

# Long-term Neurobehavioral Effects of Mild Poisonings with Organophosphate and n-Methyl Carbamate Pesticides among Banana Workers

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Organophosphate poisoning has been associated with chronic neurobehavioral dysfunction, but no epidemiologic data exist with regard to long-term consequences from carbamate poisoning. This cross-sectional study evaluated the neurobehavioral performances of 81 banana workers who, on average 27 months earlier, had received medical attention not requiring hospitalization for mild occupational poisoning by either an organophosphate or a carbamate pesticide. These performances were compared with those of 130 banana workers who had never sought medical attention for pesticide poisoning. Poisoned subjects did less well than controls on tests measuring psychomotor and visuomotor skills, language function, and affect, the differences being significant for coding skills on the Digit-Symbol test and two tests of neuropsychiatric symptoms. These deficits, in particular a marked increase of neuropsychiatric symptoms, occurred among the organophosphate-poisoned subjects, but small deficits in performance were also seen in the carbamate-poisoned subjects. The performances of the previously poisoned subjects who had had contact with cholinesterase inhibitors within three months before testing were particularly poor. These findings in workers with mild poisoning are consistent with previous findings of persistent damage to the central nervous system from organophosphate poisoning. The possibility of persistent neurobehavioral effects associated with poisonings by n-methyl carbamate insecticides cannot be excluded. Workers with histories of poisoning may be more susceptible to

neurobehavioral effects with subsequent exposures. *Key words:* carbamate; cholinesterase inhibitor; Costa Rica; cross-sectional study; developing country; epidemiology; neurobehavioral effects; occupational exposure; organophosphate; pesticides; poisoning.

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A majority of the pesticide poisonings in developing countries is caused by neurotoxic organophosphates and carbamates.<sup>1,2</sup> These cholinesterase-inhibiting pesticides are also a public health problem in some industrialized countries.<sup>3</sup> The possibility of long-term neurotoxic effects from poisonings caused by cholinesterase-inhibiting pesticides is a global concern.

Cholinesterase inhibitors bind to acetylcholinesterase, the enzyme that controls the transmission of the nerve impulses at the cholinergic synapses throughout the nervous system. This results in the accumulation of acetylcholine at the neurojunctions and the appearance of a cholinergic syndrome.<sup>4</sup> Organophosphates bind irreversibly to acetylcholinesterase, and subsequent "aging" of the phosphorylated enzyme may occur, which makes the reactivation of cholinesterases dependent on the synthesis of new enzymes. Chronic nervous system effects have been observed among workers previously poisoned with organophosphate pesticides, in case reports of poisoned patients<sup>5-7</sup> and in well-designed epidemiologic studies.<sup>8-10</sup>

Unlike organophosphates, carbamates occupy the binding sites at the target enzymes in a reversible way and without the "aging" reactions. Persistent neurologic effects are therefore assumed to be unlikely.<sup>4</sup> However, few carbamates have been submitted to extensive neurotoxicity testing.<sup>11</sup> Some case reports describe chronic neurologic effects from carbamate poisonings,<sup>11-14</sup> but no results from analytic epidemiologic studies exist in the published literature.

In Costa Rica, both organophosphate and carbamate nematocides (worm killers) are widely used on banana plantations. In 1986, the banana plantation region of

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Costa Rica had one of the highest incidence rates of poisonings reported in the world.<sup>15</sup> Since the 1990s, improvements on banana plantations have diminished the incidence of poisonings,<sup>16</sup> and those that occur are usually fairly mild. This study was undertaken to determine whether poisoning with an organophosphate or a carbamate compound is associated with long-term effects on the central nervous system.

## METHODS

The performances of previously poisoned workers on a battery of neurobehavioral tests were compared with the performances of a control group of non-poisoned workers, poisoning versus no poisoning being the measure for exposed versus unexposed.

### *Study Population*

This study was conducted in two counties in the Atlantic Region of Costa Rica. It was done on Costa Rican men, aged 15–55 years at the time of testing, who had been banana workers during at least one year and who did not have a history of any disease or condition that might induce nervous system damage or interfere with proper testing (severe head trauma, severe alcohol or drug abuse, hearing impairment, inability to “read” numbers, history of epilepsy, or serious illness such as cancer).

