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REPORT TO THE NORTH CAROLINA PRIMARY HEALTH CARE ASSOCIATION: IMPROVED DIAGNOSIS AND PREVENTION OF PESTICIDE RELATED ILLNESS AMONG NORTH CAROLINA MIGRANT FARMWORKERS

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INTRODUCTION: CHOLINESTERASE INHIBITING PESTICIDES AND THE HEALTH OF FARMWORKERS

Migrant farmworkers have the worse health of any occupational group in the nation (1). Pesticide exposure is a major source of environmental risk for the 2.5-5 million farmworkers in the US and the 80,000 in North Carolina (1,2,3,4). However, both the reduction of exposure and the management of those exposed are hampered by inadequate epidemiologic data, limited understanding of the effects of chronic exposure, and the difficulty of incorporating existing information into migrant health services. The lack of data is a major obstacle in obtaining funding and in designing effective disease prevention programs (5). Due to the non-specific nature of chronic pesticide exposure symptoms, patients are treated symptomatically; no simple, inexpensive technique is in place to diagnose pesticide exposure. The degree of occupational risk associated with pesticides is poorly defined, and under-diagnosis is the norm (6,7). In addition, the transience of migrant farmwork complicates follow-up and makes it especially important that pesticide exposure can be quickly and inexpensively assayed in migrant health clinics. Of equal importance is that the provision of specific information about pesticide exposure could be made available to patients to reduce future exposure.

LITERATURE REVIEW

Organophosphate (OP) pesticides are irreversible inhibitors and carbamates reversible inhibitors of acetylcholinesterase (AchE), the enzyme which hydrolyses the neurotransmitter acetylcholine (Ach) (11,12). Major exposures inhibit AchE almost completely, causing initial stimulation and eventual blockage of cholinergic transmission affecting the parasympathetic nervous system primarily (11,12,13). Acute exposure may cause bronchospasm and breathing impairment, blurred vision, muscle fasciculations, excessive sweating and lacrimation, increased blood pressure, contact dermatitis, ataxia, nausea/abdominal pain, diarrhea, hyperglycemia and glycosuria (1,11,13). Chronic exposure has been hypothesized to result in a variety of neurologic and cognitive deficits (14,15). Long term exposure has been implicated in several types of cancer, teratogenic effects, sterility, spontaneous abortion, and pesticide-induced delayed neuropathy (16).

Most of the pesticides (insecticides, herbicides, fungicides, fumigants, etc) used in agriculture are organophosphates or carbamates applied to crops picked by migrant farmworkers: 800 million of the one billion pounds produced annually are applied to the 20% of acreage harvested by farmworkers (6). Virtually all exposure is through dermal contact (2). OP residues have been detected in migrant camps (17) on gloves used by farmworkers (18), and a small number of studies have reported depressed cholinesterase levels in farmworkers

(19,20,21,22,23). Numerous episodes of acute poisoning, sometimes resulting in death, have been reported (7). In November 1989, 81 farmworkers in Florida were treated for acute poisoning with the pesticide Mevinphos (24). The lack of availability of hand washing water in the fields (25,26) suggests that ingestion of pesticides also occurs while eating or smoking. An additional source of risk for farmworkers results from the fact that dehydration and poor nutrition predispose to increased toxic effects (1).

This lack of data about chronic exposure and its effects is of greatest significance for farmworkers, whose entire career consists of chronic exposure to OP pesticides. The studies which have found depressed cholinesterase levels (19,20,21,22,23) have employed extremely small samples (sometimes as few as seven subjects), none have used an adequate control population, and little or no information has been obtained about specific sources of occupational exposure (non-compliance with re-entry intervals, use of protective clothing, availability of hand washing water, etc). Thus, it is not surprising that previous findings are inconclusive with regard to the health problems caused by chronic OP pesticide exposure. The inability to determine the health effects of chronic exposure have resulted partly from the small sample sizes of these studies but also from the fact that asymptomatic working individuals were targeted. In addition, several of these studies have targeted groups of agricultural workers at less risk for exposure, such as packing house workers

(22,23). Moreover, the assays employed in these studies could not be routinely used by migrant clinics. Finally, these studies have made little contribution to prevention of exposure.

