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# Neurophysiological Function in Farm Workers Exposed to Organophosphate Pesticides

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# Neurophysiological Function in Farm Workers Exposed to Organophosphate Pesticides

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**ABSTRACT. To investigate neurophysiological effects of low-level exposure to foliar organophosphate residues during one season among agricultural workers, the authors performed a cross-sectional study of 67 Hispanic farm workers and 68 age-, gender-, ethnicity-, and education-matched reference subjects. The neurophysiological examination included sensory and motor nerve conduction and neuromuscular junction testing. Erythrocyte cholinesterase activity was measured at the time of examination. No statistically significant neurophysiological differences between the exposed and reference groups were observed. Farm workers and reference subjects had similar sensory nerve latency and amplitude (sural), motor nerve conduction velocity (ulnar), and neuromuscular junction function (ulnar). No relationship between duration of exposure during the season and electrophysiological measures of nerve function was found. Exposure of farm workers to the low levels of organophosphate pesticides during one season experienced by farm workers in this study was not associated with impaired peripheral neurophysiological function.**

THE WELL-DOCUMENTED neurotoxic effects of acute organophosphate (OP) poisoning in humans result largely from the inhibition of the neuroenzyme acetylcholinesterase and the consequent accumulation of acetylcholine at cholinergic transmission sites.[1,2] A syndrome known as organophosphate-induced delayed neuropathy, unrelated to acetylcholinesterase inhibition, also occurs several weeks after acute poisoning by certain OP agents.[1]

Little is known, however, about the neurotoxic effects of persistent low-level exposure to OP pesticides. In the few studies in which such exposure has been examined, investigators have focused on pesticide production workers and pesticide applicators.[3-8] In most of these studies, investigators have reported abnormalities of the motor nerves[3-7] or sensory nerves.[5,6,8] However, there has been little research on the neurophysiological effects of persistent low-level OP exposures experienced by farm workers primarily through foliar residues, rather than from direct application.

The objective of this study was to (a) investigate whether neurophysiological abnormalities occurred in a group of farm workers after a season of low-level OP exposure and (b) estimate associations between observed motor and sensory nerve abnormalities and extent of exposure.

## Method

**Subjects.** We recruited subjects who volunteered for this cross-sectional study from the Wenatchee area

(Douglas and Chelan Counties) in central Washington State during July and August, 1994. We restricted participation to persons 16–45 y of age who had been residing in the Wenatchee area for at least 1 year and who were planning to be in the area the following spring (i.e., would be available for follow-up testing). Exclusion criteria included having mixed, loaded, or applied pesticides during the preceding 6 mo; any self-reported previous pesticide poisonings; and history of diabetes, epilepsy, or meningitis. All subjects were currently or recently employed. Recruitment, examinations, and interviews were conducted in Spanish. Informed consent was obtained from all participants (or from a parent or guardian of minors). The study protocol was approved by the University of Washington Human Subjects Committee.

Exposed subjects comprised apple thinners who were recruited from orchards or worker camps. We chose apple thinners in particular because they are exposed routinely to foliar OP pesticide residues and can experience systemic absorption even in the absence of related symptoms.[9–11] Of the 69 thinners who met the study selection criteria, 68 (99%) participated. We tested all subjects as close to the end of the thinning season as possible. We oversampled women from the farm-worker population to facilitate gender-specific analyses.

We recruited reference subjects from local workplaces, which included sewing factories; food processors; fruit-packing houses; recreational areas (e.g., golf courses, ski resorts); restaurants; and bakeries. We matched reference subjects to exposed subjects by gender, ethnicity (i.e., Hispanic, non-Hispanic Caucasian, and other), age (± 5 y), and years of education (± 3 y). Of the seventy-six persons who met the study selection criteria and who were matched to an exposed subject, 69 (91%) participated. After examination, we excluded 1 exposed and 1 nonexposed subject from the analyses because they were over 45 ys old (exposed) or were non-Hispanic (nonexposed) and could not be matched.