Subjects who had had systemic occupational poisonings caused by cholinesterase-inhibiting pesticides treated at any of the health centers in the study area at least one year prior to testing were considered eligible for inclusion in the poisoned (*exposed*) group. Poisoned subjects were identified from compulsory occupational accident reports to the National Insurance Institute. Name, personal identification number, home address, plantation, and pesticide were extracted from the reports. The poisoned workers were then traced by screening the payrolls of all banana plantations in the study area during May–June and again in August 1994. We were able to locate 94 (58%) of the 162 poisoned workers identified in the record search. Of the 82 eligible poisoned workers, one refused to participate. The final exposed group consisted of 81 poisoned workers (mean number of months since poisoning = 27, range 12–43).

A control group of non-poisoned (*unexposed*) workers was randomly drawn from the payrolls of participating banana plantations (one of the four large multinational companies in the study area and practically all other smaller companies). Subjects who had histories of receiving medical treatment for pesticide poisoning at any time of their lives were not eligible. Of the 144 eligible controls, ten refused to participate and four were excluded because a plantation manager had arbitrarily substituted them for the randomly chosen workers (response rate 90%). The final unexposed group consisted of 130 non-poisoned workers. These controls

included field workers and employees in the packing plants who had never had any contact with cholinesterase-inhibiting pesticides, as well as field workers who worked with cholinesterase-inhibiting pesticides in varying degrees. A subset of these workers reported having experienced at any time in their life mild symptoms associated with pesticide exposures without need for medical attention. Some of the results are reported separately for this subgroup of controls.

### *Potential Confounders*

Information about demographic variables and diseases or conditions that could affect performance on the tests was collected by means of a structured interview (Table 1). A complete work history was taken. Indices of lifetime exposures to cholinesterase-inhibiting pesticides as well as to other pesticides were constructed based on the number of days of exposure per year, the number of years exposed, the fraction of the day exposed per job title, and the use of protective equipment. Near visual acuity (poor, mediocre, good) was assessed with a short printed text and a chart with numbers. Confounders additional to those in Table 1 that were considered in the analysis included medical confounders such as history of convulsions and other neurologic or psychiatric disorders, malaria, chronic metabolic and infectious disorders; current use of medication; examiner; hours of sleep the night prior to testing; and sense of well-being and liquor and caffeine intakes on the day of testing.

It was not feasible to conduct the field work outside a spraying season because different plantations spray nematocides at different times. Thus, a substantial proportion of the study population had been in contact with cholinesterase-inhibiting pesticides within three months of the examination (Table 2). None of these workers reported symptoms of poisoning at the time of testing. Red blood cell and plasma cholinesterase activities were determined with the Test-Mate organophosphate kit (EQM Inc., Cincinnati, Ohio),<sup>17</sup> which uses a modified Ellman method and corrects for hemoglobin. There was no difference between the mean temperature-adjusted red blood cell cholinesterase activity levels in the poisoned and non-poisoned groups, or between subsets with and without recent exposure. The mean value of plasma cholinesterase levels of the workers with recent contact with cholinesterase inhibitors was lower than the mean for the workers without recent contact (2.11 U/mL versus 2.26 U/mL blood,  $p = 0.01$ ).

### *Poisonings*

All of the 81 poisoned subjects reported that poisonings had not been severe enough to require hospitalization. This was confirmed by a review of medical records for all but 20 subjects. Cholinesterase levels

