III. BIOLOGIC EFFECTS OF EXPOSURE

The normal function of the central nervous system (CNS) can be significantly altered by malathion intoxication. Acetylcholine (ACh), a choline ester, normally mediates neurotransmission in preganglionic autonomic fibers, postganglionic parasympathetic fibers, and in some postganglionic sympathetic fibers. [1] These fibers innervate the heart, iris, salivary glands, stomach, small intestine, urinary bladder, bronchial glands, eccrine sweat glands, and other structures. There is also evidence that ACh functions as a transmitter at neuromuscular junctions (motor endplates) and at certain synapses within the CNS. [1]

Two types of enzymes normally hydrolyze choline esters in humans: acetylcholinesterase (AChE) or true ChE (Enzyme Commission (EC) 3.1.1.7), and butyrylcholinesterase (BuChE) (EC 3.1.1.8), otherwise known as plasma ChE, serum ChE, or pseudocholinesterase. [1] AChE is found in neurons, at the neuromuscular junction, in erythrocytes, and in certain other tissues.

Inhibition of AChE leads to the accumulation of endogenous ACh [1] and, consequently, to the poisoning of the exposed individual.

BuChE is present in various types of glial or satellite cells of the central and peripheral nervous systems as well as in plasma, in the liver, and in other organs. [1] Its physiologic function is unknown; inhibition of the plasma enzyme at most sites produces no apparent functional derangement. [1] A diagram of the metabolism of malathion is presented in Figure XV-1.

Malathion itself has only a slight direct inhibitory action on erythrocyte ChE and nonspecific esterases but one of its metabolites,

malaoxon, is an active inhibitor. Malaoxon reacts with AChE to form the dimethylphosphoryl derivative, which is incapable of hydrolyzing ACh. [2-5] The resulting enzyme-inhibitor complex either may be slowly reactivated by dephosphorylation or permanently inactivated by aging via demethylation to the monomethylphosphoryl enzyme. The aging reaction is believed to play a critical role in the toxic actions of many organophosphates containing secondary or tertiary alkyl groups. [6]

Reactivation of the erythrocyte enzyme-inhibitor complex is accelerated by nucleophilic reactivators such as choline, pyridine, hydroxylamine, hydroxamic acids, and oximes. [7] Childs et al [8] found that the oximes were generally superior to the hydroxamic acids in reactivating organophosphate-inhibited ChE's. Subsequent studies have shown that pyridine-2-aldoxime methachloride (pralidoxime chloride) is highly effective in the reactivation of inhibited erythrocyte and neuroeffector ChE's if aging has not occurred. Wilson and Ginsburg [9] reported that pyridine-2-aldoxime is capable of reducing the toxicity of parathion in rats.

Extent of Exposure

Malathion (0,0-dimethyl S-(1,2-dicarboethoxyethyl) dithiophosphate), formerly known as malathon, [10] belongs to the family of organophosphorus pesticides. Pertinent physical properties of malathion are presented in Table XV-1, [11] and common trade names and synonyms for malathion are in Table XV-2. [12] Malathion is a colorless to light-amber liquid, with a solubility of 145 ppm in water at 25 C. While of limited solubility in

petroleum oils, it is miscible with most organic solvents. [13]

Malathion is produced by the reaction of 0,0-dimethyl dithiophosphoric acid and diethyl maleate dissolved in an alcohol or a ketone in the presence of a tertiary amine. An antipolymerization agent, such as hydroquinone, is usually used to inhibit polymerization of diethyl maleate. [13] Commercial malathion was introduced in 1950 as an experimental insecticide bv the American Cyanamid Company [11] and patented by the same company in 1951 (US Patent 2,578,652). By 1971, the annual production of malathion in the US totaled about 35 million pounds. [14,15]

Malathion is marketed as 99.6% technical grade liquid. Available formulations include wettable powders (25% and 50%), emulsifiable concentrates, dusts, and aerosols. [13,16]

Malathion is used in the control of certain insect pests on fruits, vegetables, and ornamental plants. It has been used in the control of houseflies, mosquitoes, and lice, [16] and on various farm and livestock animals. [17]

NIOSH estimates that approximately 75,000 workers in the US are occupationally exposed to malathion.

Historical Reports

The first indications that some organophosphorus compounds might be highly toxic appeared during the early 1930's when symptoms of ACh poisoning were experienced by the synthesizers of dimethyl and diethyl phosphorofluoridate. [18] In 1936, during an investigation of svnthetic insecticides, Schrader studied the phosphorus compounds, and in 1937 he

formula for organophosphate contact insecticides. [18]

Effects on Humans

Malathion absorption through the gastrointestinal tract, [19-26] the skin, [27-32] and the respiratory tract has been examined. [33,34] In vocational exposures, the skin is an impo rtant route of entry to the body, [35-37] and a case of intoxication following extensive skin exposure has been reported. [38] All but two reports of fatalities from malathion poisoning reviewed to date have involved ingestion [22,23,40]; two people may have died from contamination of the skin with malathion, but the attribution of the deaths to malathion remains somewhat uncertain.

Clyne and Shaffer [17] reported the experiences of three men who sprayed a liquid malathion formulation in grain-filled ship holds. Ceilings and inner walls of 34 ships' holds were sprayed intermittently during 4.5 months with either 5 or 10 quarts/ship of 57% emulsifiable liquid. The sprayers worked while lying on their backs on top of the grain and did not wear protective equipment. They did, however, wear coveralls which were changed daily; they showered after completing spraying for the day and washed their hands and faces after spraying each hold. The total duration of exposure was estimated to have been 35-40 hours. Erythrocyte and plasma ChE activities were monitored before, during, and after the project and were reported to have remained normal throughout. No symptoms of illness were detected in the men, despite what must have been very heavy dermal and respiratory exposure and probable oral exposure by swallowing airborne material.

The Expert Committee on Insecticides of the World Health Organization [41] briefly described its summary conclusions from reports on a large-scale field trial of malathion in Uganda in 1963-1965, and on routine spraying of malathion in 29,000 Central American houses from 1963 to 1967. The one safety precaution observed was to keep the compound away from foodstuffs, yet the only report of illness was in three sprayers whose uniforms were described as continually wet with malathion in kerosene for 3 days. Two of the sprayers were moderately ill for 3 days, and the third for 1 day, with symptoms of anticholinesterase poisoning. No further data were supplied in this report. Another case of symptomatic malathion poisoning directly attributable to workplace exposure involved a 16-year-old man who for 10 days was heavily exposed in Florida to malathion dust being applied to control the Mediterranean fruitfly (RM Clyne, written communication, February 1975). The young man used no protective measures to prevent exposure and, although his skin was visibly contaminated with dust, he reportedly did not bathe during the entire 10 days. He presumably inhaled a great deal of the dust and, in addition, frequently drank water directly from his cupped hands. After 10 days of exposure, he became ill with obvious manifestations of organophosphorus pesticide intoxication. He was admitted to a hospital, treated for 24 hours, and released as clinically well within 48 hours.

The State of California Department of Public Health publishes annual reports entitled Occupational Disease in California Attributed to Pesticides and Other Agricultural Chemicals--Some Current Problems. [42] The values contained in these reports are derived from data submitted by California physicians in accordance with the state's "Doctor's First Report

of Work Injury" reporting law. [43] The report required by this law is a brief summary of initial diagnostic impression and is accepted for statistical purposes by the state with little or no further communication. The data in Table III-1 were taken from the respective California Department of Health reports. In 1970, 33,085 occupational disease reports were included in the annual summary. Of these, 1,493, or 4.5%, were attributed to agricultural chemicals, but only 9, or 0.6%, of these reports involved malathion.

TABLE III-1

REPORTS OF OCCUPATIONAL DISEASE ATTRIBUTED TO MALATHION BY INDUSTRY AND YEAR, CALIFORNIA

Year	Industry Group		
	Agriculture	Manufacturing	All Other
1969	4 (162)	1 (40)	3 (29)
1970	5 (244)	0 (47)	4 (41)
1971	1 (197)	5 (31)	4 (41)

Numbers in parentheses refer to the total number of organophosphate poisonings.

Adapted from Occupational Disease in California Attributed to Pesticides and Other Agricultural Chemicals 1969--Some Current Problems [42]

A number of published case reports have been found which describe the toxic effects of acute exposure to malathion. Goldin et al, [39] in 1964, described the case of a 42-year-old woman who ingested a minimum of 120 ml of 50% malathion garden spray. She was admitted to a hospital 30 minutes

later, at which time she was comatose, markedly cyanotic, flaccid, devoid of tendon reflexes, and markedly miotic. Immediate treatment was begun with 1 mg of atropine im and 500 mg of 2-PAM iv. The history obtained from her family revealed that she had suddenly become unconscious with a major convulsion lasting for several minutes, and that excessive salivation and bronchial hypersecretion had developed rapidly thereafter. Approximately 1 hour after ingestion of the malathion, she received an additional 2,000 mg 2-PAM iv (total dose 40 mg/kg). At this stage, profuse diarrhea and massive bronchial hypersecretion which necessitated frequent tracheal-bronchial suction developed. Subcutaneous atropine therapy, 1-3 mg/hour, was begun. After 24 hours, the patient was able to move her arms and legs sluggishly on command. While pronounced tongue tremor and brisk jaw jerk were present, areflexia and pinpoint pupils persisted, as did the bronchial hypersecretion and cyanosis. Strength of muscles, other than those of respiration, returned to normal on about the 10th day after ingestion. After approximately 15 days, diaphragmatic and intercostal strength had recovered sufficiently to maintain spontaneous breathing. Neurologic recovery was gradual, with the tongue tremor and brisk jaw jerk disappearing, and the tendon reflexes returning, within about 1 week. The patient was discharged 5 weeks after admission. Laboratory investigations during the patient's hospital course included determinations of plasma and erythrocyte ChE activities. Serum ChE activity was less than 22% of laboratory normal for the first 9 days. Thereafter, the level gradually rose to 100% by the 31st day. The erythrocyte ChE activity was first measured on the 12th day, when it was found to be 10% of normal. It remained between 10 and 25% of normal until the 45th day after hospital

admission and then gradually rose to 100% by 130 days after admission. By this time the patient had been discharged. Hematocrit measurement showed a small drop after admission, from 43 to 37%, and the reticulocyte count never rose above 2%. Sternal bone marrow examination on the 25th day was essentially normal. Blood urea levels rose to 77 mg/100 ml of blood during the first 5 days, thereafter returning to normal as the nonrenal uremia due to diarrhea and hypersecretion was controlled. Electrocardiograms (ECG'S) taken immediately after admission and daily thereafter showed a prolongation of the P-R interval that persisted for 5 days, as well as changes in the S-T segment which the authors [39] reported as consistent with panmyocardial ischemia. These latter changes disappeared gradually as the patient's respiratory function improved.

