XI. APPENDIX III

DIAGNOSIS AND MEDICAL MANAGEMENT OF ORGANOPHOSPHORUS INTOXICATION

The text appearing immediately below is excerpted from a publication entitled "Prevention and Management of Organophosphate Poisoning." [168] This material, which was approved in 1970 by the AMA Committee on Occupational Toxicology of the Council on Occupational Health, originally appeared in the Journal of the American Medical Association in 1971.

Diagnosis

"A diagnosis of organophosphate intoxication is based primarily on a definite history of exposure to an organophosphate six hours or less before onset of illness and clinical evidence of diffuse parasympathetic stimulation. Laboratory verification is based on depression of plasma and red blood cell cholinesterase to a level substantially (50% or more) below preexposure values if these are available. If preexposure values are not available, one can use laboratory normal ranges, observing, of course, the usual caution in interpreting such figures. There are many different methods for estimation of cholinesterase content of blood, and associated with each method is a different set of normal values and a different set of reporting units. The laboratory report of a cholinesterase determination should

state the units involved along with the appropriate normal range. Based on the Michel method the normal range of red blood cell cholinesterase activity (delta pH/hr) is 0.39 to 1.02 for men and 0.34 to 1.10 for women. The normal range of the enzyme activity (delta pH/hr) of plasma is 0.44 to 1.63 for men and 0.24 to 1.54 for women.

"In actual practice, the cholinesterase test is often of more value as a confirmatory, rather than a diagnostic, procedure. For moderate to severe intoxication, the clinician should act on his clinical impression and on the history of exposure rather than wait for laboratory confirmation.

"Initial signs and symptoms of intoxication are headache, nausea, vomiting, sweating, blurred vision, weakness, diarrhea, abdominal pain, and pallor. In moderate to severe cases of intoxication, signs and symptoms may also include dyspnea, salivation, lacrimation, muscle fasciculation, convulsions, cyanosis, shock and cardiac arrhythmias, coma, and death. In the case of mild poisoning where the differential diagnosis may be puzzling, the results of the cholinesterase test may be necessary to establish a definite diagnosis.

"Cholinesterase is an enzyme which hydrolyzes acetylcholine. Two types are clinically significant: The first is true or acetyl cholinesterase, found principally in nervous tissue and in the red blood cell; the other, plasma or pseudo cholinesterase, is found in nervous tissue and in the

circulating plasma. Whereas the action of both is inhibited by organophosphate compounds, only depression of the amount of enzyme in the red blood cell is a specific response to these toxins. The level of the enzyme in the plasma may vary with a number of diseases or toxic states. A relatively wide variation exists in the normal levels of both enzymes from one individual to another as well as in the same individual at different times. Once enzyme activity is inhibited, the regeneration times differ between the two types. Red blood cell cholinesterase regenerates at approximately 1% per day, whereas the enzyme in the plasma regenerates at a more rapid rate, approximating 25% in the first seven to ten days.

"Circulating red blood cell and plasma cholinesterase may be conveniently thought of as a buffer system which serves to protect the individual against the nervous system effects of organophosphate toxins by binding the pesticide in the circulating blood, thereby preventing it from reaching the nervous system. Although this is an oversimplified explanation, it is a clinically useful one. In practice, an individual who has been chronically exposed to organophosphate pesticides should be withdrawn from further exposure when his cholinesterase values drop to 25% to 50% of normal, and should not be allowed to return until these values rise to at least 75% of normal. The individual who has been acutely poisoned and has shown marked cholinesterase depression should not be allowed to return to work with organophosphate pesticides until his

cholinesterase levels have returned to approximately 75% of normal.

Treatment

"Treatment of organic phosphate poisoning ranges from simple removal from exposure in very mild cases to the provision of very rigorous supportive and antidotal measures in severe cases. In the moderate to severe case, because of pulmonary involvement there may be need for artificial respiration using a positive-pressure method. Careful attention must be paid to removal of secretions and to maintenance of a patent airway. Anticonvulsants such as thiopental sodium may be necessary. The critical point is that respiration must be maintained since death usually results from weakness of the muscles of respiration and accumulation of excessive secretions in the respiratory tract. As soon as cyanosis has been overcome, 2 to 4 mg of atropine promptly should be given intravenously. This dose is approximately ten times the amount which is administered for other conditions in which atropine is considered therapeutic (emphasis in text). This dose should be repeated at five to ten minute intervals until signs of atropinization appear (dry, flushed skin, tachycardia as high as 140 beats per minute and pupillary dilatation). A mild degree of atropinization should be maintained for at least 48 hours. Atropine is contraindicated in a cyanotic patient because of the possibility

of inducing ventricular fibrillation.