The project described here had two primary goals:

1) execute an epidemiologic study of the health effects of chronic exposure and sources of occupational risk among symptomatic farmworkers and non-farmworkers;

2) test new technology in the context of migrant health delivery to improve clinical diagnosis of pesticide exposure.

REFERENCES

1. Wilk VA. The Occupational Health of Migrant and Seasonal Farmworkers in the United States. Farmworker Justice Fund, Washington DC, 1986.

2. Moses M. Pesticide-Related Health Problems and Farmworkers. AAOHN Journal 37(3):115-130, 1989.

3. Lillesand D, Dravitz L, and McClellan J. An Estimate of the Number of Migrant and Seasonal Farmworkers in the United States and the Commonwealth of Puerto Rico. Legal Services Corporation. Washington, DC, 1977.

4. Fleming MF and CS Hayak. Health care planning in North Carolina: migrant workers program. NC Med J, 45(6):371-74, 1984.

5. Reig S. Director, Office of Migrant Health. Paper presented at the National Migrant Health Conference, Hollywood Florida, April 1988.

6. Coye MJ. The health effects of agricultural production: I. the health of agricultural workers. J Public Health Policy 6(3):349-370, 1985.

7. Kahn E. Pesticide related illness in California farm workers. J of Occup Med 18(10):693-696, 1976.

8. McWilson W, Ruebush TK, Jobbs JH, Hippolyte R, Miller S. Safety measures associated with organophosphate insecticides in the Haitian malaria control programme. Bull WHO 63(2):345-351, 1985.

9. Brodgen WG. Laboratory and field microassay of cholinesterases in whole blood, plasma, and erythrocytes. Bull WHO 65(2):227-232, 1987.

10. McConnell R and Magnotti R. Screening for organophosphate insecticide overexposure under primitive conditions. Accepted for publication by AJPH.

11. Namba T. Cholinesterase inhibition by organophosphorus compounds and its clinical effect. Bull WHO 44:289-307, 1971.

12. Wills JH. The measurement and significance of changes in the cholinesterase activities of erythrocytes and plasma in man and animals. CRC Critical Reviews in Toxicology. Vol. 1, 153-202, March 1972.

13. Midtling JE, Barnett PG, Coye MJ, Velasco AR, Romero P, Clements CL, et al. Clinical management of organophosphate poisoning. West J Med 142:514-518, 1985.

14. Rodnitzky RL, Levin HS, Mick DL. Occupational exposure to organophosphate pesticides: a neurobehavioral study. Arch Env Health 30:98-103, 1975.

15. Moses M. Cancer in humans and potential occupational and environmental exposure to pesticides. AAOHN Journal 37(3):131-136, 1989.

16. Goldsmith MF. As farmworkers help keep America healthy, illness may be their harvest. JAMA 261(22):3207-3213.

17. Morse DL, McLellan R, Christopherson C. Potential pesticide exposure of migrant farmworkers living within spray areas. J Env Health, 44(6):301-4, 1982.

18. Munn S, Keefe TJ, Savage EP. A comparative study of pesticide exposures in adults and youth migrant field workers. Arch Env Health 40(4):215-220, 1985.

19. Bogden JD, Quinones MA, El Nakah A. Pesticide exposure among migrant workers in southern New Jersey. Bull Env Cont & Tox 13(5):513-517, 1975.

20. Quinones MA, Bogden JD, Louria DB, El Nakah A. Depressed cholinesterase activities among farmworkers in New Jersey. The Science of the Total Environment 6:155-59, 1976.

21. Richards DM, Kraus JF, Kurtz P, Borhani NO. A controlled field trial of physiological responses to organophosphate residues in farm workers. J Env Path & Tox 2:493-512, 1983.

22. Wicker GW, Williams WA, Guthrie FE. Exposure of field workers to organophosphorus insecticides: sweet corn and peaches. Arch Env Cont and Tox 8:175-182, 1975.

23. Wicker GW, Williams WA, Bradley JR, Guthrie FE. Exposure of field workers to organophosphorus insecticides: cotton. Arch Env Cont and Tox 8:433-440, 1975.