**Exposure assessment.** We considered a subject exposed if he or she had thinned apples for 80 or more hours during the current season (i.e., from the beginning of the calendar year to the examination date). Exposure was determined via interviewer-administered questionnaire. The interviewer asked each subject for detailed information about all farm work performed in the current season, and also obtained a work history. Information included activities, dates for each activity, names and locations of farms, use of protective clothes and practices, and any known pesticide exposures. Interviewers also asked subjects about proximity of their homes to nearby farms, use of pesticides in or around the home, use of medications, and alcohol and tobacco consumption. Erythrocyte cholinesterase activity, which has long been used for monitoring OP-exposed workers,[12] was measured for each subject, using the Testmate® Cholinesterase Kit (EQM Research [Cincinnati, Ohio]) and a venous blood sample. We adjusted cholinesterase activity for hemoglobin level.[13] Additional detailed exposure information[14] was available for 13 (19%) of the exposed and 5 (7%) of the reference subjects who participated in a concurrent occupational hygiene study. This information and crop-protection recommendations provided to orchardists[14] indicate that the thinners in this study were exposed primarily to azinphosmethyl and possibly to phosmet or methyl parathion.

**Examination of nerve function.** We used surface electrodes to examine nerves in each subject's right upper and lower limbs. The tester used standard procedures[15] to perform testing with a portable Neuropak 2 XPD electrodiagnostic instrument (Nihon Kohden [Irvine, CA]) in a temperature-controlled room (at 22 °C). The tester was blinded to subjects' exposure status.

Sensory nerve conduction was assessed using the sural nerve. The active recording electrode was placed behind the lateral malleolus, and the reference electrode was placed 3 cm distally. Supramaximal stimulation, at a rate of 1 Hz, was applied 14 cm proximal to the active recording electrode on the posterior leg. Twenty responses were averaged, with filter settings of 20 Hz–2 kHz. Antidromic onset latency (Sens Lat) and onset to peak amplitude (Sens Amp) were measured.

Motor nerve function was examined in the ulnar nerve. An active recording electrode was placed over the abductor digiti minimi, and a reference electrode was placed distally over the fifth metacarpal-phalangeal joint. Supramaximal stimuli were delivered at the wrist (8 cm proximal to the active recording electrode) and at the elbow, with filter settings of 2 Hz–10 kHz. We calculated motor nerve conduction velocity (MCV) by dividing the difference in latency between elbow and wrist stimulation by the measured distance between these two points. Repetitive activity following a single stimulus was noted when present.

To test neuromuscular junction function, the tester performed repetitive stimulation (Rep Stim) on the ulnar nerve, administering 5 stimulations at a rate of 3 Hz. Recording parameters were the same as those for motor nerve conduction studies. The nerve was stimulated at the wrist at rest (a), immediately after 30 s of maximal isometric exercise (b), and 2 min after exercise (c). The isometric exercise involved having the subject abduct the right small finger against a fixed object. The testing immediately after, and 2 min after, this exercise assessed postexercise facilitation and postexercise exhaustion, respectively. We determined the decrement in response by calculating the ratio of the amplitudes of the fourth to the first action potential for the first train (a4/a1), second train (b4/b1), and third train (c4/c1). We assessed (1) postexercise facilitation via the ratio of the amplitude of the first action potential in the second train to the amplitude of the first action potential in the first train (b1/a1) and (2) postexercise exhaustion via the ratio of the amplitude of the first action potential in the third train to the amplitude of the first action potential in the first train (c1/a1) (modification of Misra et al.)[5]

A board-certified electromyographer (LRR) reviewed waveforms in a blinded fashion after data were collected. When motor responses recorded from abductor digiti minimi with wrist stimulation differed appreciably between motor conduction studies and repetitive stim-

ulation studies (a1 response), we conservatively assumed that one of the stimuli was not supramaximal; these subjects were excluded from all analyses of that measure.

**Data analysis.** We analyzed continuous outcomes with Student's t test and multiple linear regression. We analyzed dichotomous outcomes as prevalence ratios (PRs) via stratified analysis. We treated exposure, based on the number of hours spent thinning apples in the current season, as dichotomous (< 80/≥ 80) in our comparisons of thinners with nonthinners, and as continuous or trichotomous (< 200/200–399/≥ 400) in comparisons among thinners. We denoted other farm work as an indicator variable. A 5% two-sided level of significance was used.