"Although atropine remains the drug of choice, particularly if the treatment has to be continued for more than a day or two, pralidoxime (Protopam) chloride, is a commercially available antidote which complements atropine and hastens the reactivation of cholinesterase enzymes. For adults, in the moderate to severe case, it should be used along with atropine, injected intravenously as an initial dose of 1 gm at a rate not in excess of 500 mg/minute. After an hour, a second dose of 1 gm is indicated if muscle weakness has not been relieved. After an overwhelming inhalation or skin exposure to or after ingestion of the toxic agent, the doses may be doubled. For children the dose may be 25 to 50 mg/kg of body weight. Treatment with pralidoxime chloride will be most effective if given within 24 hours after poisoning. (Its usefulness after 36 to 48 hours is questionable.) Together, the two antidotes, atropine and pralidoxime chloride, are more effective than either one alone. Morphine, aminophylline, and the phenothiazines are specifically contraindicated.

"It is of great importance to decontaminate the patient. The stomach should be lavaged and a saline cathartic administered if the toxin has been ingested. Contaminated clothing should be removed at once and the skin should be washed with generous amounts of soap or detergent and a flood of water, which is best accomplished under a shower or by submersion in a pond or other

body of water if the exposure occurred in the field. Careful attention should be paid to cleansing of the skin and hair. The patient should be attended and monitored continuously for not less than 24 hours, since serious and sometimes fatal relapses have occurred because of continuing absorption of the toxin or dissipation of the effects of the antidote.

"Atropine is antagonistic to the muscarinic effects, which include anorexia, nausea, vomiting, abdominal cramps, sweating, salivation, constricted pupils, pulmonary edema, and cyanosis. Atropine has no effect on the nicotinic manifestations, which include muscle fasciculation and weakness. Pralidoxime chloride acts to regenerate cholinesterase and to reverse muscle weakness. Muscle weakness, specifically weakness of the muscles of respiration, is responsible for respiratory impairment and death in the fatal case. A fully atropinized patient may die of respiratory insufficiency."

XII. APPENDIX IV

METHOD FOR BIOCHEMICAL DETERMINATION OF CHOLINESTERASE ACTIVITY IN BLOOD

Although NIOSH recommends only erythrocyte cholinesterase determinations for routine cholinesterase tests, plasma cholinesterase may be screened in the same blood samples as a further diagnostic indicator. The method of Wolfsie and Winter, [93] a micromodification of the Michel method, [29] is the procedure recommended for the measurement of both erythrocyte and plasma cholinesterase activities.

Reagents

All reagents should be at least ACS reagent grade.

(a) Buffer solutions

Prepare each according to the following directions, noting that pH will decrease over a period of several weeks. The pH should be checked before a solution is used, and, if it has dropped more than 0.03 pH units, the solution should be discarded and a fresh one prepared.

(1) Erythrocyte

For 1 liter of buffer, dissolve 4.1236 g sodium barbital (0.02 M), 0.5446 g potassium orthophosphate, di-H (0.004 M), and 44.730 g potassium chloride (0.60 M) in 900 ml of distilled water; add 28.0 ml of 0.1 N hydrochloric acid while shaking the solution, and add distilled water to a volume of 1 liter. The pH should be 8.10 at 25 C.

(2) Plasma

For 1 liter of buffer, dissolve 1.2371 g sodium barbital (0.006 M), 0.1361 g potassium orthophosphate, di-H (0.001 M), and 17.535 g sodium chloride (0.30 M) in 900 ml of distilled water and add 11.6 ml of 0.1 N hydrochloric acid before bringing to volume. The pH should be 8.00 at 25 C.

(b) Acetylcholine substrates

A few drops of toluene are added to each acetylcholine substrate solution as a preservative, and the solutions are kept refrigerated when not in use. These solutions should not be kept for more than a week.