24. Ryder RE. Florida clinic handles massive pesticide poisoning. Migrant Health Newsline 6(6):3-4, 1989.

25. Ciesielski SD and Seed JR. Prevalence of intestinal parasites in North Carolina migrant farmworkers. Migrant Health Newsline, 1988.

26. Ciesielski SD, Seed JR, Ortiz JC. The epidemiology of intestinal parasites of North Carolina migrant farmworkers: results of a two-year study. Submitted for publication to AJPH.

27. Poli B and Fluker S. Pesticide Safety for Farmworkers. EPA, Washington DC, 1985.

28. Eng E, Saldias G, Sarri C, Ciesielski SD. Perceptions of environmental risks in living and working conditions among Hispanic migrant farm workers. Presented at American Public Health Association conference, October 1989.

PART I: USE OF THE EQM FIELD COLORIMETER TO MEASURE CHOLINESTERASE LEVELS OF MIGRANT FARMWORKERS

A. BACKGROUND

The measurement of erythrocyte acetylcholinesterase or serum cholinesterase in the diagnosis of organophosphate (OP) and to a lesser extent carbamate pesticide poisoning or exposure is fairly standard. With regard specifically to migrant farmwork, only California has worker protection regulations based upon cholinesterase testing, requiring testing of applicators and investigation when an individual's cholinesterase level is below 30% of baseline and removal from work if the level is below 50% of baseline.[1] Reference lab testing is at present universally used in the United States; the cost per test in North Carolina ranges from $32.00 to $48.00. In developing countries the high cost of such testing, lack of availability, and logistical problems related to transport of samples has encouraged the development of simpler field methods for measuring cholinesterase. Field methods are widely used in developing countries, and include spectrophotometric analysis, tintometric methods, and use of colorimeters. There are at present problems with field use of both spectrophotometry and tintometry.[2] The WHO recommends that field measurement of OP exposure employ colorimetric measurement of RBC acetylcholinesterase.[3] Seven criteria have been offered[1,2] for reliable field measurement of cholinesterase:

1. accuracy in measurement of RBC/plasma cholinesterase
2. affordability
3. precision
4. stability of reagents, especially with regard to temperature
5. operation independent of equipment such as centrifuges or line voltage
6. portability
7. rapid determination of cholinesterase levels

The "EQM Cholinesterase Assay Kit", based on a colorimetric method, has undergone extensive field and laboratory testing. This assay was also employed in a recent major study of OP pesticide exposure and illness in Nicaragua.[4,5] The second generation model of the assay was lent to the North Carolina Primary Health Care Association for the purpose of conducting a study of illness resulting from chronic exposure to OP pesticides and also for the purpose of determining the feasibility of diagnosis of pesticide related illness in migrant health centers by means of this assay.

B. DESCRIPTION OF THE "EQM CHOLINESTERASE ASSAY KIT"

A common means of measuring the amount of an enzyme present in a sample is to measure its activity. The action of acetylcholinesterase _in vivo_ is to hydrolyze its substrate, choline ester, producing choline and carboxylic acid. The EQM assay system uses thiocholine ester as substrate, and the thiocholine released reacts with another reagent (5,5'-dithiobis(2-nitrobenzoic acid) resulting in a yellow color. The amount of light which passes through the sample is measured by the colorimeter, greater amounts of enzyme resulting in more intense color and less absorbance. The raw absorbance is converted by standard equations into the units of enzyme activity present in a sample.

The performance of the EQM colorimeter assay system can be evaluated in terms of the 7 criteria listed above, based upon our field experience of approximately six months and over three hundred testing experiences.

1. _Accuracy in measurement of RBC/plasma cholinesterase_. An important feature of the EQM colorimeter is the correction for hemoglobin. Since the concentration of erythrocytes varies between and within individuals, being affected by nutrition, hemolytic conditions, various parasites, and numerous other factors, measurement of acetylcholinesterase contained by erythrocytes ideally corrects for this variation. Correction for hemoglobin content by the EQM colorimetric method is reported to eliminate more than half of the variability of activity.[2] Through correction for hemoglobin values, the EQM colorimetric assay has been found to identify cases of poisoning missed by the tintometric method.[2]

Since the EQM colorimetric assay is a kinetic assay, it is subject to influence by ambient temperature. Enzymatic activity, including that of acetylcholinesterase, is increased at higher temperature. Results of the EQM colorimetric assay are corrected for temperature, eliminating this source of potential inaccuracy.