Crowley and Johns [19] in 1966 described the case of a 45-year-old maintenance worker who ingested between 50 and 90 cc of commercial insecticide containing 50% malathion in a petroleum hydrocarbon base. Within an hour, he developed nausea, vomiting, and profuse diarrhea; shortly after, he also developed generalized sweating, increased salivation, lacrimation, and visual blurring. He began to have wheezing, giddiness, a feeling of generalized numbness, loss of coordination, urinary and fecal incontinence, and violent shaking and tremors, but he did not lose consciousness. Two hours after ingestion, he was brought to the hospital emergency room where he was found to be hyperactive, irritable but alert, and in mild respiratory distress. His skin was slightly dusky, his pupils were pinpoint, and excessive amounts of secretions were present in his mouth, pharynx, and respiratory passages. Scattered moist rales and expiratory wheezes were heard throughout his lung fields, but chest

expansion during respiration appeared adequate. Bowel sounds were active. Neurologic examination revealed normal sensory function with respect to superficial pain, touch, vibration, and proprioception. He had generalized skeletal muscle fasciculations and mild generalized weakness. Coordination was normal. Deep tendon reflexes were absent and his plantar responses were flexor. The patient was admitted to the hospital and immediately given 2 mg atropine iv, and this was continued as necessary until 18 days after admission. During the first 24 hours after malathion ingestion, his respiration became increasingly labored, apparently because of the increasing volume and tenacity of respiratory tract secretions together with pulmonary congestion and weakness of respiratory muscle contractions. The level of consciousness exhibited by the patient fluctuated widely from alertness to complete stupor. Skeletal muscle weakness increased, so that by 38 hours after ingestion of the insecticide the patient exhibited severe weakness of the trunk muscles and all extremities, with somewhat more power in the pelvic girdle and leg muscles and in muscles of the arms. He was able to initiate only a very weak muscular contraction, which fatigued after 5-10 seconds, followed by tremors involving the entire limb. This in turn was succeeded by flaccid paralysis of the limb. He had moderate ptosis of the eyelids with normal power of the extraocular muscles. There was weakness of the upper and lower facial muscles. He was able to open his mouth for 3-5 seconds but unable to protrude his tongue. On attempts to phonate, his palate elevated symmetrically. Therapy with 2-PAM was begun 48 hours after ingestion of malathion with 500 mg administered iv at 48 and 54 hours and 1 g iv each hour for 3 hours beginning 72 hours after ingestion. It was discontinued thereafter. The patient's level of

consciousness continued to fluctuate for 5 days. He then began to regain muscle strength and endurance. Erythrocyte ChE activity, which measured less than 10% of laboratory normal in the first 48 hours, began to rise after the first 7 days. He was discharged from the hospital on the 28th day after ingestion and was not seen again until the 56th day. At that time, his erythrocyte ChE activity had risen to 66% of normal. No description of the clinical course between the 7th and 56th days was given, and no further followup visits were reported.

Amos and Hall [20] described the case of a 14-year-old boy weighing 165 pounds who was admitted to the hospital 20 minutes after ingestion of approximately 4 ounces (120 ml) of a malathion preparation of unstated composition. At the time of admission, he had abdominal cramps, nausea, vomiting, excessive salivation, difficulty in breathing, and severe muscle weakness. Within several minutes of admission to the emergency room, he became cyanotic, developed pulmonary edema, and lapsed into coma. There were muscular fasciculations of the face, eyelids, and neck accompanied by mild muscular twitchings of the entire body. Pupils were pinpoint, round, regular, equal, and nonresponsive to light. Neurologic investigation revealed the absence of deep tendon reflexes and the presence of marked motor weakness. The patient was atropinized with 1 intravenous (iv) dose of 0.4 mg and with intramuscular (im) doses of 0.4 mg, 0.6 mg, and 0.6 mg in the 1st hour; and doses of 2 mg iv every 30 minutes from the 3d hour after admission onward; the authors reported total atropine dosage of 54 mg in the first 48 hours. After 3 hours, the state of deep coma, areflexia, and nonresponse to painful stimuli began to abate. The deep tendon reflexes returned and a slight increase in the size of the pupil with

pupillary reaction to light was noted. The patient regained consciousness for several minutes and then became combative, confused, and disoriented. Several hours later, he appeared alert with no evidence of motor paralysis. The day after admission, the patient began to deteriorate again, becoming progressively weaker with shallow respiration, rapid thready pulse, and a cloudy sensorium. There was the return of excessive salivation, difficult respiration, pinpoint pupils, muscular fasciculations of the face and neck, and twitching of the entire body as if convulsions were imminent. The patient was critically ill for the next 12 hours and required tracheostomy. Atropinization was continued and, after another 24 hours, his vital signs improved and respiration was again satisfactory. On the 3d hospital day, miotic pupils were again noted; his respiration was shallow and abdominal in type with no intercostal muscle function. No muscle twitching was present, but there was profound muscular weakness with inability to move the lower extremities more than a few inches from the bed. Ptosis of the eyelids suggested a severe myasthenia gravis-type weakness. At this time, the patient was rational with clear sensorium. Because of progressive respiratory failure complicated by pneumonia, it was decided to treat the patient with 2-PAM, 500 mg iv during 5 minutes, whereupon muscle function improved. During the next 24 hours, respiratory paralysis recurred, so that repetition of treatment with 2-PAM seemed necessary and was performed. After 72 hours, the hemogram, urinalysis, and ECG were normal. Atropine dosage was reduced to 0.5 mg/hour, and on the 7th day to 1 mg in every 4 hours. The patient was discharged from the hospital on the 15th day in satisfactory condition.

Harris et al [40] described the case of a 45-year-old woman who had ingested an indeterminate amount of malathion. She was admitted to hospital no more than 6 hours after ingestion and, by then, was unconscious and areflexic. She was in total cardiac and respiratory arrest. No involuntary motor activity was evident, although seizures did develop about 8 hours later. Some mucus about the mouth had been seen by the patient's husband at home, but no salivation or frothy sputum was observed thereafter. Pupils were of intermediate size and unreactive to light. There was no excessive sweating, diarrhea, or retching. She had not been incontinent. Moderate bradycardia, without gross arrhythmia, was noted. Laboratory tests at the time of admission showed hyperglycemia and 4+ glycosuria, but subsequent estimations of concentrations of glucose in the plasma were within the normal range. Ventilation via an endotracheal tube was required and levarterenol was administered iv. The authors [40] noted that voluntary respiratory effort was inadequate throughout most of the patient's hospital course. For the next 36 hours, maintenance of adequate blood pressure was dependent upon administration of levarterenol. The patient was initially given 4 mg of atropine iv. Throughout hospitalization, therapy was continued with 2-PAM, 1 g iv every 12-24 hours, and atropine, 1-2 mg iv or im every 1-4 hours. Two days after her admission, the patient showed slight voluntary movement and responded momentarily to external stimuli. Her pupils were dilated and there seemed to be stronger respiratory effort. However, ventricular fibrillation occurred 4 hours thereafter and, following this, the patient remained comatose. ChE activity was absent from both erythrocytes and plasma. The patient expired 5.5 days after malathion ingestion. Autopsy revealed

generalized edema, including severe pulmonary edema, and bronchopneumonia. These were probably indirect effects of shock and respiratory failure. A small subdural hematoma was also found, but no associated abnormalities to indicate head trauma were observed.

Windsor [44] reported the case of a 37-year-old woman admitted to the hospital after swallowing 60-85 ml of a malathion preparation (55% malathion, 35% crude naphthal extract), a dose the author calculated as equivalent to 35-50 g of malathion. She was an epileptic who was taking barbiturates, chloral hydrate, chlorpromazine, phenelzine, and phenytoin, and was under psychiatric care and had attempted suicide with chlorpromazine 2 years prior to this incident. On admission, the patient was described as "severely collapsed" and deeply cyanosed, with profoundly depressed respiration and bubbling rales throughout the chest. Pupils were pinpoint, but reflexes were detectable and symmetrical. Endotracheal intubation and bronchial aspiration were performed, and she was placed on a respirator. Gastric lavage resulted in the recovery of large, but unspecified, amounts of the malathion mixture. Atropine was administered in 2-mg doses every 15 minutes; pralidoxime chloride was administered iv in doses of 1 g at 2 and 6 hours after admission. The patient's pupils dilated and respiration improved, although during this time she had "several bouts" of diarrhea and generalized muscular fasciculations. The respirator was removed, but had to be replaced 24 hours after admission. Atropinization was continued until the morning after admission. It was resumed 14 hours later at a rate of 2 mg every 15 minutes, together with 1 g of pralidoxime every 4 hours. Serum ChE activity was determined to be about 20% of the normal minimum 3 days after malathion ingestion and

increased to about 45% of normal 1 week after the incident. Pralidoxime chloride was discontinued on the 5th hospital day, but atropinization was maintained until the 11th day, by which time a total of 1,065 mg of atropine had been administered. The respirator was withdrawn, and her recovery proceeded uneventfully.