(1) Erythrocyte

Prepare a 0.11-M acetylcholine chloride solution (2.000 g in 100 ml of distilled water).

(2) Plasma

Prepare a 0.165-M acetylcholine chloride solution (3.000 g in 100 ml of distilled water).

(c) Saponin solution

For both plasma and erythrocyte cholinesterase determinations, prepare a 0.010% saponin solution (100 mg in 1,000 ml of distilled water).

Apparatus

(a) Centrifuge capable of 3,500 rpm and of holding capillary sample tubes.

(b) A pH meter, calibrated to 0.01 pH units.

(c) 0.02-ml Sahli-type hemoglobin pipet.

(d) Constant-temperature bath, 25 C.

(e) 100- and 1,000-ml volumetric flasks.

(f) Heparinized capillary tubes.

(g) A Bunsen burner.

Sample Handling and Preparation

Blood is collected from a clean, dry fingertip in a heparinized, glass capillary tube. The blood is allowed to flow into the capillary tube until the tube is approximately 3/4 full, leaving one end free by 1-1.25 inches to permit flame-sealing of the tip of the tube without overheating the blood sample.

The finger should be pricked deeply and care should be taken to collect only free-flowing drops of blood to guard against the initiation of the clotting process before the blood contacts the heparin on the wall of the capillary.

One end of the capillary is plugged with solid (room temperature) paraffin and the other (free) end is sealed in the flame of a Bunsen burner. The capillary may now be labeled with an adhesive tape tag bearing a serial number or name and date. The sample should then be centrifuged at 3,000-3,500 rpm for 50-60 minutes or the equivalent. When the sample has been so treated, it may be shipped to a laboratory, if necessary, or stored for several days (preferably in a refrigerator) without appreciable change.

Analysis

For analysis, the capillary is cut cleanly with a sharp ampule file. From the packed-cells section of the capillary, draw 0.02 ml directly into

a Sahli-type hemoglobin pipet. The ends of the capillary must be cut evenly to provide satisfactory juxtaposition with the tip of the pipet. Discharge the contents of the pipet directly into 1.0 ml of 0.01% saponin in a microbeaker, and rinse the pipet well (three times) into the solution. Glass vials, 1 inch (2.5 cm) deep by 3/4 inch (19 mm) in diameter, are convenient for electrometric testing. They will fit in the carrier of a standard pH meter and, when used with a clean rubber stopper, will eliminate transfer of the sample from a test tube for each pH measurement. Plasma is taken from the appropriate section of the capillary in the same manner as the packed erythrocytes and is discharged into 1.0 ml of distilled water, the Sahli pipet being rinsed into the solution (three times) as with the erythrocytes.

Erythrocyte Cholinesterase Assay

(a) One milliliter of hemolyzed erythrocyte suspension is added to 1 ml of erythrocyte buffer solution and placed in a 25 C water bath.

(b) After a 10-minute equilibrium period, the initial pH (pH(i)) is determined to the nearest 0.01 pH unit with the pH meter.

(c) Two-tenths milliliter of 0.11 M acetylcholine solution is added with rapid mixing; the time is recorded.

(d) The reaction proceeds for 1-1.5 hours before the final pH (pH(f)) is noted.

The beaker containing the solution should be shaken when the glass electrode is introduced to speed equilibration.

Note: Erythrocyte buffer solution is designed to yield a pH of 8.00

after the addition of hemolyzed human erythrocytes.

Plasma Cholinesterase Assay

(a) One milliliter of diluted plasma is mixed with 1 ml of plasma buffer solution.

(b) The solution is allowed to equilibrate in a 25 C water bath for 10 minutes.

(c) At the end of 10 minutes, the initial pH (pH(i)) is noted to the nearest 0.01 pH unit.

(d) Two-tenths milliliter of 0.165 M acetylcholine solution is added with rapid mixing.

(e) The reaction mixture is incubated for 1-1.5 hours before the final pH (pH(f)) is noted.