2. Affordability. The EQM Cholinesterase Assay used during this project included the Model 176 LED source colorimeter, purchase price $995.00 (including reagents and some materials). Cost per assay is estimated by the company at $0.41. This cost includes the initial purchase of the assay and also material not included in the assay (cotton swabs, lancets, etc.). It is not possible to provide another estimate based upon our experience, since the assay was loaned and other materials were donated. Our experience may not have approximated the above figure exactly, it was clear that on a per test basis the cost was extremely small, certainly less than $1.00.

3. Precision Laboratory and field studies have found that the precision of the EQM assay is comparable to that of reference laboratories in measurement of RBC cholinesterase.[2] Some features, discussed above, which increase the accuracy of measurement (correction for hemoglobin and temperature) also increase the precision. If cholinesterase measurements are to be obtained in migrant camps the adjustment for temperature becomes more important.

Unlike erythrocyte acetylcholinesterase, plasma cholinesterase is affected by numerous factors other than OP or carbamate pesticides. In addition, there is greater interpersonal and intrapersonal variation in plasma cholinesterase levels than is found with erythrocyte acetylcholinesterase. Thus, erythrocyte acetylcholinesterase is the analyte of choice for the assessment of exposure to or illness from cholinesterase inhibiting pesticides. The coefficient of variation by use of the EQM colorimetric method is less than ten percent.

4. Stability of reagents, especially with regard to temperature. We were unable to make any determination regarding stability of reagents in any formal manner. However, there was no instability observed over time.

5. Operation independent of equipment such as centrifuges or line voltage. The assay obtained from EQM clearly fulfilled this criterion. It functioned as a self-contained, easily transported unit, without the need for additional laboratory equipment or materials. Only common and inexpensive items were required in addition to those which are included in the assay kit itself.

6. Portability. The second generation model we used came with a small carrying case; the colorimeter and some supplies could fit in this. It was usually necessary to carry another box for glass bottles, bleach, gloves, and other supplies that could not fit in the carrying case. While is was somewhat cumbersome to move these supplies, this would not be an important consideration for clinical use of the colorimeter. In any case the new model is smaller and requires less additional gear.

7. Rapid determination of cholinesterase levels. Other studies have found that 10 determinations can be made per hour.

We made fewer, primarily because we administered a lengthy questionnaire at the same time, and also because our research site was within an adjacent building rather than within the clinic. We expect that a determination could be made within clinics in about five minutes. Again, the new model colorimeter is much easier to use and requires fewer procedures, which will reduce the time of determination.

To these seven criteria should be added another, on the basis of our experience with the system:

8. Mechanical reliability of the system. Extensive trials have demonstrated that the principles upon which the EQM assay is based are sound.[2,4,5] However, technical malfunctions occurred during our use of the colorimeter. These malfunctions were not related to the principle of the system itself, but rather to secondary mechanical hardware. Since field workers obtained their own cholinesterase values each day prior to testing subjects, technical problems were easily detected and were in general unable to introduce error. The two malfunctions which led to the greatest delay in progress of the project were fractures in the filter mountings of the colorimeter and malfunction of the toggle switches which served as control devices for the colorimeter. These and other malfunctions were repaired free of charge by EQM, but did result in some delay, without which the projected sample size of 400 rather than the actual final sample size of 244 would probably have been achieved.

C. THE USE OF THE EQM COLORIMETER IN MIGRANT HEALTH CENTERS

Had they not been corrected, these malfunctions would obviously raise serious questions about the feasibility and advisability of implementation of the EQM colorimeter for the diagnosis of pesticide related illness in migrant health centers. However, on each occasion in which a problem emerged, the personnel at EQM investigated thoroughly the circumstances and cause of malfunction. As mentioned above, the model used for this project was the second generation model. Since the primary market for a field assay is in developing countries, affordability is a fundamental consideration. We were one of the first to use the second generation model, which differed from the first generation model in several aspects of a secondary mechanical nature and which had been implemented to reduce the price of the assay. Partly as a result of our experience with the assay, changes have been made and the third generation model should correct the problems that we encountered.