Prevalence ratio analyses were also performed; we used the bottom 10th and 50th percentiles of sural nerve function—as determined by latency (Sens Lat10 and Sens Lat50, respectively) and amplitude (Sens Amp10 and Sens Amp50, respectively)—where longer latency and smaller amplitude indicate impaired function. We performed similar analyses for ulnar motor conduction velocity (MCV10 and MCV50, respectively), for which a deficit is indicated by slower velocity. In prevalence ratio analyses of neuromuscular function, we considered a4/a1, b4/b1, and c4/c1 abnormal if decrement exceeded 10%; b1/a1 and c1/a1 were considered abnormal if the difference (increase or decrease) exceeded 50% (based on Kimura[16]).

The other factors we considered in the analysis were age, gender, height, other (nonthinning) farm work performed in the current season, history of farm work, history of working with pesticides (mixing, loading, or applying), proximity of the home to nearby farms, use of pesticides in or around the home, and alcohol and tobacco consumption. For analyses restricted to light or moderate alcohol consumers, we defined heavy consumption as either (a) 6 or more drinks per drinking session or (b) 3 or more drinks per session and 3 or more sessions per week. Factors we examined in exposed-only analyses also included timing of thinning relative to testing, use of protective clothes and practices, number of days work clothes were worn between washing, and frequency of bathing. We analyzed data with the SPSS for Windows statistical program (SPSS [Chicago, Illinois]).

One reference and 1 exposed subject refused to give blood samples. One reference subject, whose adjusted cholinesterase activity was extremely elevated as a result of an abnormally depressed hemoglobin count, was excluded from all cholinesterase analyses. One exposed subject refused all nerve-conduction tests. One reference subject refused any further Rep Stim tests after the first test, and 1 exposed subject refused after the second test. Three reference and 2 exposed subjects, for whom Rep Stim stimuli were not supramaximal, were excluded from those analyses. The tester was unable to obtain clear sural readings on 5 exposed and 3 reference subjects. These subjects did not differ appreciably from the remainder of study subjects in other characteristics.

## Results

**Characteristics of subjects.** As can be seen in Table 1, the exposed and comparison groups were well matched for ethnicity, gender, and age. A history of farm work prior to the 1994 thinning season was reported by 55 (82.1%) of the exposed versus 45 (66.2%) of the reference subjects. Other (nonthinning) farm work in the current season was also more common among exposed subjects (i.e., reported by 47 thinners [70.1%] versus 15 reference subjects [22.1%]). None of the subjects smoked more than one pack of cigarettes per day; 13 (19.4%) exposed, compared with 18 (26.5%) reference subjects reported smoking one pack or less per day. The two groups reported similar alcohol consumption histories; approximately one-quarter of the subjects in each group reported heavy consumption.

Of the 67 thinners tested, 29 (43.3%) were female (Table 2). Females performed significantly fewer hours of thinning than did males (median hours—230.0 versus 355.8, respectively), and they started and ended earlier in the season (median weeks since starting—11.1 versus 8.3, respectively; median weeks since end-

**Table 1.—Selected Characteristics of Study Subjects**

| Characteristic | Exposed* | | Nonexposed* | |
|---|---|---|---|---|
| | *n* | % | *n* | % |
| Hispanic race | 67 | 100.0 | 68 | 100.0 |
| Gender | | | | |
| Female | 29 | 43.3 | 31 | 45.6 |
| Male | 38 | 56.7 | 37 | 54.4 |
| Age (y) | | | | |
| < 25 | 26 | 38.8 | 26 | 38.2 |
| 25–34 | 30 | 44.8 | 29 | 42.6 |
| ≥ 35 | 11 | 16.4 | 13 | 19.1 |
| Height (cm) | | | | |
| < 155 | 17 | 25.4 | 14 | 20.6 |
| 155–169 | 35 | 52.2 | 38 | 55.9 |
| ≥ 170 | 15 | 22.4 | 16 | 23.5 |
| Did farm work in previous seasons | 55 | 82.1 | 45 | 66.2 |
| Performed other (nonthinning) farm work this season | 47 | 70.1 | 15 | 22.1 |
| Current smoking (packs/d) | | | | |
| 0 | 54 | 80.6 | 50 | 73.5 |
| ≤ 1 | 13 | 19.4 | 18 | 26.5 |
| Alcohol consumption history | | | | |
| Never, or < 1 oz/mo | 28 | 41.8 | 30 | 44.1 |
| Stopped > 6 mo ago | 6 | 9.0 | 6 | 8.8 |
| Current | 33 | 49.3 | 32 | 47.1 |
| Heavy alcohol use† | 17 | 25.4 | 15 | 22.1 |