Calculations

The final units derived from this assay are delta pH/hour:

$$\text{Delta pH/hour} = \frac{c(pH(i) - pH(f)) - bc}{t(f) - t(i)}$$

where:

pH(i) = initial pH

pH(f) = final pH

t(f) - t(i) = time elapsed in hours between reading pH(i) and reading pH(f)

b = nonenzymatic hydrolysis corresponding to pH(f)

c = correction for variations in delta pH/hour with pH, corresponding to pH(f)

The b and c correction factors are given in Table XII-1. [1] Average baseline values of erythrocyte and plasma cholinesterase activity determined by this method for healthy nonexposed men and women, are given in Table XII-2. [93,112]

TABLE XII-1

CORRECTION FACTORS FOR USE IN THE EQUATION FOR DELTA pH/HOUR

pH(f)	Erythrocyte Cholinesterase Corrections		Plasma Cholinesterase Corrections	
	b	c	b	c
7.9	0.03	0.94	0.09	0.98
7.8	0.02	0.95	0.07	1.00
7.7	0.01	0.96	0.06	1.01
7.6	0.00	0.97	0.05	1.02
7.5	0.00	0.98	0.04	1.02
7.4	0.00	0.99	0.03	1.01
7.3	0.00	1.00	0.02	1.01
7.2	0.00	1.00	0.02	1.00
7.1	0.00	1.00	0.01	1.00
7.0	0.00	1.00	0.01	1.00
6.8	0.00	0.99	0.01	1.00
6.6	0.00	0.97	0.01	1.01
6.4	0.00	0.97	0.01	1.02
6.2	0.00	0.97	0.01	1.04
6.0	0.00	0.99	0.01	1.09

Adapted from reference 29

TABLE XII-2

MEAN BASELINE ERYTHROCYTE AND PLASMA CHOLINESTERASE VALUES IN MEN AND WOMEN (DELTA pH/HR)

	Erythrocyte Cholinesterase	
	Men	Women
Mean	0.861	0.843
	Plasma Cholinesterase	
Mean	0.953	0.817

Adapted from references 93 and 112

The average erythrocyte cholinesterase activity value for men is drawn from Wolfsie and Winter. [93] The value for women uses the product of the average erythrocyte cholinesterase activity figure for men [93] times the ratio (of mean delta pH/hour for women to mean delta pH/hour for men) formed from the data of Rider et al. [112] The use of the data from Wolfsie and Winter [93] allows for increased packing of erythrocytes in the capillary tubes above that obtained in more usual types of centrifuge tubes as well as for possible contamination of erythrocytes by plasma. Plasma cholinesterase values were selected from Rider et al, [112] since their larger data base probably provides a closer approximation of the true population mean of normal values for plasma cholinesterase activity. For the same reason, their data appear to provide the most reliable women/men ratio for erythrocyte cholinesterase activity.

XIII. APPENDIX V

MATERIAL SAFETY DATA SHEET

The following items of information which are applicable to a specific product or material shall be provided in the appropriate block of the Material Safety Data Sheet (MSDS).

The product designation is inserted in the block in the upper left corner of the first page to facilitate filing and retrieval. Print in upper case letters as large as possible. It should be printed to read upright with the sheet turned sideways. The product designation is that name or code designation which appears on the label, or by which the product is sold or known by employees. The relative numerical hazard ratings and key statements are those determined by the rules in Chapter V, Part B, of the NIOSH publication, An Identification System for Occupationally Hazardous Materials. The company identification may be printed in the upper right corner if desired.

(a) Section I. Product Identification

The manufacturer's name, address, and regular and emergency telephone numbers (including area code) are inserted in the appropriate blocks of Section I. The company listed should be a source of detailed backup information on the hazards of the material(s) covered by the MSDS. The listing of suppliers or wholesale distributors is discouraged. The trade name should be the product designation or common name associated with the material. The synonyms are those commonly used for the product, especially formal chemical nomenclature. Every known chemical designation or

competitor's trade name need not be listed.

(b) Section II. Hazardous Ingredients

The "materials" listed in Section II shall be those substances which are part of the hazardous product covered by the MSDS and individually meet any of the criteria defining a hazardous material. Thus, one component of a multicomponent product might be listed because of its toxicity, another component because of its flammability, while a third component could be included both for its toxicity and its reactivity. Note that a MSDS for a single component product must have the name of the material repeated in this section to avoid giving the impression that there are no hazardous ingredients.