In addition to correcting on a design level every problem that we encountered, other changes have been made which will make the third generation model colorimeter more feasible for clinical use. While the second generation model was not a complicated laboratory procedure at all, the number of steps, the exactness required, and the computations necessary might introduce measurement error or discourage busy clinicians from conducting the assay. The new model protects against measurement error by using a larger aliquot of blood, internal correction for temperature and hemoglobin eliminating the need for computation, and reduces the number of steps in the procedure. These changes make the clinical use of the colorimeter feasible, and such implementation advisable with regard to technical reliability.

Additional features have been added to the third generation model which are not as relevant to clinical diagnosis per se, but increase the possible scope of investigation of pesticide related illness. The new model allows determination of the presence of OP or carbamate pesticides in water or soil. Migrant health clinics with strong outreach programs and experience in public health activities could use these features both to identify the source of contamination, to prevent further exposure, and to add to the body of knowledge about pesticide exposures among farmworkers.

It can be safely stated that, more than any other single issue, pesticides occupy a dominant place in the field of migrant health at the present time. A review of conference topics from several regional and national meetings shows that more presentations are made concerning pesticides than any other single topic. Nearly all health education programs directed at farmworkers devote a substantial portion or even the entirety of their content to pesticide related issues. It is equally evident that pesticides have become the focus for advocacy groups, whether legal or public health oriented. Thus, it is clear that a considerable portion of migrant health funding, from whatever source, is devoted to pesticide related issues.

This being the case, it would likely appear paradoxical to an outside observer, or to those critical of this activity, that a case of pesticide related illness has never been diagnosed at the Tri-County Community Health Center (TCCHC) in Newton Grove, NC (at least physicians working there for the previous five years have no recollection of such a case). The considerable difficulties encountered in the diagnosis of pesticide related illness, especially those resulting from chronic exposures, are described more fully in the Introduction. However, it seems to the authors of this report that even taking these difficulties into account, the activities which transpire on the level of advocacy and health education have not been met with any remotely proportionate efforts in the critical field of the diagnosis of pesticide related illness itself. It is apparent that migrant health as a field is committed to the concept of pesticide related illness, for which the evidence on the level of acute poisoning is strong and unequivocal. However, an equal commitment to improving the diagnosis of chronic pesticide related illness has not been apparent. In large part this absence of activity is related to the difficulties in diagnosis, a difficulty which has in considerable measure derived from the inability to feasibly measure the only biological marker of exposure, cholinesterase, in a clinical setting. However, the recent technological advances in field cholinesterase measurement, such as the EQM Cholinesterase Assay, solve almost entirely the prior logistical, financial and scientific constraints on the measurement of cholinesterase in a clinical setting. This being the case, migrant health centers must not only make use of technological advance, for technology only provides a part of the solution, but begin to improve the clinical protocols for the diagnosis of pesticide related illness.

It is now financially feasible for every migrant health center in the United States to possess the technology to measure cholinesterase and routinely check workers. Before the diagnosis of pesticide related illness can become a routine part of migrant health care delivery, a substantial body of clinical experience must be accumulated. Until cholinesterase measurement is routinely conducted in migrant health centers, it will be impossible to rectify the present inadequate state of clinical capability in this regard.

In the second part of this report the results of the study of cholinesterase and their relation to occupational factors and morbidity is discussed more fully. These results are preliminary, and additional analysis is necessary. However, even on the basis of these preliminary results some conclusions can be drawn of relevance to the use of cholinesterase measurement in a clinical setting. The study described in Part II is the largest and most thorough study of the health effects of chronic OP exposure among farmworkers, but the results to date conform well with those of prior studies. We demonstrated that farmworkers exposed to OP pesticides have acetylcholinesterase levels significantly lower than non-farmworkers, as reported by other studies. However, preliminary analysis has not, and it appears unlikely that additional analysis will demonstrate a statistical association between low cholinesterase levels and reported symptoms or reported exposures. There are two possibilities for the absence of such associations: that chronic exposures do not result in morbidity, or that the large range of normal cholinesterase values precludes the demonstration of such effects in a cross sectional study obtaining a single measurement from subjects. Neither possibility can be confirmed or confuted on the basis of the data available, and a fuller discussion is found in Part II. However, that farmworkers as a group were demonstrated to have depressed cholinesterase levels, and that depression of cholinesterase at some point (varying by individual and perhaps impossible to specify without baseline measurements) results in morbidity is certain.