Namba et al [25] reported the case of a 63-year-old man who was poisoned and admitted to a hospital after he drank between 120 and 180 ml of a malathion preparation (50% malathion, 42.4% xylene, and 7.6% inert ingredients) diluted in milk. Although roughly equivalent to 60-90 g of malathion, the quantity actually absorbed was probably less, according to the authors, as the subject had vomited and gastric lavage had been performed on him. The patient, an epileptic for 20 years, had been treated for that disorder with diphenylhydantoin and primidone. Upon admission to the emergency room, he was reportedly comatose and dyspneic. His blood pressure and radial pulse could not be recorded, mucous membranes were cyanotic, eye movements were absent, his conjunctivae were injected, and he had pinpoint pupils which were unresponsive to light. A foul-smelling, thick, whitish secretion filled his mouth and pharynx. The heart was not enlarged as determined by percussion, but heart sounds were distant and muffled by bilateral coarse rales and ronchi. The patient's limbs were flaccid and unresponsive to painful stimuli; neither tendon nor pathologic reflexes were observed. Also absent were the oculocephalic and caloric stimulation reflexes, as well as muscle fasciculations. He was incontinent and vomited copious amounts of garlic-smelling fluid. Treatment was begun with cardiac massage, assisted respiration, frequent tracheobronchial suctioning, and administration of 2 mg of atropine sulfate.

Electrocardiographic abnormalities (incomplete right bundle branch block, S-T depression) were found, as were further neurologic abnormalities. During his first hospital day, the patient received 22 mg of atropine sulfate and 3 g of pralidoxime chloride iv. Plasma ChE activity was reduced to 13% of normal on the first day and 4% on the second, while the erythrocyte ChE level was reduced to 7% of normal on the second and 3% on the third day. An injection of 1 g of pralidoxime chloride on the second day was purportedly responsible for a "temporary" 24% elevation of both erythrocyte and plasma ChE activities. At the time of the victim's death, 6 days after exposure, no lasting recovery of plasma (erythrocyte not mentioned) ChE activity had been achieved, nor had the ECG abnormalities resolved. During the course of treatment, he had received 24.4 mg of atropine sulfate iv and 12.0 mg im, and 6 mg of pralidoxime chloride iv. Autopsy disclosed basal adhesive meningitis (believed to have been associated with his history of epilepsy), arteriosclerotic changes in the coronary arteries without evidence of myocardial infarction, ulceration of the pharynx and trachea because of endotracheal intubation, right lung bronchogenic carcinoma (with left lung, hilar, and paratracheal node metastases), telangiectases in the buccal mucosa and jejunum, granulomas of the spleen and liver because of old histoplasma infection, and status post posterior colic gastrojejunostomy. ChE activities were reduced to 32% of normal in the cerebrum, 3% in the cerebellum, 1% in muscle, 19% in the liver, and 13% in the kidney on post mortem determination.

Richards [45] described the case of a 46-year-old woman who was admitted to the hospital after drinking 50 ml of 50% malathion solution. She was cyanotic and had marked respiratory depression. Artificial

ventilation was instituted, and a total of 790 mg of atropine administered in the first 6.5 days of treatment was followed by 60 mg more in the 2 days following. Five hours after the malathion ingestion, 1 g of 2-PAM was administered as well. The patient recovered, despite her moribund status on admission.

Two incidents of malathion poisoning in Sarawak, Malaysia, subsequent to deliberate ingestion were reported by Mathewson and Hardy. [46] The first was that of a 31-year-old woman who drank 56 ml of 57% malathion solution, an amount equivalent to 35 g of malathion. On admission to the hospital a half hour later, she was conscious and not cyanosed. Gastric lavage was performed, and atropine administration im was begun at a rate of 1.8 mg every 10 minutes. One and one-half hours after admission, by which time she had received 18 mg of atropine, the patient showed cyanosis, sweating, profuse salivation, pinpoint pupils, and muscular fasciculation. The subject was placed on artifical respiration, and atropine administration was continued at the same dose but iv. The next morning, despite apparent improvement in her condition, with better muscle power, less constricted pupils, and adequate self-ventilation (permitting withdrawal of the respirator), she deteriorated within a half hour after the respirator was removed and had to be reintubated. Paraldehyde was administered (6 ml iv) to stop generalized convulsions, and atropinization was still continued every 10 minutes. Tracheostomy was performed on the 3d hospital day, and on the 4th day the patient was able to breathe spontaneously for only 4-5 minutes every 2 hours. Peripheral muscle power was described as poor, although she could lift her hands off the bed. Atropine was continued, with the dosage adjusted according to her pupil

size. Spontaneous breathing returned for gradually longer periods, but artificial respiration could not be completely withdrawn until the 19th day. A total of 589 mg of atropine was administered to the woman in 37 days, mostly by iv or im routes during the first two weeks, and by stomach tube thereafter. She was discharged to a mental hospital 46 days after admission.

The second case [46] was that of a 16-year-old boy who drank 6 or 7 tablespoonsful of 57% malathion solution, a dose of about 30 g of malathion. On admission to the hospital 1.5 hours later, he was cyanosed and incontinent, with shallow bubbling respiration and pinpoint pupils. He was placed on a respirator and given 1 g of pyridine aldoxime ethanosulphate (P2S) iv. Atropine was given 2.5 mg in every 15 minutes. Gastric lavage was performed. An additional gram of P2S was administered iv after 2.5 hours; artificial ventilation was discontinued after 4.5 hours. Atropinization was continued, and the patient still had pinpoint pupils. Eight hours later, artificial respiration had to be resumed and 1.2 g of P2S was administered in divided doses. A fall in blood pressure 3 hours later required a 24-hour infusion of metaraminol during which time an additional 1 g dose of P2S was given. Not until the 19th hospital day could the use of the respirator be discontinued; thereafter, small doses of atropine were administered, to a total of 613 mg by day 24 after admission.

Goldman and Teitel [47] reported in 1958 that a 34-month-old child, weighing 21 kg (unusually heavy for this age), consumed 8 cc of 50% malathion in xylene. He was immediately given a glass of milk, a hard-boiled egg, and milk of magnesia. Fifteen minutes after ingestion, the child became limp and was taken to a physician, who noted muscle

flaccidity, miosis, and drooling. Stomach lavage was performed and the odor of malathion was noted in the material removed. One hour and forty minutes after ingestion of the pesticide, the patient was admitted to the hospital emergency room, stuporous, retching, and with rapid, noisy respiration. Miosis, hypersalivation, and excessive mucus secretion were observed, as were moist rales and expiratory wheezes. The child's deep tendon reflexes were absent, but abdominal and cremasteric reflexes were present and equal, and both the gag reflex and the response to painful stimuli were present. Incontinence and vomiting occurred. Gastric lavage was again administered. The progressive increase in respiratory distress and slight cyanosis prompted administration of atropine (0.15 mg iv and 0.15 mg subcutaneously) 1 hour after admission, although it was noted that spontaneous resolution of these signs had begun immediately prior to drug treatment. Two and one-half hours after admission, the child was conscious, alert, oriented, and in no respiratory distress. His lungs were clear, pupils in mid-miosis, and salivation was normal. Three hours after admission, respiratory distress again appeared, but without rales. It was felt that this episode was in part a reaction to the local irritating effect of the mixture of malathion and xylene. High humidity, created by use of a nebulizer, was used to alleviate this, and by the 5th hour after admission (6 hours, 40 minutes after ingestion), the child was asymptomatic and without clinical signs of intoxication.

These case reports [19,20,25,39,40,44-47] are consistent with the signs and symptoms attributable to the inhibition of AChE. The authors of these papers concluded, therefore, that malathion produces its acute toxic effects by this means; NIOSH concurs in this opinion.

The major signs and symptoms of malathion poisoning are attributable to the potentiation of responses to the ACh released from preganglionic and postganglionic cholinergic and somatic motor nerve endings whenever nerve volleys reach the periphery. In milder cases, the postganglionic stimulation may predominate. As listed in the case reports described above, these signs and symptoms include nausea, [19,20,27] vomiting, [19,20,25] diarrhea, [19,25,39] excessive sweating, [19,20,23,27] salivation, [19,20,23-25,27] miosis, [20,23-25,27,39] increased bronchial secretion, [19,20,24,25,27,39] bronchial constriction, [20,24,27] and the appearance of generalized muscular fasciculations followed by weakness. [19,20,23,24,27,39] Central nervous system effects include anxiety, [20] restlessness, [19,24] headache [27] and, in more serious cases, tremors, [19,20,39] confusion, [19] drowsiness, [19,20] slurred speech, [19] coma, [20,23,25,27,39,40] loss of reflexes, [19,20,24,25,27,39,40] and convulsions. [24,40]

The full range of manifestations of ChE inhibition is given in Table III-2.