*Exposed = subjects with ≥ 80 h of thinning this season, and nonexposed = subjects with < 80 h of thinning this season.
†Defined as either (a) six or more drinks/drinking session or (b) three or more drinks/session and three or more sessions/wk during the previous 6 mo.

**Table 2.—Exposure Characteristics of Apple Thinners**

| Characteristic | Males and females ($n$ = 67) | | Females ($n$ = 29) | | Males ($n$ = 38) | |
|---|---|---|---|---|---|---|
| | $n$ | % | $n$ | % | $n$ | % |
| Hours of thinning this season | | | | | | |
| < 200 | 13 | 19.4 | 9 | 31.0 | 4 | 10.5 |
| 200–399 | 35 | 52.2 | 14 | 48.3 | 21 | 55.3 |
| ≥ 400 | 19 | 28.4 | 6 | 20.7 | 13 | 34.2 |
| Weeks since start of thinning | | | | | | |
| < 7 | 17 | 25.4 | 5 | 17.2 | 12 | 31.6 |
| 7–9 | 23 | 34.3 | 5 | 17.2 | 18 | 47.4 |
| ≥ 10 | 27 | 40.3 | 19 | 65.5 | 8 | 21.1 |
| Weeks since end of thinning | | | | | | |
| < 1 | 26 | 38.8 | 5 | 17.2 | 21 | 55.3 |
| 1–3 | 22 | 32.8 | 8 | 27.6 | 14 | 36.8 |
| ≥ 4 | 19 | 28.4 | 16 | 55.2 | 3 | 7.9 |
| Did other (nonthinning) farm work this season | 47 | 70.1 | 18 | 62.1 | 29 | 76.3 |

**Table 3.—Neurophysiological Outcomes of Study Subjects**

| Neurological measure | Exposed* | | Nonexposed† | | Mean difference | | | |
|---|---|---|---|---|---|---|---|---|
| | $\bar{x}$ | $SD$ | $\bar{x}$ | $SD$ | Unadjusted | 95% CI | Adjusted | 95% CI‡ |
| Sural nerve | | | | | | | | |
| Latency (ms) | 2.7 | 0.3 | 2.6 | 0.2 | 0.0 | 0.0, 0.1 | 0.1 | 0.0, 0.2 |
| Amplitude (μV) | 24.5 | 11.1 | 22.7 | 11.3 | 1.7 | −2.3, 5.7 | 0.9 | −3.7, 5.5 |
| Ulnar nerve | | | | | | | | |
| Velocity (m/s) | 60.0 | 3.8 | 61.2 | 4.1 | −1.1 | −2.5, 0.2 | −1.3 | −2.9, 0.3 |
| Rep stim | | | | | | | | |
| a4/a1 | 1.01 | 0.05 | 1.01 | 0.05 | 0.00 | −0.01, 0.02 | −0.01 | −0.03, 0.01 |
| b4/b1§ | 1.08 | 0.38 | 1.07 | 0.23 | 0.01 | −0.10, 0.12 | 0.00 | −0.13, 0.14 |
| c4/c1 | 1.04 | 0.12 | 1.04 | 0.09 | 0.00 | −0.04, 0.04 | 0.01 | −0.04, 0.05 |
| b1/a1§ | 0.93 | 0.30 | 0.90 | 0.30 | 0.03 | −0.07, 0.14 | 0.01 | −0.11, 0.14 |
| c1/a1 | 0.94 | 0.21 | 0.89 | 0.27 | 0.06 | −0.03, 0.14 | 0.06 | −0.04, 0.16 |