**TABLE 1 Characteristics of the Study Population**

| Characteristic  | Poisoned<br><i>n</i> = 81 | Non-poisoned<br><i>n</i> = 130 |
|---|---------------------------|--------------------------------|
| Age, mean (SD)  | 28.3 years (7.6)          | 29.0 years (10.0)              |
| Formal education, mean (SD)   | 5.5 years (2.1)           | 6.0 years (2.2)                |
| Contact with cholinesterase inhibiting pesticides within 3 months before testing      | 54.3%                     | 27.7%                          |
| Cumulative lifetime exposure to cholinesterase-inhibiting pesticides <sup>a</sup>     |                           |                                |
| Lowest tertile  | 18.5%                     | 41.5%                          |
| Mid tertile   | 44.5%                     | 28.5%                          |
| Highest tertile   | 37.0%                     | 31.0%                          |
| Cumulative lifetime exposure to non-cholinesterase-inhibiting pesticides <sup>a</sup> |                           |                                |
| Low   | 32.1%                     | 34.6%                          |
| Medium  | 32.1%                     | 33.9%                          |
| High  | 35.8%                     | 31.5%                          |
| Red blood cell cholinesterase level, mean (SD)  | 29.6 U/mL (3.4)           | 29.6 U/ml (2.9)                |
| Plasma cholinesterase level, mean (SD)  | 2.18 U/ml (0.41)          | 2.22 U/ml (0.41)               |
| Alcohol consumption   |                           |                                |
| Current drinkers  | 55.6%                     | 55.4%                          |
| Ever drinkers   | 66.7%                     | 68.5%                          |
| Ever heavy drinkers   | 7.4%                      | 14.6%                          |
| Ever smokers  | 43.2%                     | 42.3%                          |
| Time of day of testing  |                           |                                |
| Morning   | 53.1%                     | 26.9%                          |
| Afternoon   | 46.9%                     | 73.1%                          |
| Worked with solvents > 3 months daily and continuously                                | 27.2%                     | 28.5%                          |
| Loss of consciousness > 1 hour  | 8.6%                      | 2.3%                           |
| Poor or mediocre near visual acuity   | 6.2%                      | 7.7%                           |

<sup>a</sup>Exposure index = SUM (number of days per year × number of years × fraction of the day per job title × weight for no use of protective equipment).

were seldom reported, and clinical descriptions in out-patient records were not adequate to further assess severity of the poisonings. However, the workers were interviewed regarding the poisonings with a checklist of 23 typical cholinergic symptoms.<sup>4,18</sup> The mean number of symptoms was 13.7 (SD 3.9), with almost identical numbers for the subjects with and without review of their medical records. The most frequently reported poisoning symptoms included lightheadedness (94%), nausea (91%), general weakness (91%), abdominal pain (89%), excessive sweating (83%), salivation (79%), headache (78%), vomiting (75%), blurred vision (75%), muscle twitching (67%), muscle cramps (56%), and difficulty breathing (51%). Seizures and involuntary loss of urine, which are symptoms of severe poisoning, were acknowledged by 3% and 6%, respectively. Fifteen percent answered positively to “sneezing,” which had been included as a symptom not likely to be related to poisoning in order to be able to adjust for overreporting of poisoning.

Sources of information about the specific pesticides responsible for the poisonings were the medical file, the report to the National Insurance Institute, the worker’s interview, and information provided by the

company about which nematocide had been used on the day of the poisoning. When the sources did not agree, the company’s information was used. Hospital information was used in a documented epidemic of carbofuran poisonings. In all but three cases, the poisonings could be classified as an organophosphate (*n* = 54) or a carbamate (*n* = 24) (Table 2). Of the organophosphate poisonings, 18 were caused by terbufos, six by fenamifos, four by ethoprophos, two by diazinon, and one each by cadusaphos and chlorpyrifos. Twenty-two pesticides were classified only as “organophosphates.” Of the carbamate poisonings, 23 were caused by carbofuran and one by oxamyl.

#### *Neurobehavioral Testing*

Previously trained technicians, who were blind to the poisoning status of the subjects, examined all workers between May and September 1994. Written informed consent was obtained. The costs of travel, food, and lost wages were reimbursed, but the workers did not receive any additional incentive.