Nalin's [50] classification of the clinical status of a series of 264 people who ingested malathion with suicidal intent is exceptionally complete. He found that severity of illness could be classified as mild (56%), moderate (14%), or severe (30%) based on the clinical criteria tabulated in Appendix VI. Patients with mild cases frequently had about them the characteristic pungent odor of malathion. Nausea, vomiting, and dizziness were common.

Moderately ill patients [50] had a strong odor of malathion, which was present in the milky gastric aspirate. They had sialorrhea with

TABLE III-2

SIGNS AND SYMPTOMS ASSOCIATED WITH ACUTE AND SUBACUTE EXPOSURES TO MALATHION

Effector Organ	Signs or Symptoms
(1) Muscarinic manifestations	
(a) Gastrointestinal	Anorexia, nausea, vomiting, abdominal cramps, diarrhea, tenesmus, involuntary defecation, eructation, "heartburn," sensation of substernal pressure
(b) Sweat glands	Increased sweating
(c) Salivary glands	Increased salivation
(d) Lacrimal (tear) glands	Increased lacrimation
(e) Cardiovascular system	Bradycardia, fall in blood pressure
(f) Bronchial	Sensation of tightness in chest, wheezing suggestive of bronchoconstriction, dyspnea, cough, increased bronchial secretion, pulmonary edema
(g) Pupils	Pinpoint (miosis) and nonreactive to light
(h) Ciliary bodies	Blurring of vision
(i) Bladder	Increased frequency of and involuntary urination
(2) Nicotinic manifestations	
(a) Striated muscle	Muscular twitching, fasciculation, cramping, weakness (including muscles of respiration)

TABLE III-2 (CONTINUED)

SIGNS AND SYMPTOMS ASSOCIATED WITH ACUTE AND SUBACUTE EXPOSURES TO MALATHION

Effector Organ	Signs or Symptoms
(b) Sympathetic ganglia	Pallor, tachycardia, elevation of blood pressure
(c) Adrenals	Elevation of blood pressure, elevation of blood glucose
(3) CNS manifestations	
	Uneasiness, restlessness, anxiety, tremulousness, tension, apathy, giddiness, withdrawal and depression, headache, sensation of "floating," insomnia with excessive dreaming (nightmares), ataxia, slurred and slow speech with repetition, drowsiness, difficulty in concentrating, confusion, emotional lability, coma with absence of reflexes, Cheyne-Stokes respirations, convulsions, hyperpyrexia, depression of respiratory and circulatory centers (with hypopnea or apnea and fall in blood pressure)

Derived from Grob et al [48] and Namba et al [49]

"foaming at the mouth," bronchospasm and bronchorrhea with rales or rhonchi, sweating, tearing, tachycardia, and sometimes mild stupor. Pupils were pinpoint except in a few milder cases. In 30% of the patients, transient glycosuria, as determined by reaction with glucose oxidase strips, was found. Three of sixteen of these patients had elevated concentrations of glucose in their blood. Blood urea concentrations

remained normal, but lactic dehydrogenase activities were elevated in the seven moderately ill patients tested. Serum calcium was determined in only seven cases and was not found to be elevated. Hematocrits were normal, but the white blood counts were reported to be acutely high. Specific values were not reported for these determinations.

Severely ill patients [50] had all of the above symptoms and signs, as well as hypotension, coma, cyanosis, areflexia, and sometimes convulsions, fasciculations, involuntary defecation, and Cheyne-Stokes respiration. Death occurred in 80% of the severely intoxicated patients despite intensive therapy. Nine of the severely ill patients had elevated blood pressures (mean, 170/110). Seven of these nine cases (four of them fatal) were females. Twitching of the eyelids, ankle clonus, muscle spasms, and acroparesthesias were occasionally noted.

Respiratory insufficiency was the cause of death in most reported cases of fatal malathion poisoning. [25] This terminal event was a consequence of one or more of the following: bronchial hypersecretion, weakness or paralysis of the intercostal muscles and diaphragm, and depression of the respiratory centers of the brain. [51]

The time of onset of signs and symptoms of poisoning after malathion exposure has varied from a few minutes [20,39,52] to several hours, [19] and to as long as 14 hours in the case of a child poisoned by the application of a 50% malathion-xylene hairwash solution. [27]

Hyperglycemia and glycosuria have been reported by three authors in connection with six patients with malathion poisoning, [25,27,40] although the pathophysiologic significance of this occurrence is unknown. Reports on three of these six patients noted the absence of acetone in the urine.

No comment on this finding was made in the other two cases. One case of glycosuria without hyperglycemia has been reported. [40] In one near-fatal accidental ingestion of malathion, the blood sugar was normal, and only a small amount of sugar was found in the urine. [19] Nalin [50] found transient glycosuria in 30% of the moderately ill cases. Five other reports of malathion intoxication [20,23,24,45,53] did not mention blood or urine glucose determinations. The degree of elevation of blood sugar that occurs in some cases of acute malathion poisoning has been shown to be much less than the levels associated with diabetic coma. [25]

Trinh Van Bao et al [54] conducted a study of 14 patients admitted to the hospital for malathion intoxication from either vocational exposure or attempted suicide. Blood samples were taken from all 14 patients within 3-6 days and thereafter at approximately 30 and 180 days from 12 of the group, one having died and another having declined to participate further in the study. Lymphocyte cultures were examined for chromosomal abnormalities and compared with those from 13 male and 2 female healthy unexposed controls. Stable chromosomal aberrations (ie, those containing deletions, translocations, and inversions) increased significantly immediately after intoxication, remained at a high level after 1 month, but returned to levels comparable with those seen in the controls after 6 months. The incidence of stable aberrations was not correlated with the dose of malathion. Of the patients examined, all were treated with atropine, and 9 (8 of the 12 completing the study) of the 14 also received Toxogonin (bis(4-hydroxyimino methylpyridinium(1)methyl)ether dichloride). The authors commented that they had not observed chromosomal damage in patients not poisoned by malathion but "treated with atropine shock." They

had no information to enable them to allow for the possible effects by Toxogonin, solvents, or vehicles involved in the poisoning and treatment on chromosomal structure.

Czeizel (unpublished manuscript) reported further data on the same 14 patients studied by Trinh Van Bao et al. [54] The malathion implicated in these studies was manufactured in Czechoslovakia. "Peripheral blood" samples, as cited by Trinh Van Bao et al, [54] revealed an "outstandingly high number of structural chromosome aberrations." The results of undefined animal experiments with malathion dissolved in cooking oil showed a higher frequency of break-isobreaks and deletions in bone marrow cells after 24 and 48 hours of treatment, and a higher occurrence of XY univalents and translocations in testicular tissue. This unpublished manuscript stated no doses, no correlation of the frequency of abnormalities with dose, and no numbers or types of experimental animals involved. In the absence of significant experimental details, such as dose and number of animals, and of independent confirmation in other laboratories, the significance of these unpublished observations for the human population is unknown. To this time, no human reproductive effects attributable to malathion seem to have been reported.

Mattson and Sedlak [55] measured the ether-extractable phosphates in the urine of an adult man who had been administered malathion in a single oral dose of 58 mg (0.84 mg/kg). A total of 23% of the ingested dose was recovered in the ether-extractable, urinary phosphate fraction of the urine during the first 16.3 hours. Ninety-seven percent of this recovered dose was excreted in the first 7.5 hours. A smaller dose of 11 mg (0.16 mg/kg) gave a similar excretion pattern. Based on experiments in rats injected ip

or fed 32P-labeled malathion, the authors [55] found an average of 69 and 36%, respectively, of the malathion excreted in the urine to be recoverable in the ether-extractable fraction.

Moeller and Rider [21] fed 10 healthy men daily doses of malathion dissolved in corn oil to determine the amount of malathion which can be ingested over an extended period of time without causing ChE activity depression. Five subjects each received 16 mg/day of malathion for 47 days, and five were given 24 mg/day for 56 days. Control ChE activities for these same subjects were determined twice weekly for 2 weeks preceding the experiment. The observed decrease in ChE activity reached a maximum 3 weeks after discontinuation of administration of malathion. Both erythrocyte and plasma ChE activities rebounded to normal quite rapidly thereafter, erythrocyte ChE reaching the baseline value and plasma ChE leveling off at about 93% of baseline within 10 days. The erythrocyte ChE overshot the baseline value during the month following a 20-day postdose observation period, whereas the plasma ChE remained at 90-95% of baseline during this period. It is possible that some reservoir of malathion or some persistent effect of malathion on AChE and BuChE was exhausted about 21 days after the cessation of daily doses.

Studies of humans exposed to malathion aerosols were reported by Golz. [56] For 42 consecutive days, three groups of four men each received a total of 84 1-hour exposures to malathion aerosols sprayed once into exposure-room air at calculated initial concentrations of 5.3, 21.2, or 84.8 mg/cu m. Since the aerosol concentrations represented a static exposure which must have declined rapidly, the actual dose received by each subject cannot be calculated. No significant decrease of ChE activity in

either plasma or erythrocytes was noted, nor were any cholinergic signs or symptoms found at any time during the study.