*Sural latency—$n$ = 62, amplitude—$n$ = 60; ulnar velocity—$n$ = 66; a4/a1—$n$ = 64, b4/b1—$n$ = 64, c4/c1—$n$ = 63, b1/a1—$n$ = 64, and c1/a1—$n$ = 64.
†Sural latency—$n$ = 65, amplitude—$n$ = 61; ulnar velocity—$n$ = 68; a4/a1—$n$ = 65, b4/b1—$n$ = 63, c4/c1—$n$ = 64, b1/a1—$n$ = 63, and c1/a1—$n$ = 63.
‡Adjusted for age, height, alcohol consumption, other (non-thinning) farm work in the current season, and history of farm work.
§One reference subject removed from analysis as an outlier.

ing—5.6 versus 0.3, respectively). Females were also less likely to have engaged in other farm work during the season: 18 (62.1%) females reported other farm work, compared with 29 (76.3%) males. Females reported washing their work clothes more frequently than did males (median days of use between washings—1 versus 2, respectively), but they reported bathing with equal regularity (median days between bathing—1).

**Cholinesterase activity.** Mean cholinesterase activity was slightly, but significantly, lower in the exposed group (29.9 IU/g Hgb) than in the reference group (31.3 IU/g Hgb), with a mean difference of −1.4 (95% confidence interval [95% CI] = −2.5, −0.3). When we restricted analysis to exposed subjects, we observed a nonsignificant trend between number of thinning hours and cholinesterase activity, with a slope of −0.24 IU/g Hgb/100 h of thinning (95% CI = −0.64, 0.15). This relationship remained nonsignificant when we examined it separately by gender, although the effect was in the expected direction for both. The group mean difference of 0.2 IU/g Hgb (95% CI = −1.1, 1.5) between male and female thinners was also not significant.

Time since start of thinning, time since end of thinning, number of days work clothes were worn between washings, and bathing pattern were not predictive of cholinesterase activity and did not confound the relationship between thinning hours and cholinesterase activity was not confounded.

**Nerve conduction.** All values for Sens Lat were within the normal range.[15] One thinner and 2 reference

**Table 4.—Neurophysiological Outcomes of Apple Thinners**

| Neurological measure | Hours of thinning: < 200 $\bar{x}$ | < 200 SD* | 200–399 $\bar{x}$ | 200–399 SD† | ≥ 400 $\bar{x}$ | ≥ 400 SD‡ | Regression estimate (change/100 h thinning): Unadjusted b | Unadjusted 95% CI | Adjusted§ b | Adjusted§ 95% CI |
|---|---|---|---|---|---|---|---|---|---|---|
| Sural nerve | | | | | | | | | | |
| Latency (ms) | 2.7 | 0.3 | 2.7 | 0.2 | 2.7 | 0.3 | 0.0 | –0.1, 0.0 | 0.0 | –0.1, 0.0 |
| Amplitude (μV) | 19.7 | 10.7 | 26.5 | 11.7 | 23.8 | 9.7 | –0.4 | –2.1, 1.4 | 0.0 | –1.8, 1.9 |
| Ulnar nerve | | | | | | | | | | |
| Velocity (m/s) | 59.7 | 4.6 | 60.1 | 3.9 | 60.3 | 3.1 | 0.2 | –0.4, 0.8 | 0.3 | –0.3, 0.9 |
| Rep stim | | | | | | | | | | |
| a4/a1 | 1.02 | 0.05 | 1.01 | 0.05 | 1.01 | 0.04 | 0.00 | –0.01, 0.01 | 0.00 | –0.01, 0.01 |
| b4/b1 | 1.10 | 0.28 | 1.10 | 0.47 | 1.01 | 0.25 | –0.03 | –0.09, 0.03 | –0.03 | –0.10, 0.03 |
| c4/c1 | 1.03 | 0.05 | 1.06 | 0.16 | 1.01 | 0.08 | –0.01 | –0.02, 0.01 | –0.01 | –0.03, 0.01 |
| b1/a1 | 0.87 | 0.33 | 0.95 | 0.28 | 0.94 | 0.31 | 0.02 | –0.02, 0.07 | 0.02 | –0.02. 0.07 |
| c1/a1 | 0.98 | 0.09 | 0.92 | 0.27 | 0.96 | 0.13 | 0.00 | –0.03, 0.03 | 0.00 | –0.03, 0.04 |