A test battery was assembled to assess the main domains of neurobehavioral functions, including

**TABLE 2 Numbers of Subjects in the Exposure Categories and Subgroups**

| Exposure Category   | Number of Subjects  |     |       |
|---|---|-----|-------|
|   | Contact with Cholinesterase Inhibitors within 3 Months before Testing |     | Total |
|   | No  | Yes |       |
| Poisoned with medical attention   | 37  | 44  | 81    |
| Poisoned by organophosphates  | 24  | 40  | 54    |
| Poisoned by n-methyl carbamates   | 12  | 12  | 24    |
| Poisoned by nonspecified cholinesterase inhibitor                       | 2   | 1   | 3     |
| Nonpoisoned   | 94  | 36  | 130   |
| Never any symptoms from pesticide exposures                             | 53  | 23  | 76    |
| Ever an episode of pesticide-related symptoms without medical attention | 41  | 13  | 54    |

memory, attention, psychomotor and visuomotor abilities, language, and affect (Table 3), based on the results from previous research on persistent neurotoxic effects from organophosphate poisonings.<sup>8-10</sup> We used the World Health Organization Neurobehavioral Core Test Battery (WHO-NCTB),<sup>19</sup> which has produced good results in different cultural contexts, except for the Profile of Mood States.<sup>20</sup> Instead, we used the Swedish Questionnaire-16,<sup>21</sup> validated in Nicaragua,<sup>22</sup> and the Brief Symptom Inventory (BSI).<sup>23</sup> In view of the many relatively uneducated subjects in our study population, we administered the BSI (a self-administered test) orally with the help of a visual scoring scale. To assess the neurobehavioral domains more comprehensively, this core battery was complemented with other standardized,

**TABLE 3 Characteristics of the Study Population**

| Neurologic Function                  | Test  |
|--------------------------------------|---|
| Memory                               |   |
| Visual <sup>19</sup>                 | Benton visual retention                     |
| Verbal <sup>24</sup>                 | Rey verbal learning                         |
| Attention                            |   |
| Visual <sup>25</sup>                 | Digit vigilance                             |
| Verbal <sup>19</sup>                 | Digit span                                  |
| Psychomotor                          |   |
| Coordination <sup>19</sup>           | Santana dexterity                           |
| Steadiness <sup>19</sup>             | Pursuit aiming II                           |
| Speed <sup>26</sup>                  | Finger tapping                              |
| Reaction <sup>19</sup>               | Simple reaction time                        |
| Visuomotor                           |   |
| Coding <sup>19</sup>                 | Digit-symbol                                |
| Planning <sup>26</sup>               | Trails-A                                    |
| Problem solving <sup>27</sup>        | Block design                                |
| Language <sup>27</sup>               | Vocabulary                                  |
| Neuropsychiatric symptoms            |   |
| Neurotoxic symptoms <sup>21,22</sup> | Questionnaire-16                            |
| Affect <sup>23</sup>                 | Brief symptom inventory (BSI) <sup>23</sup> |

internationally well-known tests, as listed in Table 3.<sup>19,21-27</sup> The Wechsler vocabulary test was included as a “hold” test to assess premorbid intelligence.<sup>28</sup> All tests had been used in an earlier, similar study of more severe hospitalized poisoning cases in Nicaragua.<sup>9</sup>

### Statistical Analyses

Potential associations between poisoning and neurobehavioral deficits were assessed by means of multiple linear regression analyses. Confounding was assessed by determining for each outcome whether the crude difference of means changed meaningfully following the addition of potential confounders to a univariate model with poisoning status as the explanatory variable, or to a model including poisoning status, age, and education.<sup>29</sup> Age, education, cumulative lifetime exposure to cholinesterase inhibitors, alcohol intake, solvent exposure, loss of consciousness, examiner, time of the day of testing, and plasma cholinesterase level at the time of examination were entered as covariates in all the models. In addition to adjustment for plasma cholinesterase activity in the multivariate analysis, confounding by recent exposure to cholinesterase-inhibiting pesticides was controlled by stratification into subsets with and without contact with cholinesterase inhibitors within the preceding three months. Seven subjects whose near visual acuity was poor were eliminated from the Benton, Pursuit aiming, Digit vigilance, Digit-symbol and Trails-A tests.