In 1960, Mattson and Sedlak [55] described a chemical method to measure exposure to malathion, which entailed the study of the urinary excretion of malathion-derived materials in humans. The method was developed using laboratory animals. In the first experiment conducted, one male and one female rat were given 100 mg/kg of 32P-labeled malathion intraperitoneally (ip) daily for 5 consecutive days, and one male and one female rat were given peanut oil only by the same route. A device which separated urine and feces was used, and both were collected for each 24-hour period. This separation, however, did not rule out the possibility of cross-contamination. The urine was first assayed for the total amount of radioactive materials present. The percentage of dose recovered ranged from 20 to 73 (average 46%) for the 5-day period. The authors [55] noted that, as no extraordinary attempt was made to obtain quantitative recovery of all the urine, some of the variation in percentage recovery might have been due to collection losses. They reported that assay for total malathion metabolites established the presence of large amounts of these in the rat urine. Samples at 48, 72, and 96 hours after cessation of treatment showed a rapid decline in the amount of metabolites recovered, although it was still measurable after 96 hours.

The acidified urine was extracted with carbon tetrachloride followed by ether. [55] The extracted urine was analyzed colorimetrically by both the procedure of Fiske and Subbarrow [57] and the Chen et al modification [58] of the Ammon and Hinsberg method, [59] with the latter method determined to be four times as sensitive as the former. The urinary

metabolites were calculated in terms of malathion.

Ether extracted 13-43% of the daily dosage from acidified urine for the treatment period. This represented 47-78% (average 66%) of the total malathion-derived material excreted in the urine. Using this extremely sensitive method, ether-soluble, malathion-derived materials were also found to be excreted up to at least 96 hours after cessation of treatment.

The dose was next reduced to 25 mg/kg, with the other experimental parameters maintained the same. Urinary excretion of the total daily intake ranged from 37 to 47% (average 42%), and daily urine samples taken at 24-hour intervals after treatment showed amounts of malathion-derived material detectable by radioassay. Ether-extractable materials ranged from 38 to 75% of the total daily intake, averaging 69%, which the authors [55] concluded was within the same range (average 66%) as for the higher level of intake. The authors deemed the amounts of ether-extractable urinary phosphates to be almost directly proportional to the dose of malathion at both doses used.

Malathion labeled with 32P administered orally to male and female rats at a dose of 100 mg/kg was excreted to the extent of 24 and 48%, respectively, in the urine, of which 31 and 36%, respectively, was ether-soluble. [55]

According to the authors, [55] colorimetric determination of phosphates in the ether-soluble materials gave good correlation with the radioassay. Ether extracts from the urine of control animals were reported to show no detectable amounts of organic phosphate.

The same method [55] was applied to human urine obtained from a man who ingested 58 mg of technical malathion (0.84 mg/kg). This dose was

below that capable of causing any detectable ChE inhibition. Preexposure urine samples showed no detectable amounts of ether-extractable phosphates. The amount of ether-extractable phosphates was seen by the authors [55] to rise quickly after dosage and then decrease to the predosage level. A total of 23% of the dose was recovered after 16.5 hours and 97% of this was excreted in the first 7.5 hours. The rate in mg/hour was greatest in the sample taken 1.3-3 hours after ingestion. A smaller dose of 11 mg of malathion (0.15 mg/kg) gave a similar result so far as the percentage recovered was concerned. However, metabolites could be detected for a shorter time after ingestion of the smaller dose of malathion, even though the more sensitive method for detection was used.

Application [55] of malathion in a talc dust to the skin of a man resulted in the appearance of ether-extractable phosphatic materials in the urine. One hundred milligrams of malathion as a 1% dust was applied for 14 hours; 1% of the dose was recovered during this period. It was reported that with higher amounts of malathion the percentage recovery of metabolites from the urine was of the same order. No ChE inhibition was detected.

Using the method described by Mattson and Sedlak, [55] Hayes et al [32] studied the urinary excretion of malathion-derived material after dermal application of various doses on human volunteers, as well as the effect of these applications on blood ChE activity. The malathion (minimum 95% purity) was formulated in talc at concentrations of 0, 1, 5, and 10%. Each volunteer was issued 90 g of powder 5 days/week for up to 16 weeks. After showering and contributing a blood sample if necessary, each volunteer dusted his entire body except head, neck, and genitalia with

powder. This required only 25-30 g, and the rest was sifted into the clothing. Hands and forearms were washed immediately after dusting. Prior to and after the experiment, physical examinations were conducted. Blood ChE activity was measured by the Nelson procedure [60] several days each week for the first few weeks, and weekly thereafter. Urine samples were collected in a hospital during an approximate 6-hour period every 6th night, both volume and sampling duration being recorded.

The initial review of symptoms [32] indicated no complaints resulting from significant illness and no increased prevalence of symptoms in one group. Only two complaints, burning of the skin and visible dermatitis, were noted during the course of the experiment. Results of the bromsulphalein test remained normal (less than 10% retention) throughout the course of the experiment.

A Student's t-test showed that erythrocyte ChE activity values determined [32] on paired samples by the methods of Nelson [60] and Michel [61] could not be distinguished. The experimental design did not allow prediction of either the minimum dose necessary to produce a statistically significant inhibition of AChE activity or the dosage necessary to produce clinical symptoms of intoxication.

The highest rate of malathion excretion [32] was 9.57 mg/hour, and the highest concentration of ether-extractable phosphate was 107 ppm calculated as malathion. The coefficient of correlation between body surface and the excretion of malathion-derived material ranged from 0.43 to 0.64 for the groups receiving 5 and 10% malathion; no correlation was found in the 1% group.

The mean malathion excretion rate [32] was 0.30 ± 0.19 mg/hour for 1% dusting powder, 1.74 ± 1.65 mg/hour for 5%, and 1.99 ± 1.25 mg/hour for 10% powder. Given 28 g as the amount of dusting powder actually applied, the formulations made available 0.28-2.8 g of malathion. Mean minimum amounts of 7.8, 41.76, and 48 mg/day, respectively, of the malathion in the 1, 5, and 10% formulations, were excreted as measured ether-extractable material. As shown by Mattson and Sedlak, [55] these figures are in the order of one-third lower than the actual excretion rate because of the extraction of only a part of the malathion and its metabolites. Only with the 10% malathion dust was there any evidence of possible depression of ChE, suggesting that the human body can be safely exposed to up to 2.8 g of malathion in talc powder/day applied to the skin. This is equal to an absorbed dose of 40 mg/kg for a standard 70-kg man.

Mammalian tissues effectively detoxify malathion. [62,63] An enzyme which hydrolytically cleaves one ethyl alcohol moiety from malathion has been partially purified from human liver specimens by Main and Braid. [62] In addition to malathion, the enzyme preparation (termed aliesterase) was found to hydrolyze several aliphatic and aromatic esters. The half-maximal velocity (Km) of the enzyme was estimated to be 48 μM. The maximum capacity of human liver homogenates to degrade malathion was estimated to be 7.9 μM/g of liver/minute. Since only 1 mole of a titratable acid was released for each mole of malathion, the authors concluded that the reaction product was malathion monoacid. Since neither malathion monoacid nor malaoxon monoacid is a ChE inhibitor, the catalytic detoxification of malathion by aliesterase is an important aspect of mammalian resistance to malathion-induced intoxication. Because most other commercial

organophosphates do not contain a carboxy ester as a functional group, they are not substrates for aliesterase and are not detoxified by this mechanism. This fact probably contributes to the relatively low order of toxicity of malathion for mammals in comparison with that of other organophosphates. [64]

Matsumura and Ward [63] investigated the in vitro degradation of malathion by fresh human liver samples. Tissue samples from livers frozen no later than 16 hours post mortem were homogenized in Kronecker's saline solution at a concentration of 20 mg of tissue/ml. Aliquots of 1 ml of homogenate were incubated with 0.00001 M malathion for 1 hour. Unreacted malathion and soluble metabolites were extracted twice from the aqueous fraction with an equal volume of chloroform. The combined solvent phase was reextracted with sodium phosphate buffer. The aqueous phase was washed with chloroform. All solvent phases were combined, dried over sodium sulfate, and analyzed by thin layer chromatography. The results were given by the authors in terms of percentage of substrate added. The authors [63] stated that the accuracy of recovery of their procedure was 96.7 ± 8.5%.

Six samples of human liver homogenate tested [63] hydrolyzed 99.57, 95.28, 98.45, 98.44, 97.94, and 98.53% of the added malathion. Of the hydrolysis products, 3.97-5.71% appeared as desmethyl malathion, 0.07-1.23% as diethyl mercaptosuccinate, 0.08-0.48% as diethyl malate, 85.51-90.76% as carboxyesterase products, and 0.09-7.38% as "others." Although the data suggested that malathion is efficiently detoxified by humans through the action of the carboxyesterase. Walker et al [65] found that in the human liver, this same carboxyesterase is beta-aliesterase. [64,66] Although the results of the Matsumura and Ward in vitro study [63] reinforce the

knowledge that carboxyesterase activity is of prime importance in the detoxification of malathion in humans, they cannot be taken as an absolute indication of the proportions of metabolites to be expected in vivo.