The obvious implication of the above is that a single cholinesterase measurement in a clinical setting will be of little diagnostic utility for confirming illness resulting from <u>chronic</u> exposure. However, the corollary of this proposition is not, as is sometimes suggested, that <u>baseline</u> measurements are necessary to make cholinesterase testing useful. For obvious reasons, obtaining baseline measurements on farmworkers will be impossible under present or any likely future conditions. There is no simple or uncomplicated solution, unfortunately, to the biologic constraints which exist. However, if baseline measurements are not possible, it is possible to conduct followup testing of patients after the initial measurement conducted in conjunction with assessment of symptomaticity and other clinical procedures.

In this regard the portability of the EQM Colorimeter becomes important. The colorimeter can be powered by battery, making rapid field measurement in migrant camps feasible. In a clinic such as the TCCHC in Newton Grove, the presence of an outreach staff makes possible routine followup of presumptive cases of pesticide related illness. The testing procedure itself is extremely simple and does not require prior lab or clinic experience. Ideally one outreach worker would be assigned the task of followup measurements to minimize variation in measurement.

Changes in cholinesterase measurement and changes in symptomaticity observed in followup can be used to help confirm presumptive pesticide related illness. If cholinesterase levels increase and stabilize after the initial clinical encounter, it would be possible to retroactively determine the patient's baseline. The length of followup required would vary, and there are obvious logistical considerations which will influence protocols. However, a feasible and reliable protocol for the diagnosis of pesticide related illness in a clinical setting, based upon initial cholinesterase measurement and examination as well as followup cholinesterase measurement, can only be formulated on the basis of experience. When migrant health centers acquire the technology upon which to base the initial attempts at such a protocol, the diagnosis of illness resulting from chronic exposureto OP pesticides can become a routine clinical entity, the health of farmworkers will be protected in this regard, and the limited funds for migrant health can be allocated in a rational manner.

REFERENCES

1. Medical supervision of pesticide workers:guidelines for physicians. Berkeley, CA. State of California Department of Health. Epidemiological Studies Laboratory, 1974;AFC 59-122.

2. Magnotti RA, Dowling K, Eberly JP, McConnell RS. Field measurement of plasma and erythrocyte cholinesterases. Clinica Chimica Acta 1988;315:315-332.

3. World Health Organization. Technical report series No. 356 by Expert Committee on the safe use of pesticides in public health. Geneva: WHO, 18\967;15.

4. McConnell R, Pacheco FA, Magnotti R. Cropduster aviation mechanics: high risk for pesticide poisoning. Am J Public Health 1990:80(10):1236-39.

5. Cole D, McConnell R, Murray DL, Pacheco FA. Pesticide illness surveillance; the Nicaraguan experience. PAHO Bulletin 1988;22(2):119-130.

PART II. RESULTS OF CROSS SECTIONAL STUDY OF ERYTHROCYTE CHOLINESTERASE LEVELS AND THEIR ASSOCIATIONS WITH REPORTED SYMPTOMS AND OCCUPATIONAL EXPOSURES

INTRODUCTION

Pesticide related illness has become a major source of concern in the migrant health field. Attention has focused on cholinesterase inhibiting pesticides such as organophosphates and carbamates. The effects of acute exposure are well known and the identification of such effects, while probably underestimating their frequency, is fairly reliable. However, although chronic exposures to these pesticides is far more common, there is little known about the nature or frequency of adverse health effects resulting from chronic exposure. The cross-sectional study described below attempted to describe the health effects of such exposure among migrant farmworker patients at two health clinics in North Carolina.