Unstandardized and standardized coefficients along with their 95% confidence intervals were estimated for all poisoned workers, for organophosphate- and carbamate-poisoned workers, and for the subgroups with and without recent contact with cholinesterase inhibitors (see Table 2). In addition, we performed analyses comparing the performances of the poisoned subjects with those of the subset of controls restricted to people who had never experienced any symptom attributable to pesticide exposure. The analyses were conducted with the BMDP statistical software.<sup>30</sup>

**TABLE 4 Neurobehavioral Performances of Banana Plantation Workers Previously Poisoned with a Cholinesterase-inhibiting Pesticide, as Compared with Non-poisoned Workers**

| Neurobehavioral Test                           | Poisoned<br>Mean (SD)<br>n = 81 | Non-poisoned<br>Mean (SD)<br>n = 130 | Crude<br>Difference<br>of Means* | Regression<br>Coefficient <sup>b</sup> | 95% CI <sup>c</sup> | Direction <sup>d</sup> |
|--|---------------------------------|--------------------------------------|----------------------------------|--|---------------------|------------------------|
| Benton visual retention (no. correct)          | 6.3 (2.2)                       | 6.9 (2.1)                            | -0.5*                            | -0.4                                   | -1.0, 0.2           | -                      |
| Rey verbal learning (no. after<br>distraction) | 8.5 (2.9)                       | 8.6 (2.5)                            | -0.1                             | -0.2                                   | -0.9, 0.6           | -                      |
| Digit vigilance (s)                            | 218 (49)                        | 218 (55)                             | 0                                | -9                                     | -24, 7              | +                      |
| Digit span (forward)                           | 4.2 (1.6)                       | 4.5 (2.0)                            | -0.3                             | -0.1                                   | -0.7, 0.4           | -                      |
| Santana dexterity (both hands)                 | 53.02 (7.9)                     | 54.7 (8.5)                           | -1.7                             | -1.3                                   | -3.7, 1.2           | -                      |
| Pursuit aiming II (no. correct dots)           | 60.8 (14.1)                     | 63.3 (16.4)                          | -2.5                             | -5.3                                   | -14.2, 3.6          | -                      |
| Finger tapping (both hands)                    | 92.1 (14.4)                     | 94.0 (13.2)                          | -1.9                             | -2.4                                   | -6.4, 1.7           | -                      |
| Mean simple reaction time (ms)                 | 314 (78)                        | 301 (65)                             | 13                               | 21*                                    | -1, 43              | -                      |
| Digit-symbol                                   | 28.3 (8.7)                      | 30.8 (12.0)                          | -2.5*                            | -2.7†                                  | -5.3, -0.1          | -                      |
| Trails—A (s)                                   | 82.3 (34.31)                    | 75.5 (31.5)                          | 9.8                              | 7.1                                    | -2.4, 16.7          | -                      |
| Block design                                   | 14.1 (8.0)                      | 15.3 (7.8)                           | -1.2                             | -1.5                                   | -3.6, 0.7           | -                      |
| Vocabulary                                     | 26.2 (12.9)                     | 29.2 (16.5)                          | -3.0                             | -3.1                                   | -7.4, 1.2           | -                      |
| Questionnaire—16 (no. yes<br>answers)          | 9.1 (4.2)                       | 6.5 (3.8)                            | 2.6‡                             | 1.7‡                                   | 0.5, 3.0            | -                      |
| Brief symptom inventory<br>(grand total)       | 45.8 (25.3)                     | 26.8 (40.7)                          | 19.0‡                            | 15.7‡                                  | 5.5, 25.8           | -                      |

<sup>a</sup>The poisoned subjects performed worse on all tests, except digit vigilance, where no difference was observed.

<sup>b</sup>Adjusted for age, education, long-term exposure to cholinesterase inhibitors, alcohol intake, solvent exposure, loss of consciousness, examiner, time of day of testing, and plasma cholinesterase levels.

<sup>c</sup>CI: Confidence interval.

<sup>d</sup>(-) The poisoned subjects performed worse; (+) the poisoned subjects performed better.