Milby and Epstein [67] studied the contact-sensitizing effect of malathion on skin under both experimental and field conditions. The experimental study utilized 87 healthy male volunteers divided into four groups. The first group was exposed for 2 days to 10% malathion in ethanol on a skin site previously irritated by a 3-second freeze with dichlorodifluoromethane (Freon 12). There were no controls for the first group. Group 2 was exposed to the 10% malathion solution applied to a nonirritated site. Groups 3 and 4 received the Freon 12 irritation and were then exposed to 1.0 and 0.1% solutions of malathion in ethanol, respectively. All applications were made under occlusive dressings. After 30 days, all subjects were retested at a new site with a nonirritating dose of malathion (1% in ethanol). The areas were graded after 2, 4, and 6 days for intensity of cutaneous response (1+, erythema and edema, to 4+, bullae). Ten percent malathion was found to induce contact sensitization readily and the reactions were marked. To further examine the degree of sensitization, five highly malathion-sensitive subjects were tested with weak solutions of malathion in acetone and in water. A 1:10*6 (* means to the power of) concentration in acetone evoked 4+ reactions, and a commercial product consisting of 0.9% malathion in water with an unidentified suspending agent evoked positive responses as well. The field study comprised two occupationally exposed groups, the first composed of 157 sprayers, mechanics, and supervisors from mosquito abatement districts (93% of the total work population of those districts), and the second of 43

poultry-rancher volunteers (10% of the area's ranchers) who had used malathion for at least one season during the past 3 years. Each subject was interviewed for history of past or present asthma, hay fever, allergy, skin disease, and systemic poisoning by pesticides. Records were made of each individual's exposure to malathion and other organophosphate pesticides. Exposure was quantified according to the total years of malathion use and an estimate of the number of times it was used. The subjects then were tested with an emulsion of 1% malathion in distilled water applied on a cloth pledget under adhesive tape to the skin of the upper arm for 2 days. Three days later, the reaction was observed and graded. Four of the 157 workers in the first group (3%) showed positive reactions. They were all considered heavily affected by the grader. Three of the four had previous histories of undiagnosed skin eruptions. The greatest number of sensitized individuals was found in a district which exclusively employed No. 2 diesel oil as the spray vehicle. The authors speculated that the properties of the oil may have contributed to the apparent skin sensitization. Two of the 43 poultry ranchers had positive reactions (5%). While these reactors had not been as heavily exposed as the mosquito abatement reactors, they also had histories of unexplained dermatitis. The results of these studies indicate that malathion may act as a contact skin sensitizer in humans, and that clinically significant dermatitis may occur under conditions of heavy field use. A chi-square test associating patch-test reactivity and history of dermatitis in mosquito abatement district workers was significant at the 5% level.

Several investigators have studied the exposure of workers to malathion in agricultural pest and vector control operations. [35-37,68,69]

These measurements of respiratory and skin exposure were undertaken to determine the margin of safety between exposure dose and estimated toxic dose. ChE activities, where determined, verified the absence of significant depression as a consequence of exposure to occupational field-encountered concentrations of malathion. These reports are described in Chapter IV.

Epidemiologic Studies

Only two epidemiologic reports on the effects of occupational exposure to malathion were found. Grech [69] examined alterations of the activities of serum BuChE, SGOT, SGPT, and serum aldolase, and the concentration of serum albumin in 12 agricultural workers exposed to malathion over a period of 6 months. Two groups of controls were used, consisting of 30 blood samples each, the first from randomly selected healthy blood donors and the second from healthy blood donors engaged in manual labor. The mean BuChE activity of the agricultural workers at the end of the exposure period was not significantly different from that of either group of controls. However, the author [69] stated that the enzyme activity of any single subject changed significantly after exposure. A reduction in BuChE activity was noted in 11 of 12 agricultural workers, and 6 showed a sustained fall until the end of the study. Two showed slight increases over their preexposure activities at the end. Both SGOT and SGPT were found to be without significant difference between the two groups of controls, or between the mean values of the agricultural workers and those of the controls. The largest percent changes in the mean values of the agricultural workers during exposure to malathion were decreases in the

activities of serum aldolase, SGOT, and SGPT, with that of BuChE changing the least of the four serum enzymes studied. No significant differences were observed in serum albumin concentrations. This study indicates that BuChE depression secondary to malathion exposure under field conditions does occur.

In 1968, the New Jersey State Department of Health [70] studied the medical records of 35 malathion- and/or Thiophos- (parathion-) manufacturing employees, 11 of whom had been employed in this capacity for 15 years or more. Medical examinations by the company had been done on a preemployment basis, and at 1-1.5 yearly intervals thereafter. The Department of Health reviewed the medical records of 16 employees who had averaged 15.5 years of work in the organophosphate-production facilities. Eleven of these men had been exposed primarily to malathion, with sporadic exposure to the organophosphates Abate, Zinophos, and parathion. After carefully reviewing and analyzing the company medical records, the Department of Health concluded that the 16 men were suffering no adverse chronic health effects from exposure to the materials used in the manufacture of the organophosphates. Beginning in March 1953, monthly ChE testing was done on workers in the manufacturing unit, which had begun operation in 1951. All employees were required to wear protective coveralls, helmets, and boots upon entering the work area, as well as rubber gloves and respirators when in the work area or engaged in manufacturing operations. Accidental splash exposures occurred in two of the long-term workers: one because of a leak (following which the employee showered and changed, but developed headache, nausea, and redness and itching of the arms and chest); the other by malathion being sprayed on the

face (no symptoms developed). ChE activities measured 33 and 34 days later, respectively, were within normal limits (erythrocyte 0.93 delta pH, plasma 1.1 delta pH in the first worker; bromthymol blue test, 90-100% activity in the second).

Animal Toxicity

Malathion is absorbed through the intestinal tract, [10,33] the skin, [10,33,71] and the respiratory tract. [33] Following absorption, it is transported via the circulatory system to the liver, where it is metabolized in part to products which are not inhibitors of ChE [72] and in part to its oxygen analog, malaoxon, a potent ChE inhibitor. [73] In the liver, both malathion and malaoxon are rapidly hydrolyzed, and thus detoxified, by an esterase enzyme system, [73,74] which itself is inhibited to some extent by malaoxon. [73,74] Hydrolysis of malathion is not restricted to the liver but occurs also in human brain, where two highly active esterases, two moderately active ones, and six others of lesser activity have been separated electrophoretically by Sakai and Matsumura. [75] This rapid enzymatic hydrolysis accounts for the low toxicity of malathion [76] to mammals as compared with insects, which have relatively less capacity to hydrolyze malathion. [77] The metabolites of malathion are listed in Table XV-4. Compounds such as EPN which inhibit this system of enzymatic hydrolysis tend to enhance the toxicity of malathion. [78-80] Compounds which induce the enzymes tend to decrease malathion's toxicity. [66]

After formation in the lungs, kidneys, liver, and other organs, malaoxon is transported via the circulatory system to the nervous system

and to muscle, where it sets in motion the following sequence of events: inhibition of ChE activity at synapses and motor endplates; accumulation of ACh at these sites; and the appearance of signs and symptoms of ChE inhibition or ACh poisoning. [64] The biochemical mechanism by which malaoxon inhibits the enzyme ChE involves phosphorylation of the active site of the enzyme. The phosphorylated enzyme is incapable of hydrolyzing ACh. [64] In time, spontaneous reactivation of the enzyme occurs or the inhibited enzyme "ages," becoming nonsusceptible to reactivation by 2-PAM and other oximes. [81]

The signs of acute intoxication which appear in animals poisoned with these agents are similar to those which occur in humans. These are listed in Table III-2. [82] Other manifestations of malathion toxicity which are unrelated to ChE inhibition are discussed later in this chapter.

The LD50's for rats, mice, and guinea pigs by various routes of exposure are given in Table XV-5.

The sex difference in susceptibility to the effects of malathion is generally confined to rodents. Sex differences in toxicity have been found for most organophosphorus pesticides. In a study [78] of the inhibition of the ChE's of the blood by malathion, the authors observed that larger doses were required to produce given inhibition of the enzymes in the male than in the female because of the male's higher rate of metabolism of malathion by oxidative microsomal enzymes. [79] Species differences have been investigated and attributed to differences in rates of detoxification by hydrolytic enzymes.

Golz [33] reported Hazleton's results on the acute iv toxicity of malathion in dogs. A dose of 100 mg/kg had no apparent effect, while 200

mg/kg produced severe signs and marked ChE inhibition and 250 mg/kg was lethal. No details of the purity of the malathion or of the ages, sexes, or numbers of dogs used were provided, nor was there any clarification of the observed signs.

The subchronic toxicity of malathion in rodents also has been investigated. Ten male and ten female rats [33] were given 90% technical malathion at a concentration of 5,000 ppm in the diet for 4-6 weeks. The average daily intake was 62 mg/kg for the male rats and 68 mg/kg for the females. All animals survived until killed after the 4th, 5th, or 6th week of feeding. No signs of adverse effects and no physical or behavioral changes were noted. ChE activity determinations were made on brain, plasma, and erythrocytes. Essentially all tests showed ChE inhibition of at least 50%, presumably compared with control animals although this was not stated. Also unspecified were the sex distribution and distribution relative to week of killing for the five animals showing 100% erythrocyte ChE inhibition. In another study, [33] an unspecified number of rats of unstated age and sex were fed 90% technical malathion at 5,000 ppm in the diet (77.9 mg/kg/day) for 63 weeks. At termination, the exposed animals weighed 12% less than the controls. ChE activity was not determined. The author reported that no other effects were detected. Another group of six rats of unspecified age and sex was given 99% malathion at 20,000 ppm in the diet (275 mg/kg/day) for 68-70 weeks. Four of the animals died before the conclusion of the experiment; the two survivors weighed 36% less than the control rats.

