service	1 (1.0)	0
Food preparation/restaurant work	10 (6.2)	0
Maintenance/cleaning	14 (8.6)	0
Mechanic	9 (5.6)	0
Production	30 (18.5)	0
Professional	3 (1.9)	0
Sales	9 (5.6)	0
Transportation/material moving	7 (4.3)	0
Unemployed	8 (4.9)	0

Table 2

Number of recent residential pesticide exposures^a, comparing non-farmworkers and farmworkers in North Carolina, aggregated over 2012 and 2013.

	Non-farmworkers <i>n</i> (%)	Farmworkers <i>n</i> (%)
June exposures		
0	24 (64.9)	12 (3.6)
1	9 (24.3)	252 (76.0)
2+	4 (10.8)	68 (20.5)
July exposures		
0	120 (79.0)	8 (3.0)
1	26 (17.1)	163 (61.5)
2+	6 (4.0)	94 (35.5)
August exposures		
0	126 (76.8)	10 (4.6)
1	27 (16.5)	162 (74.0)
2+	11 (6.7)	47 (21.5)
September exposures		
0	226 (80.1)	11 (4.8)
1	44 (15.6)	178 (78.1)
2+	12 (4.3)	39 (17.1)

^aExposures defined as pesticides applied within the past week in the residence, in the yard and to agricultural fields within one-half mile of the residence.

Table 3

Comparing farmworker and non-farmworker likelihood of 15% reduction in total cholinesterase, acetylcholinesterase, and butyrylcholinesterase activities during months June to September and overall; unadjusted and adjusted for recent residential pesticide exposure, n=320, North Carolina, 2012 and 2013.

	Model 1 ^a		Model 2 ^b	
	Odds Ratio (95% Confidence Limits)	p-value	Odds Ratio (95% Confidence Limits)	p-value
Total Cholinesterase				
June	0.61 (0.19, 1.99)	0.4093	0.54 (0.16, 1.80)	0.3159
July	1.54 (0.90, 2.64)	0.1131	1.35 (0.73, 2.50)	0.3398
August	3.80 (2.27, 6.38)	<.0001	3.31 (1.82, 6.04)	<.0001
September	1.38 (0.88, 2.17)	0.1620	1.19 (0.71, 2.01)	0.5096
Overall	1.49 (1.04, 2.14)	0.0304	1.30 (0.83, 2.04)	0.2502
Acetylcholinesterase				
June	1.01 (0.38, 2.71)	0.9809	0.92 (0.34, 2.51)	0.8779
July	1.31 (0.81, 2.12)	0.2775	1.20 (0.70, 2.06)	0.5137
August	3.13 (1.94, 5.06)	<.0001	2.80 (1.65, 4.77)	0.0001
September	1.56 (1.05, 2.33)	0.0286	1.38 (0.87, 2.20)	0.1694
Overall	1.60 (1.18, 2.16)	0.0027	1.44 (0.99, 2.10)	0.0576
Butyrylcholinesterase				
June	0.88 (0.35, 2.24)	0.7897	0.93 (0.35, 2.43)	0.8784
July	2.02 (1.31, 3.10)	0.0014	2.18 (1.31, 3.63)	0.0027
August	3.14 (2.00, 4.92)	<.0001	3.33 (2.00, 5.53)	<.0001
September	1.20 (0.83, 1.74)	0.3258	1.27 (0.80, 2.03)	0.3074
Overall	1.61 (1.20, 2.17)	0.0017	1.71 (1.15, 2.54)	0.0078

^aModel included year, month, farmworker status, and month x farmworker interaction.

^bModel included year, month, farmworker status, month x farmworker interaction, and recent residential pesticide exposure.