\* $p < 0.10$ ; † $p < 0.05$ ; ‡ $p < 0.01$

## RESULTS

The poisoned group as a whole performed less well than did the non-poisoned comparison group on 13 of the 14 neurobehavioral tests (Table 4), but the differences on most tests were small. The largest deficits among the poisoned subjects were observed for tests of psychomotor and visuomotor functions, language, and affect. The poisoned subjects performed significantly less well on the Digit-symbol test (beta = -2.7, 95% CI -5.3, -0.1) and, in addition, reported significantly more neuropsychiatric symptoms than did the non-poisoned workers on the Questionnaire-16 (beta = 1.7, 95% CI 0.5, 3.0) as well as the BSI (beta = 15.7, 95% CI 5.5, 25.8).

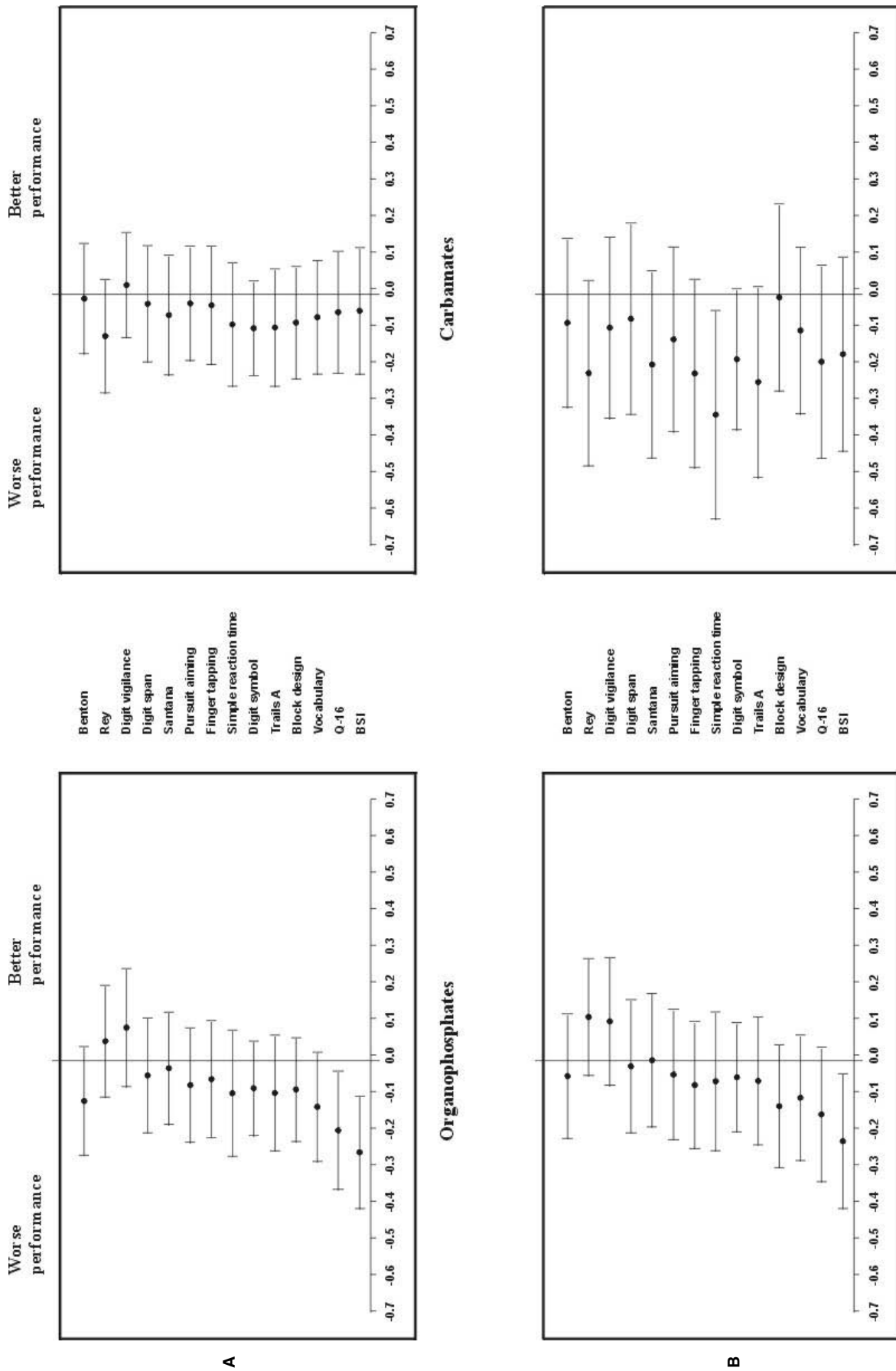
Figure 1a shows the adjusted standardized coefficients after stratification into poisonings by organophosphate and carbamate pesticides. The deficits occurred among both the organophosphate- and the carbamate-poisoned subjects, but more clearly among the organophosphate-poisoned subjects. Neuropsychiatric symptoms on the Questionnaire-16 and the BSI were significantly increased among the organophosphate-poisoned subjects, but the excesses among the carbamate-poisoned subjects were small and not statistically significant. Figure 1b shows the results after stratification into subsets with and without handling of cholinesterase-inhibiting pesticides within three months before testing. Deficits for the poisoned sub-