METHODS

Subject Selection

The study population consisted of a randomly selected group of walk-in patients (migrant farmworkers) at the Tri-County Community Health Center in Newton Grove NC and Goshen Medical Center in Faison NC during the harvest season of 1990. An unexposed cohort of non-farmworkers was randomly selected from patients at these same clinics. Informed consent was obtained from subjects in their preferred language.

Field Cholinesterase Testing

The cholinesterase assay is described in Part I. Blood obtained by finger prick (5 microliters), and hemoglobin was measured using the colorimeter. Activity of cholinesterase in the subject's blood sample as measured by absorbance was corrected for ambient temperature and hemoglobin.

Clinical Data From Patient Records

Clinical data was obtained from patient records for study subjects at both the Tri-County Community Health Center and the Goshen Medical Center. Data obtained consisted primarily of temperature and blood pressure. Serum and blood glucose were obtained where available.

Questionnaires

Pre-tested questionnaires were administered to subjects in their preferred language. Data was obtained relating to demographic variables, all relevant clinical manifestations (using recognition rather than recall strategies), work history, pesticide exposure, and a variety of occupational factors. The latter category included employer compliance with re-entry intervals, subjects' use of protective clothing, availability of hand washing water in the fields, etc.

RESULTS

A total of 244 subjects (202 farmworkers and 42 non-farmworkers) participated in the study, whose field component lasted from 6/25/90 - 11/15/90. The demographics of farmworker and non-farmworker subjects are found in Table 1. The two groups were fairly similar with regard to gender, ethnicity, and age.

The symptoms which may result from exposure to cholinesterase inhibiting pesticides are listed in Table 2, with the frequency of their occurence among farmworkers and non-farmworkers. In most instances such symptoms had a much higher prevalence among farmworkers.

A high frequency of exposure to pesticides was reported by subjects (Tables 3 and 4). Exposures may be divided into those deriving directly from occupational conditions, such as being sprayed directly or working in fields in which a chemical smell is present. A surprisingly high percentage of farmworkers (46%) reported being sprayed directly or working immediately adjacent to rig spraying within one month prior to study enrollment. Moreover, one-third of such subjects reported that illness resulted from such exposure. Fifty percent of subjects reported working in fields in which a chemical smell was apparent within one month of enrollment. Hand washing water was not always available for 58% of subjects.

Reported exposure deriving from individual behavior was also frequent in some cases (Table 4), although in general the actions completely under the control of the worker were more likely to have a higher percentage of protective behaviors. Protective actions that depend upon occupational conditions, such as hand washing water, were less often reported than those under the workers control, such as wearing shoes and long pants.

The mean cholinesterase level among the farmworker group was 30.28 vs 32.3 among the comparison group (Table 5), a statistically significant difference ($p = .013$, T-test). Although only a small percentage (5%) of farmworker subjects reported mixing, handling, or applying pesticides themselves, these subjects had cholinesterase values that were significantly below those of other farmworker subjects.

At this stage of analysis no associations have been demonstrated between cholinesterase levels and symptoms. Examples of cholinesterase values and their relation to symptoms are found in Table 6.

DISCUSSION

The results presented above are preliminary and incomplete. However, it is possible to reach several conclusions at this stage. It is evident that the symptoms which can result from pesticide exposure are quite frequent among farmworkers, and in most cases are more frequent than among non-farmworkers. However, since many other factors intrinsic to farmwork, such as dehydration, dust, contact dermatitis, etc, can produce such symptoms there is little that can be inferred about their etiology. It is also clear that exposures to pesticides are frequent in farmwork. Approximately half of the farmworker subjects reported substantial exposure within the previous month, in the form of direct aerial spraying or indirect rig spraying. That such exposures were indeed substantial is indicated by the fact that one third of those with such exposure experienced subsequent illness. Thus, in the sample of farmworkers as a whole 17% experienced illness they attributed to a specific pesticide exposure during a one month period. Even allowing for a large amount of misattribution it is appropriate to conclude that occupational exposures to pesticides routinely result in illness among farmworkers.