Clyne and Shaffer [17] reported the results of 2-year feeding studies of technical grades of malathion (90% technical and 99+% technical, both as

25% wettable powder formulations) in male and female albino rats. Survival, effects on food intake and growth, and degrees of plasma, erythrocyte, and brain ChE inhibition determined when the animals were killed were measured for comparison with control group values. The 90% technical grade malathion was administered at concentrations of 100 ppm (6 mg/kg of malathion) to 20 males, of 1,000 ppm to 20 males and 10 females (60 mg/kg and 80 mg/kg, respectively), and of 5,000 ppm (350 mg/kg) to 20 males. The control group comprised 20 males and 10 females. Of the controls, 10/20 males and 5/10 females were alive after 2 years. Survival rates in the 100-, 1,000-, and 5,000-ppm groups were: 15/20 males, 11/20 males and 8/10 females, and 14/20 males, respectively. No effects on food intake or growth were reported below the 5,000-ppm concentration; at that level, a retardation of growth was observed. Plasma, erythrocyte, and brain ChE activities were depressed 10-30% in the 100-ppm group. The 1,000-ppm males showed 10-30% plasma, 60-95% erythrocyte, and 10-30% brain ChE inhibition, while the females in this group showed no plasma inhibitions, but otherwise were the same as the males. Inhibition of 60-95% in both plasma and brain ChE's and total inhibition of erythrocyte ChE were found in the 5,000-ppm males.

The 99+% technical grade malathion [17] was administered at concentrations of 500 ppm to four male and four female rats (30 and 40 mg/kg, respectively), of 1,000 ppm to four males and four females (60 and 80 mg/kg, respectively), of 5,000 ppm to three males and four females (380 mg/kg for both sexes), and of 20,000 ppm to three males and three females (720 and 1,800 mg/kg, respectively). Survivals after 2 years for these groups were 2/4 males and 3/4 females at 500 ppm, 2/4 males and 1/4 females

at 1,000 ppm, 3/3 males and 3/4 females at 5,000 ppm, and 0/3 males and 2/3 females at 20,000 ppm (all males having died within 20 days at this level). No effects on food intake or growth were noted in males at 500 ppm, or in either males or females at 1,000 ppm. No results for the 500-ppm females were given. At doses of 5,000 ppm, both males and females exhibited reduced food intakes, and growth in males was retarded. Because of the early death of the 20,000-ppm males, only data for reduction of food intake and retardation of growth for the females were given. No inhibition of plasma or brain ChE was noted, but erythrocyte ChE was inhibited by 60-95% in both sexes at the 500-ppm concentration. At 1,000 ppm, no inhibition of plasma ChE, 60-95% inhibition of erythrocyte ChE in males and complete inhibition in females, and 30-60% inhibition of brain ChE in males and 10-30% in females were reported. ChE inhibitions at 5,000 ppm were 10-30% in plasma and 100% in erythrocytes of both sexes; brain ChE was inhibited 30-60% in males and 10-30% in females. In the surviving female fed 20,000 ppm, both plasma and brain ChE activities were inhibited by 60-95% and erythrocyte ChE by 100%. Gross and microscopic tissue examinations reportedly revealed no anatomic lesions attributable to the action of malathion. [17]

The experimental results of Fogelman on the effects of chronic feeding of malathion to rats were submitted to NIOSH (CB Shaffer, written communication, March 1976). Groups of three or four weanling albino rats (Carworth Farm strain) of each sex were fed diets containing 500, 1,000, or 5,000 ppm of 99% malathion for up to 104 weeks. In a parallel experiment with 90% malathion, 20 male rats and up to 10 female rats by group were fed 0, 100, 1,000, or 5,000 ppm of malathion for up to 104 weeks. At autopsy,

tissues from the liver, kidney, adrenal, spleen, large intestine, small intestine, brain, lung, bladder, and for females from the uterus, were examined microscopically from six animals of each group. Although no evidence of neoplasia was reported, the number of animals used was insufficient to allow a firm conclusion with respect to the induction of cancer by malathion in either this or any other species.

Using 32P-malathion, Mattson and Sedlak [55] measured the urinary excretion of all 32P-containing metabolites from one male and one female rat that had been injected ip with 100 mg/kg of the material. The total number of rats in each group and sex distribution were not specified. During the 24 hours after each of the five daily injections, the rats excreted a mean of 46% of the daily dose. After the end of daily dosing, the excretion rate fell rapidly but was still measurable after 96 hours. Male and female rats given 100-mg/kg oral doses of 32P-malathion excreted totals of 24 and 48%, respectively, in their urine. One male and one female rat injected ip with 25 mg/kg of 32P-malathion on each of 5 consecutive days excreted a mean of 42% of the injected dose during each 24-hour period following a dose. [55] The male excreted an average of 45% and the female 38%. After the daily injections were stopped, the label was detected in the urine for up to 5 days; between 24 and 120 hours after the last dose, the male excreted 1.5% of the daily dose and the female 5.1%. In the urine obtained after ip injections of 32P-labeled malathion, an average of 68% of the radioactivity was extracted from the urine by diethyl ether, whereas only 34% was similarly extractable after oral administration. Although a device was used to separate the urine and feces during collection, the authors [55] admitted that some cross-contamination

may have occurred. Fecal excretion of 32P-labeled material was not measured.

Hazleton and Holland [10] reported the results of inhalation studies on rats, guinea pigs, mice, rabbits, and dogs. Unspecified numbers of rats and guinea pigs were exposed to air bubbled through 90% technical malathion at 30 C, 8 hours/day, 5 days/week, for 2 weeks. No fatalities, no signs suggestive of ChE toxicity, and no significant lowering of ChE activity were observed. The estimations of ChE activity were made by an unspecified electrometric method, and the chamber concentration of malathion was not measured. Mice, rats, guinea pigs, and rabbits (numbers unspecified) were exposed to an aerosol of 90% malathion at a concentration of approximately 60 ppm for 6 hours/day for 2 days. The only responses noted were sneezing and rhinorrhea. Erythrocyte, plasma, and brain ChE activities were observed to be within normal limits for rats and guinea pigs, but they were not reported for mice and rabbits. Rats, guinea pigs, and a dog were exposed for 7 hours/day, 5 days/week, for 4 weeks to aerosolized malathion at a concentration of 5 ppm. The dog and guinea pigs lacrimated, but no tremors, salivation, or other signs of intoxication by organophosphate compounds were seen. No significant reduction in the ChE activities of plasma, brain, or erythrocytes were found, and no gross pathologic changes were detected at necropsy. Microscopic examination disclosed thickening and leukocytic infiltration of the intraalveolar septa.

Rats, guinea pigs, and dogs [10] exposed for 7 hours/day, 5 days/week, for 6 weeks to an aerosol containing dust equivalent to 5 ppm of malathion showed no gross evidence of toxicity. Moderate inhibition of plasma, erythrocyte, and brain ChE's were noted in the rats. The guinea

pigs had no significant inhibition of these ChE activities; one of two dogs had slightly inhibited plasma and erythrocyte ChE activities. Again, analytical methods were not detailed.

Weeks et al [83] exposed groups of 6 male New Zealand white rabbits and 20 Coturnix quail to aerosols of technical (95%) malathion and of 6% malathion in No. 2 fuel oil for periods of 6 hours. At concentrations of 6 and of 34 mg/cu m of aerosolized technical malathion, there were no toxic signs or significant decreases in blood ChE activities of the rabbits. At 34 mg/cu m, reductions in plasma ChE activity which averaged 51% were noted in the quail immediately after exposure, but no reduction was observed at 24 hours nor at 7 days postexposure. Plasma ChE activity was reduced in quail by 84% immediately following and by 63% at 24 hours after exposure to malathion at 65 mg/cu m, but it was not affected 7 days postexposure. No toxic signs or changes in blood ChE activities were found at this concentration (65 mg/cu m) in rabbits, although at 123 mg/cu m, significant decreases in ChE activities were found in plasma at 24 hours, and in erythrocytes at 24 and 72 hours and at 7 days. Quail exposed to 123 mg/cu m had significantly decreased plasma ChE activities immediately following exposure and at 24 hours, but not at 7 days. Microscopic examination of the tissues and organs of the test animals killed 7 days after exposure showed no pathologic changes due to the compound. From these data, quail appeared to react to malathion exposure in qualitatively the same way as rabbits, but to be slightly more sensitive to the compound at these concentrations and this duration of exposure.

Six-hour exposures [83] of groups of the same species and numbers of animals to aerosols of 6% malathion in No. 2 fuel oil resulted in the same

minimal effects of ChE depression in quail at concentrations equivalent to 24 and 34 mg/cu m of malathion as did exposures to aerosols of the 95% compound. Particle sizes were 12 μm mass median diameter (mmd) for the 95% formulation, and 25 μm mmd for the 6% formulation. Thus, it would appear that particle size had no effect on the responses of quail and rabbits to these aerosols. Exposures to 6% malathion in No. 2 fuel oil at levels of 66 and 128 mg/cu m resulted in mortalities due to respiratory distress, rather than to ChE inhibition, in both quail and rabbits. The respiratory difficulty appeared to have been caused by the heavy concentration of fuel oil in the formulation. One may conclude, then, that the possible toxic effects from the fuel oil may be a greater risk than those from malathion itself under these experimental conditions.

Oral administration of single doses of 0, 12, 120, 300, 600, or 1,200 mg/kg of technical malathion in corn oil to groups of six rabbits of the same strain was undertaken by Weeks et al [83] for comparison with the aerosol exposure results. They found that an oral dose of 300 mg/kg caused approximately the same inhibition of AChE as did the estimated dose of about 15-20 mg/kg inhaled by rabbits exposed to an aerosol containing 123 mg/cu m for 6 hours. These results indicated a greater hazard from inhalation than from ingestion of equivalent amounts of malathion.