jects were observed in both subsets, but were more apparent in the subset of workers who had been in recent contact with cholinesterase-inhibiting pesticides. However, the excess reporting of neuropsychiatric symptoms by poisoned workers was similar for those with and without recent exposure to cholinesterase inhibitors.

The analyses restricted to the subgroup of the reference workers who had never experienced poisoning symptoms in connection with pesticides yielded results similar to those for all controls in the models. (Results are not shown.)

## DISCUSSION

Workers previously poisoned with cholinesterase-inhibiting pesticides tended to perform less well on psychomotor and visuomotor tests as compared with a non-poisoned control group. These, mainly nonsignificant, differences were observed in the organophosphate-poisoned subgroup, but small deficits in performance were also observed for the carbamate- (mostly carbofuran-) poisoned subjects. A marked increase in neuropsychiatric symptoms was observed in the organophosphate-poisoned workers. Neurobehavioral deficits of previously poisoned workers, as compared with non-poisoned workers, were more apparent within the subset who had worked with cholinesterase inhibitors within the three months before testing.



**Exposure to cholinesterase inhibitors in last three months**

**No recent exposure to cholinesterase inhibitors**

Figure 1—Neurobehavioral performances of previously poisoned banana plantation workers as compared with those of non-poisoned workers, stratified (A) by type of cholinesterase inhibitor and (B) by recent contact with cholinesterase inhibitors. Standardized regression coefficients with 95% confidence intervals adjusted for age, education, long-term exposure to cholinesterase inhibitors, alcohol intake, solvents, loss of consciousness, examiner, time of day of testing, and plasma cholinesterase levels.

Overall, these results are consistent with growing literature that suggests that poisoning with organophosphate insecticides causes persistent damage to the nervous system.<sup>5-14,31</sup> The diffuse profile of deficits that we observed is in accordance with the findings of other studies.<sup>8-10</sup> Also, in these three previous studies significant deficits on the Digit-symbol test were observed. This study was designed to be comparable to one of these studies, of Nicaraguan agricultural workers who had been hospitalized because of organophosphate poisoning.<sup>9</sup> Our study of less severely poisoned patients (not requiring hospitalization) demonstrated an overall pattern of neurobehavioral effects similar to that in the Nicaraguan study, but the deficits were smaller. Steenland et al. also found evidence of increasing neuropsychological impairment with increasing severity of organophosphate poisoning.<sup>10</sup>

This is the first epidemiologic study that examined neurologic effects from carbamate poisoning, though the number of cases (24) was small. The deficits were small and the confidence intervals wide, but chronic neurotoxic effects, suggested by previous case reports,<sup>11,14</sup> cannot be excluded. Further epidemiologic and experimental data are needed, especially for carbofuran, which accounted for all but one of the carbamate poisonings in our data.

The deficits in the performances of poisoned subjects in the subset with contact with cholinesterase inhibitors within the three months before testing were considerably larger than were those in the subset without recent exposure to cholinesterase-inhibiting pesticides (see Figure 1b). Within this subset with recent contact with cholinesterase inhibitors, the previously poisoned workers had a lower mean value of plasma cholinesterase activity than did the controls (2.05 U/mL versus 2.20 U/mL,  $p = 0.12$ ), which may indicate higher recent exposures among the poisoned subjects. However, the results were adjusted for plasma cholinesterase levels and, in addition, no difference was found for red blood cell cholinesterase activity, which is a better marker of acute effects on the nervous system.<sup>4</sup> Therefore, even if there were an additional effect from recent exposure, the observed deficits cannot be explained by recent exposure.