*Sural latency—$n$ = 12, amplitude—$n$ = 11; ulnar velocity—$n$ = 13; a4/a1—$n$ = 13, b4/b1—$n$ = 13, c4/c1—$n$ = 13, b1/a1—$n$ = 13, and c1/a1—$n$ = 13.
†Sural latency—$n$ = 32, amplitude—$n$ = 32; ulnar velocity—$n$ = 34; a4/a1—$n$ = 33, b4/b1—$n$ = 33, c4/c1—$n$ = 32, b1/a1—$n$ = 33, and c1/a1—$n$ = 32.
‡Sural latency—$n$ = 18, amplitude—$n$ = 17; ulnar velocity—$n$ = 19; a4/a1—$n$ = 18, b4/b1—$n$ = 18, c4/c1—$n$ = 18, b1/a1—$n$ = 18, and c1/a1—$n$ = 18.
§Adjusted for age, height, alcohol consumption, and other (non-thinning) farm work in the current season

subjects had subnormal Sens Amps (< 5 μV), whereas 1 thinner and 1 reference subject had slightly subnormal MCVs (< 50 m/s). We observed these abnormalities in 5 different subjects.

Treated as continuous variables, none of the neurophysiological measures (i.e., Sens Lat, Sens Amp, MCV, a4/a1, b4/b1, c4/c1, b1/a1, and c1/a1) was significantly different between the thinners and reference subjects (Table 3). Height, which was associated significantly only with Sens Lat and Sens Amp, did not confound the relationship between thinning and any nerve function. No effect of alcohol consumption on nerve function was observed, nor expected, given the levels of alcohol consumption seen in this study population. Adjusted and unadjusted estimates were very similar.

When analyses were restricted to thinners, no significant dose-response relationship was observed between thinning hours and any neurophysiological measure (Table 4). Sural amplitude (Sens Amp) increased from the lowest exposure group to the intermediate (200–399 h) group, but it then decreased between the intermediate group and the highest-exposure group. This increase was not significant, and the mean Sens Amp for thinners with 200 or more thinning hours (25.5 μV) was not appreciably higher than the mean for the nonexposed group (22.7 μV) ($p$ =.20).

Among thinners, other (nonthinning) farm work was predictive of a small increment in a4/a1, even with inclusion of thinning hours in the model (mean difference = 0.04 [95% CI = 0.01, 0.06]). However, the group means were well within the normal range for both those who engaged in other farm work and those who did not. None of the other potential confounding factors was predictive of nerve function.

In comparisons of the bottom 10th and 50th percentiles of nerve function, only MCV50 showed any appreciable association with thinning (PR = 1.4 [95% CI = 1.0, 1.9). This association was not evident in exposure-response comparisons among thinners. Repetitive activity of the ulnar nerve following a single stimulation was common in both exposed and nonexposed subjects (14.9% and 22.1%, respectively) and was not significantly associated with thinning.

Results were similar when we performed analyses separately by gender. The exclusion of heavy alcohol consumers and former pesticide handlers from the analyses also had no appreciable effect on results, nor did the restriction of analyses to thinners who had thinned within the past 4 wk. In analyses of neuromuscular function, use of the amplitude of the second action potential, instead of the fourth (i.e., a2/a1, b2/b1, and c2/c1), produced results comparable with those in which the fourth was used.

## Discussion

We designed the current study to (a) test whether peripheral neurophysiological abnormalities were present in farm workers after a season of low-level organophosphate pesticide exposure and (b) assess dose-response trends.