This conclusion is supported by the significant difference in the cholinesterase values of farmworkers and non-farmworkers. However, at this stage of analysis no associations have been found between specific symptoms and cholinesterase values. However, it would be premature to conclude at this stage that these results indicate that there is no association between cholinesterase levels and symptoms. The association or lack thereof will be determined by the remaining phases of analysis.

TABLE 1. DEMOGRAPHICS OF STUDY SUBJECTS

	FARMWORKERS (N = 202) %	NON-FARMWORKERS (N = 42) %
GENDER		
male	77	49
female	32	51
AGE (mean)		37
BIRTHPLACE		
United States	43	52
Mexico	47	36
Central America	06	12
Other	07	0
ETHNIC GROUP		
US born blacks	35	26
US born whites	02	21
Hispanic	62	50
Other	02	03

TABLE 2. FREQUENCY OF REPORTED SYMPTOMS

SYMPTOM EXPERIENCED WITHIN ONE WEEK OF STUDY	FARMWORKERS (N = 202) %	NON-FARMWORKERS (N = 42) %
HEADACHE	48	38
reporting > 2 headaches	28	24
✗ headache persisting > 2 days	42	14
FEVER	21	12
NAUSEA	27	21
STOMACH PAIN	27	31
DIARRHEA	14	05
BLURRED VISION average duration	27	17
RED/IRRITATED EYES	31	36
✗ RASH mean days rash lasted	42	05
INCREASED SALIVATION	18	05
ABNORMAL INCREASED SWEATING	32	12
MUSCULAR SYMPTOMS[1]	42	33
✗ COORDINATION PROBLEMS	14	05
DIFFICULTY BREATHING[2]	20	24
CHEST PAIN[3]	16	14
DIZZINESS	30	12
IMPAIRED MEMORY	10	10
✗ INCREASED ANXIETY	27	07
PHONY VARIABLES		
INCREASED NAIL FRIABILITY	11	05
ABNORMAL TASTE PERCEPTION	18	12

[1] tremor, cramping, weakness
[2] excluding subjects with previous diagnosis of pulmonary disorder
[3] excluding subjects with previous diagnosis of heart disease

TABLE 3. PERCENT OF FARMWORKER SUBJECTS REPORTING OCCUPATIONAL EXPOSURES TO PESTICIDES

VARIABLES RELATING TO WORK CONDITIONS	
DIRECTLY SPRAYED OR WORKING IN IMMEDIATE VICINITY OF SPRAYING	46
symptoms resulting from spraying	33
WORKING IN FIELDS WITH CHEMICAL ODOR	50
MIXING OR APPLICATION OF PESTICIDES	05
HAND WASHING WATER NOT ALWAYS AVAILABLE	58

TABLE 4. PERCENT OF FARMWORKER SUBJECTS REPORTING OCCUPATIONAL EXPOSURES TO PESTICIDES[1]

VARIABLES RELATING TO INDIVIDUAL BEHAVIORS	
NOT ALWAYS WEARING:	
GLOVES	83
LONG SLEEVED SHIRT	56
SHIRT	18
LONG PANTS	17
WEARING SHOES	20
NOT ALWAYS WASHING HANDS:	
BEFORE EATING IN FIELDS	50
BEFORE SMOKING IN FIELDS[2]	96
BEFORE EXCRETORY ACTIVITY	85
NOT ALWAYS LAUNDERING CLOTHING BEFORE WEARING AGAIN	38
NOT ALWAYS WASHING HARVESTED PRODUCE BEFORE CONSUMPTION	50

[1] reported exposures within the 30 days prior to interview
[2] of those who smoke

TABLE 5. CHOLINESTERASE VALUES AMONG SUBJECTS

	FARMWORKERS (N = 202)	NON-FARMWORKERS (N = 42)	T-test p value
ALL SUBJECTS	30.28	32.3	.013

TABLE 6. CHOLINESTERASE VALUES AMONG SUBJECTS REPORTING SYMPTOMS WHICH CAN RESULT FROM ORGANOPHOSPHATE PESTICIDE EXPOSURE

SYMPTOM	+ SYMPTOM	- SYMPTOM	T-test p value
INCREASED SALIVATION	30.6	30.07	.55
DIZZINESS	29.7	30.2	.49