Several possible sources of bias could have influenced the results. For 25% of the poisoned workers, the medical record could not be obtained to confirm the clinical diagnosis of poisoning with a cholinesterase inhibitor. However, the diagnosis was confirmed for all 61 workers for whom medical records were located. Exclusion from the analyses of unconfirmed poisonings did not produce appreciable changes in the effect estimates. Medical doctors in banana plantation areas are unlikely to misdiagnose pesticide poisoning, which is common and has characteristic clinical features. Therefore, misdiagnosis of pesticide poisoning is an unlikely source of bias.

Banana plantation workers are a distinctive social group, unlike other agricultural or industrial worker populations. The random selection of the controls from the same population guaranteed identical socioeconomic and occupational backgrounds, which indirectly controls for a number of important potential confounders. As a result, however, pesticide exposures in the comparison group were substantial. Some controls had experienced pesticide-related symptoms, albeit not requiring medical attention. The mean number of symptoms was considerably smaller than among the poisoned who had sought medical attention (7 versus 14), and the symptoms were mostly nonspecific (headache, nausea, lightheadedness, weakness, vomiting), but an overlap of this subgroup of comparison workers with the poisoned group is conceivable. However, when restricting the analysis to the subset of controls never having experienced symptoms, the differences between poisoned and non-poisoned workers did not increase, as would be anticipated if such bias existed.

Forty-two percent of the identified cases of poisonings could not be located. There were no objective data for comparison of located and non-located workers, which might have made it possible to evaluate the direction of this potential selection bias. It is likely that most of those persons still working on banana plantations or living in the region were located. However, the most affected workers may have been more likely to leave the banana region due to potential intolerance of previously poisoned subjects to subsequent pesticide exposures.<sup>6,7,32</sup> This might have led to the examination of a healthy subset of the previously poisoned group, minimizing differences between poisoned workers and controls.

More non-poisoned than poisoned workers had received some high school education. In addition, the lower scores on the vocabulary test for the poisoned group could indicate lower premorbid intellectual aptitude of the poisoned subjects.<sup>28</sup> However, it has been shown that vocabulary test scores are sensitive to solvent-induced central nervous system effects.<sup>33,34</sup> Also, two of the three major epi-studies of organophosphate poisonings found considerable deficits of the poisoned subjects on the Wechsler vocabulary test,<sup>8,9</sup> while the third did not include a language test.<sup>10</sup> In any case, adding the vocabulary test score to the model in the multivariate analyses did not substantially change the coefficients of the poisoning status after controlling for education.

In practice, the cleverest workers often are selected for dangerous tasks such as nematocide spraying and, thus, may be more likely to get poisoned. To evaluate this possibility, we compared the performances of controls with recent exposure to cholinesterase inhibitors (who were mostly nematocide applicators) with those of controls without recent exposure to cholinesterase inhibitors. The recently exposed did considerably

better on all cognitive tests. Thus, even though the controls were somewhat better educated, it seems unlikely that they had greater cognitive ability, because only relatively few controls were nematocide sprayers versus a majority of the poisoned subjects.

The size of our total study population was sufficient to detect a 10% difference in performances with good precision on most of the tests,<sup>35</sup> but the numbers of exposed subjects in the subgroups became small. In spite of the wide confidence intervals, the point estimates of the effects were, in general, consistent over the various categories (see Figure 1), which supports the validity of the observed results.

In conclusion, this study lends additional support to previous findings that organophosphate poisonings have adverse chronic effects on a broad spectrum of nervous system functions. We observed such effects among workers with only mild poisonings. We could not exclude the possibility of persistent neurobehavioral effects associated with poisonings by n-methyl carbamate insecticides. Workers who have histories of poisoning may be more susceptible to neurobehavioral effects with subsequent exposures.

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