The results indicate that OP pesticide exposure during a growing season at the low levels observed in these workers was not related to detectable impairment in peripheral nerve conduction or neuromuscular function. Differences between thinners and nonthinners and between exposure groups among thinners were small, compared with the normal variability of these measures. No dose-response relationship, based on time spent thinning in OP-sprayed orchards, was evident in these workers.

Results from other studies involving persistent low-level OP exposures are mixed. In several of these studies, investigators examined subjects who were also exposed to other neurotoxins. Jager et al.[4] reported repetitive activity, reduced voltage potential, and decreased b1/a1 of the ulnar nerve among workers in an organophosphate/organochlorine pesticide-manufacturing plant. Roberts[7] found decreased ulnar voltage potential and MCV among OP pesticide factory workers. Drenth et al.[3] observed both abnormally low and high voltage potentials in the ulnar nerve among a group of pesticide mixers and applicators who handled a broad spectrum of neurotoxic pesticides, which included organophosphates, organochlorines, and organic mercury and tin compounds. In a study of fenthion applicators, Misra et al.[5] found no evidence of peripheral neuropathy and no significant differences in nerve-conduction measures between exposed and nonexposed workers. However, they did report a number of peroneal and median nerve-conduction measures that were significantly and adversely associated with exposure when they used applicators as their own reference group (after several weeks of nonexposure). Ring et al.,[6] who studied the association between electromyogram (EMG) measures and mild symptoms of OP intoxication among aerial OP spray workers, found correlations between symptom severity and several measures of nerve impairment, including decreased MCV and increased ulnar motor and sensory distal latency. In contrast, Jusic et al[17] found no neuromuscular abnormalities in either the median or ulnar nerves among a group of OP pesticide handlers and applicators. Stalberg et al.[8] found no motor nerve deficits, and they found only a slight reduction in sural sensory conduction velocity among OP applicators.

In the present study, we examined subjects exposed primarily to OPs and not engaged in direct handling of pesticides (i.e., mixing, loading, applying, or manufacturing); that restriction may account for the absence of apparent effects. The subjects examined by Drenth et al.[3] and Jager et al.,[4] while showing only mild cholinesterase depression, were also exposed to other neurotoxic pesticides at unreported levels; that additional exposure may account for the observed neurophysiological abnormalities. Subjects in the remaining studies in which positive results were reported apparently received substantially higher exposures to OPs than did the subjects in our study. Misra et al.[5] observed approximately 31% lower cholinesterase levels in OP applicators than referents; these levels increased 23% after 3 wk of nonexposure. Stalberg et al.,[8] who found only a slight decrease in sensory conduction velocity in OP applicators, reported a decrease of 8% in cholinesterase levels after OP exposure. In contrast, thinners in our study had cholinesterase levels only 4% lower than in reference subjects. Ring et al.,[6] who did not report cholinesterase levels, included in their study a substantial number of spray workers who displayed mild symptoms of OP intoxication in a study undertaken after a series of fatal crashes by aerial OP applicators. Roberts[7] studied pesticide factory workers, who likely experienced higher exposures than the subjects in our study, but no dose estimates were provided. In the present study, we also excluded persons who had ever experienced a pesticide poisoning and who, therefore, were at risk of acute organophosphate-induced delayed neuropathy.

Nonetheless, thinners in our study were exposed to detectable levels of OPs. In the concurrent occupational hygiene investigation by Simcox et al.,[14] which examined some of the same subjects as were in this study, they found significantly elevated levels of the azinphosmethyl metabolite, dimethylthiophosphate (DMTP), in the urine of thinners, compared with nonthinners (median = 0.32 μg DMTP/ml urine; range = 0.00–3.96 versus median = 0.02; range = 0.00–0.18, respectively) ($p < .001$). They also found detectable foliar OP residues in the orchards in which the thinners were working (median = 0.56 μg/cm$^2$; range: 0.24–3.35). These orchards were sprayed with OP pesticides one to four times (median = 2.5) during the thinning period, with re-entry times of 2–49 d (median = 16). The remaining thinners included in the current study likely received exposures similar to those described above. We are unaware of any similar investigations in apple thinners; however, the urinary metabolite levels reported in our study are appreciably lower than those observed in comparable studies among peach thinners.[18,19]