Chemical substances should be listed according to their complete name derived from a recognized system of nomenclature. Where possible, avoid using common names and general class names such as "aromatic amine," "safety solvent," or "aliphatic hydrocarbon" when the specific name is known.

The "%" may be the approximate percentage by weight or volume (indicate basis) which each hazardous ingredient of the mixture bears to the whole mixture. This may be indicated as a range or maximum amount, eg, "10-40% vol" or "10% max wt" to avoid disclosure of trade secrets.

Toxic hazard data shall be stated in terms of concentration, mode of exposure or test, and animal used, eg, "100 ppm LC50-rat," "25 mg/kg LD50-skin-rabbit," "75 ppm LC man," "permissible exposure from 29 CFR 1910.1000," or if not available, from other sources of publications such as the American Conference of Governmental Industrial Hygienists or the American National Standards Institute Inc. Flashpoint, shock sensitivity,

or similar descriptive data may be used to indicate flammability, reactivity, or similar hazardous properties of the material.

(c) Section III. Physical Data

The data in Section III should be for the total mixture and should include the boiling point and melting point in degrees F hrenheit (Celsius in parentheses); vapor pressure, in conventional millimeters of mercury (mmHg); vapor density of gas or vapor (air = 1); solubility in water, in parts/hundred parts of water by weight; specific gravity (water = 1); percent volatiles (indicated if by weight or volume) at 70 degrees Fahrenheit (21.1 degrees Celsius); evaporation rate for liquids or sublimable solids, relative to butyl acetate; and appearance and odor. These data are useful for the control of toxic substances. Boiling point, vapor density, percent volatiles, vapor pressure, and evaporation are useful for designing proper ventilation equipment. This information is also useful for design and deployment of adequate fire and spill containment equipment. The appearance and odor may facilitate identification of substances stored in improperly marked containers, or when spilled.

(d) Section IV. Fire and Explosion Data

Section IV should contain complete fire and explosion data for the product, including flashpoint and autoignition temperature in degrees Fahrenheit (Celsius in parentheses); flammable limits, in percent by volume in air; suitable extinguishing media or materials; special firefighting procedures; and unusual fire and explosion hazard information. If the product presents no fire hazard, insert "NO FIRE HAZARD" on the line labeled "Extinguishing Media."

(e) Section V. Health Hazard Information

The "Health Hazard Data" should be a combined estimate of the hazard of the total product. This can be expressed as a TWA concentration, as a permissible exposure, or by some other indication of an acceptable standard. Other data are acceptable, such as lowest LD50 if multiple components are involved.

Under "Routes of Exposure," comments in each category should reflect the potential hazard from absorption by the route in question. Comments should indicate the severity of the effect and the basis for the statement if possible. The basis might be animal studies, analogy with similar products, or human experiences. Comments such as "yes" or "possible" are not helpful. Typical comments might be:

> Skin Contact--single short contact, no adverse effects likely; prolonged or repeated contact, possibly mild irritation.
>
> Eye Contact--some pain and mild transient irritation; no corneal scarring.

"Emergency and First Aid Procedures" should be written in lay language and should primarily represent first-aid treatment that could be provided by paramedical personnel or individuals trained in first aid.

Information in the "Notes to Physician" section should include any special medical information which would be of assistance to an attending physician including required or recommended preplacement and periodic medical examinations, diagnostic procedures, and medical management of overexposed employees.

(f) Section VI. Reactivity Data

The comments in Section VI relate to safe storage and handling of hazardous, unstable substances. It is particularly important to highlight instability or incompatibility to common substances or circumstances, such as water, direct sunlight, steel or copper piping, acids, alkalies, etc. "Hazardous Decomposition Products" shall include those products released under fire conditions. It must also include dangerous products produced by aging, such as peroxides in the case of some ethers. Where applicable, shelf life should also be indicated.

(g) Section VII. Spill or Leak Procedures

Detailed procedures for cleanup and disposal should be listed with emphasis on precautions to be taken to protect employees assigned to cleanup detail. Specific neutralizing chemicals or procedures should be described in detail. Disposal methods should be explicit including proper labeling of containers holding residues and ultimate disposal methods such as "sanitary landfill," or "incineration." Warnings such as "comply with local, state, and federal antipollution ordinances" are proper but not sufficient. Specific procedures shall be identified.