Although in this study we observed a high prevalence of repetitive activity in the ulnar nerve, this phenomenon did not appear to be associated with pesticide exposure. In fact, the prevalence of repetitive activity in thinners was slightly lower than in the comparison group. Also, this repetitive activity was not related to a history of farm work or to prior pesticide handling. Although this phenomenon has been observed in other studies of persistent OP exposure,[4,5] such a high prevalence in this study sample is surprising and difficult to explain, given its rarity among healthy individuals.[16]

The observed repetitive activity could be an artifact of nerve-conduction measurement. These tests can be technically difficult to perform, and there are numerous factors that can produce false positive or false negative results (e.g., movement of electrodes during stimulation, improper electrode placement, changes in skin resistance or intramuscular temperature).[8] Arguing against this is the high degree of agreement between subjects who showed repetitive activity in the present study, and in a follow-up study performed the following year with a different nerve-conduction technician (kappa = 0.7, $p < .001$; $n = 86$). Furthermore, standard deviations for all neurophysiological measures were typical of those reported elsewhere for normal populations,[15] with the exception of a somewhat higher standard deviation associated with Sens Amp.

The threshold effect for Sens Amp among thinners has not been reported previously, and it most likely resulted from the small size ($n = 11$) of the lowest-exposure thinning group, with the consequent instability of its mean Sens Amp estimate.

One important limitation of this study was the lack of specific exposure data for the vast majority of subjects (87%). A subject's duration of thinning work is an

imprecise measure of actual pesticide exposure. In this study, we failed to observe any neurophysiological impairment associated with either duration of thinning (continuous, in hours) or employment in thinning (dichotomous, yes/no). It is possible that any effects among thinners in the present study were too small for us to detect, given our sample size and the normal population variability of these neurophysiological measures. More exposed subjects ($n$ = 67) were included in our study than in many of the previous studies in which abnormalities were reported (e.g., Jager et al.[4] ($n$ = 36); Misra et al.[5] ($n$ = 24); Stalberg et al.[8] ($n$ = 11)), suggesting that effects, if present, were small. Misra et al.[5] observed effects only when the exposed acted as their own reference group (and not when they were compared with an unexposed reference group).

The large number of reference subjects in this study who had a history of farm work in prior seasons or who had performed other, nonthinning, farm work in the current season could have attenuated differences in neurophysiological function between exposure groups. If persistent low-level OP exposure at the levels observed in this study does, in fact, cause chronic neurophysiological deficits, then overmatching may be present. However, the fact that most reference subjects had normal neurophysiological measures suggests that this is not the case (at least among this young a group [mean age = 28.2 y] with relatively few years of possible agricultural employment). In addition, the high prevalence of repetitive activity among reference subjects was also present among those reference subjects who had never engaged in farm work (i.e., 6 of 15).

Occupational OP exposure among reference subjects was likely minimal. None of the reference subjects reported any contact with pesticides in the workplace. Workers in fruit-packing houses and food processors typically do not handle the produce until it has been washed. In the analysis, we accounted for any nonthinning farm work in the current season.

Cholinesterase activity was significantly lower in the exposed group than in the reference group ($p$ = .01); however, the difference was small, and this biomarker was not adequate for us to judge individual exposures. Cholinesterase activity was not associated with time since the end of thinning, despite the fact that a large percentage (28.4%) of exposed subjects had stopped thinning more than 4 wk prior to examination. One would expect blood cholinesterase activity to return steadily to normal as red blood cells are replaced approximately every 120 d. However, blood cholinesterase activity shows marked intra- and interindividual variation.[13] Without a baseline value for an individual, it is generally not possible to assess the extent of OP-induced cholinesterase inhibition and, therefore, OP exposure. In this study, cholinesterase activity was not predictive of neurophysiological function.

## Conclusion

Exposure of farm workers during one season to the low levels of OP pesticides experienced by our subjects did not appear to be associated with impaired peripheral neurophysiological function. However, these findings may not apply to other settings, such as in developing countries or among pesticide mixers, loaders, or applicators, for which pesticide exposures may be substantially higher.

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