(h) Section VIII. Special Protection Information

Section VIII requires specific information. Statements such as "Yes," "No," or "If necessary" are not informative. Ventilation requirements should be specific as to type and preferred methods. Respirators shall be specified as to type and NIOSH or US Bureau of Mines approval class, ie, "Supplied air," "Organic vapor canister," etc. Protective equipment must be specified as to type and materials of construction.

(i) Section IX. Special Precautions

"Precautionary Statements" shall consist of the label statements selected for use on the container or placard. Additional information on any aspect of safety or health not covered in other sections should be inserted in Section IX. The lower block can contain references to published guides or in-house procedures for handling and storage. Department of Transportation markings and classifications and other freight, handling, or storage requirements and environmental controls can be noted.

(j) Signature and Filing

Finally, the name and address of the responsible person who completed the MSDS and the date of completion are entered. This will facilitate correction of errors and identify a source of additional information.

The MSDS shall be filed in a location readily accessible to employees exposed to the hazardous substance. The MSDS can be used as a training aid and basis for discussion during safety meetings and training of new employees. It should assist management by directing attention to the need for specific control engineering, work practices, and protective measures to ensure safe handling and use of the material. It will aid the safety and health staff in planning a safe and healthful work environment and in suggesting appropriate emergency procedures and sources of help in the event of harmful exposure of employees.

MATERIAL SAFETY DATA SHEET

I PRODUCT IDENTIFICATION	
MANUFACTURER'S NAME	REGULAR TELEPHONE NO. EMERGENCY TELEPHONE NO.
ADDRESS	
TRADE NAME	
SYNONYMS	

II HAZARDOUS INGREDIENTS		
MATERIAL OR COMPONENT	%	HAZARD DATA

III PHYSICAL DATA			
BOILING POINT, 760 MM HG		MELTING POINT	
SPECIFIC GRAVITY (H_2O=1)		VAPOR PRESSURE	
VAPOR DENSITY (AIR=1)		SOLUBILITY IN H_2O, % BY WT	
% VOLATILES BY VOL		EVAPORATION RATE (BUTYL ACETATE=1)	
APPEARANCE AND ODOR			

IV FIRE AND EXPLOSION DATA

FLASH POINT (TEST METHOD)		AUTOIGNITION TEMPERATURE	
FLAMMABLE LIMITS IN AIR, % BY VOL.	LOWER		UPPER
EXTINGUISHING MEDIA			
SPECIAL FIRE FIGHTING PROCEDURES			
UNUSUAL FIRE AND EXPLOSION HAZARD			

V HEALTH HAZARD INFORMATION

HEALTH HAZARD DATA

ROUTES OF EXPOSURE

INHALATION

SKIN CONTACT

SKIN ABSORPTION

EYE CONTACT

INGESTION

EFFECTS OF OVEREXPOSURE

ACUTE OVEREXPOSURE

CHRONIC OVEREXPOSURE

EMERGENCY AND FIRST AID PROCEDURES

EYES

SKIN:

INHALATION:

INGESTION

NOTES TO PHYSICIAN

VI REACTIVITY DATA

CONDITIONS CONTRIBUTING TO INSTABILITY

INCOMPATIBILITY

HAZARDOUS DECOMPOSITION PRODUCTS

CONDITIONS CONTRIBUTING TO HAZARDOUS POLYMERIZATION

VII SPILL OR LEAK PROCEDURES

STEPS TO BE TAKEN IF MATERIAL IS RELEASED OR SPILLED

NEUTRALIZING CHEMICALS

WASTE DISPOSAL METHOD

VIII SPECIAL PROTECTION INFORMATION

VENTILATION REQUIREMENTS

SPECIFIC PERSONAL PROTECTIVE EQUIPMENT

RESPIRATORY (SPECIFY IN DETAIL)

EYE

GLOVES

OTHER CLOTHING AND EQUIPMENT

IX SPECIAL PRECAUTIONS

PRECAUTIONARY
STATEMENTS

OTHER HANDLING AND
STORAGE REQUIREMENTS

PREPARED BY _____

ADDRESS _____

DATE _____