Murphy et al [5] compared the abilities of malathion and its oxygen analog, malaoxon, injected ip, to inhibit the ChE activity in groups of four mice, of two or four chickens, and four of each of two fish species. They found that mice appeared to be 25 times more susceptible to malaoxon than to malathion, chickens 13 times, and bullheads about 80 times. The findings on sunfish were not appropriate for this kind of analysis, but the

authors indicated that this species was much more susceptible to the effects of malaoxon than to those of malathion. These results were adduced from brain ChE inhibition assays conducted in duplicate according to the manometric method of DuBois and Mangun. [84]

The occurrence and significance of in vivo interactions between malathion and other organophosphorus pesticides [53,80,85-91] and between malathion and certain drugs and chemicals [92,93] have been the subjects of numerous reports.

In 1957, Frawley et al [86] studied the effects of both single and coadministration of malathion and O-ethyl O-p-nitrophenyl phenylthiophosphonate (EPN) on 35 Osborne-Mendel strain adult male rats, and in male and female dogs. Rats in groups of five received ground diets containing 500 ppm malathion, 100 ppm malathion, 25 ppm EPN, 5 ppm EPN, 25 ppm EPN with 500 ppm malathion, or 5 ppm EPN with 100 ppm malathion. During a 4-week pretreatment period, five whole blood ChE activity determinations were done on each rat. One group was kept as a control. ChE activity of whole blood was measured on each animal after 1, 2, 4, 6, and 8 weeks on the test diet. Marked ChE inhibition (21% of pretreatment level) was observed in the group fed the combination of 25 ppm EPN and 500 ppm malathion, and minimal but significant inhibition occurred in the 25-ppm EPN group.

One dog of each sex was placed for 12 weeks on a diet containing 250 ppm of malathion, 100 ppm of malathion, 25 ppm of malathion, 50 ppm of malathion, 20 ppm of EPN with 100 ppm of malathion, or 3 ppm of EPN with 8 ppm of malathion, followed by a control diet for 8 additional weeks. Two dogs of each sex were maintained as controls throughout the experiment.

Both plasma and erythrocyte ChE determinations were made on five blood samples drawn during the course of 4 pretreatment weeks, and after 1, 2, 4, 5, 6, 8, 10, and 12 weeks on the experimental diet and 3 and 8 weeks on the terminal control diet. Only slight, but significant, inhibition of erythrocyte ChE was found at the 250-ppm dietary level, while 250 ppm of malathion and 50 ppm of EPN together caused up to 60% inhibition of plasma ChE and 93% inhibition of erythrocyte ChE activities. A combination of 100 ppm of malathion and 20 ppm of EPN caused questionable inhibition of plasma ChE, but up to 68% inhibition of erythrocyte ChE. Up to 24% inhibition of erythrocyte ChE was noted after 8 weeks on the diet containing 8 ppm of malathion and 3 ppm of EPN, but the ChE activity of the erythrocytes returned incompletely to pretreatment levels during the recovery period, thereby complicating the interpretation of the data. No animals exhibited gross manifestations of poisoning during the experiment.

Shortly thereafter, Cook et al [94] and Murphy and DuBois [78] demonstrated the capacity of the liver in vitro to alter malathion to a form not susceptible to conversion to malaoxon or any other active inhibitor of ChE. This activity was inhibited by EPN and Diazinon more markedly than by any other organophosphate compounds tested. Murphy and DuBois [78] also demonstrated that single ip doses of 13 mg/kg of EPN (1/2 LD50) clearly reduced the capacity of male and female Sprague-Dawley rats to detoxify malaoxon. This suggested that EPN increased the toxicity of malathion by inhibiting the enzymes responsible for its detoxification. Principal among these enzymes are the carboxyesterases. Cook et al [94] found that a number of organophosphorus compounds, after oxidation with bromine if they were thiophosphates, were inhibitors not only of ChE but

also of malathionase; here, EPN and Diazinon were particularly effective. Cook and Yip [74] showed that the change in the constitution of malathion by malathionase was caused by the formation of a monoacid homolog in the removal of one alcoholic residue from the diethylsuccinate portion of the malathion molecule. DuBois [80] found that when 50 compounds being tested for use as pesticides were given simultaneously in pairs to rats, potentiation occurred with four pairs. Among these were malathion with EPN, Dipterex, or Co-Ral. Certain other organophosphorus pesticides, such as Ronnel, [91] Delnav, [91] Dipterex, and Baytex, [79,88,95-98] and drugs of the phenothiazine family, [99] when administered for a period of days before exposure to malathion, have been found to potentiate the toxicity of malathion.

In addition to their studies on human liver homogenates, which were discussed in the preceding section, Main and Braid [62] analyzed the in vivo inhibition of serum aliesterase by malathion in male Sprague-Dawley rats. Initial serum aliesterase activities were measured, and single oral doses of 1,250, 1,500, or 1,700 mg/kg of 96.8% malathion were administered. Aliesterase activity was measured colorimetrically (pH 6.3 and 25 C) at 0.5, 1, 2, 3, 4, and 5 hours after dosing. The results are shown in Table III-3. The data suggest that high doses of malathion inhibit aliesterase, an enzyme which detoxifies it in vivo. The data may have been influenced by an epizootic infestation in the animal colony, but this was not experimentally verified.

Male rats (weighing 200 ± 50 g) [62] were administered practical grade tri-o-tolyl phosphate (TOTP) in single oral 0.5-mg/kg doses. Serum, erythrocyte, and brain ChE, and liver and serum aliesterase activities were

TABLE III-3

IN VIVO INHIBITION OF SERUM ALIESTERASE BY MALATHION IN RATS

Malathion (mg/kg)	Initial Activity (μ moles/ml of serum/min)	% Initial Activity Remaining					
		0.5 hr	1 hr	2 hr	3 hr	4 hr	5 hr
1,700	12.67	73.2	48.2	31.2	25.8	21.3	-
1,700	3.06	25.5	0.0	0.0 Death after 2 hr			
1,500	11.18	69	56.7	37.8	35.5	36.8	35.0
1,500	13.70	48	39.2	31.5	30.3	28.5	-
1,250	0.79	Death before 0.5 hr					
1,250	5.80	76.5	35.3	12.0	-	6.5	9.8

Adapted from Main and Braid [62]

measured during 5-25 hours. Serum aliesterase activity was reduced to zero within 60 minutes, and that of liver to 12% of its initial activity. After 24 hours, liver aliesterase activity was only 1.5% of the initial value. The effect of an identical TOTP pretreatment on the oral LD50 of 96.8% malathion was then examined in rats fasted for 24 hours before administration of malathion. Without TOTP, the oral LD50 of malathion was determined to be 1,600 mg/kg. This value fell to 35 mg/kg when malathion was given 1 hour after 0.5 mg/kg TOTP, and further still to 20 mg/kg when

malathion was administered 24 hours after the same dose of TOTP. While the eightyfold decrease in the LD50 of malathion cannot be attributed unequivocally to inhibition of the aliesterase, these data support this hypothesis by agreeing with the findings by Cook et al, [95] Murphy and DuBois, [78] and Frawley et al [86] of potentiation of the toxicity of malathion by EPN, demonstrated by these authors [78,86,94] to follow inhibition by EPN of carboxy-ester hydrolysis of malathion.

Welch and Coon [66] studied the effects of drug pretreatment on malathion toxicity. Young adult, unfasted, male Swiss-Webster mice (weighing 18-25 g) were pretreated for 4 days with SKF-525A, chlorcyclizine, phenobarbital, or cyclizine before administration of technical malathion (95%). Malathion (1,500 mg/kg) was given orally in corn oil 24 hours after the last pretreatment dose, which had been given in amounts equivalent to about 1% of body weight: chlorcyclizine, cyclizine, SKF-525A (each at 25 mg/kg, twice a day for 4 days), and phenobarbital (35 mg/kg, twice a day for 4 days). They concluded that mice pretreated with chlorcyclizine and phenobarbital were less susceptible to the lethal activity of malathion than control mice.

Interactions in which organophosphorus compounds affect the metabolism of other drugs or chemicals have not been reported to any great extent. Rosenberg and Coon [100] reported that malathion, along with OMPA, EPN, chlorthion, and phostex, but not DFP and TEPP, increased markedly the time during which a given dose of hexobarbital removes the ability of mice to right themselves from a position on one side. Malathion did not alter the toxicity of hexobarbital. Stevens et al [101] reported that several organophosphorus pesticides, including malathion, impaired the metabolism

of hexobarbital and aniline, as evaluated by hexobarbital sleeping time and para-hydroxylation of aniline. Male Swiss-Webster mice (25-35 g), Wistar rats (150-200 g), albino rabbits (900-1,200 g), and mongrel dogs (9-14 kg) were the experimental animals. Malathion (99.6%) dissolved in corn oil was administered orally at a dose of 5 ml/kg. First, the oral LD50 in mice was determined to be 5,896 μmoles/kg. Mice were then dosed with 1/8, 1/4, or 1/2 the LD50 of malathion. The hexobarbital sleeping time (HST) was measured at 30-minute intervals, beginning 1 hour after malathion administration. HST was designated as "the time interval elapsed from the loss to the recovery of the righting reflex after an ip injection of 100 mg/kg of hexobarbital sodium." [100] The results indicated that the pretreatment with malathion significantly increased the HST.

Stevens et al [101] reasoned that this increase in HST caused by malathion might be related to an interference with the metabolism of hexobarbital in the liver and demonstrated this through in vitro experiments. Malathion at a concentration of 2 x 10*5 M (* means to the negative power of) significantly inhibited hexobarbital metabolism by liver in vitro. No inhibition by 2 x 10*6 M malathion, however, was observed. Further experiments showed that malathion (0.01-M range) consistently inhibited the metabolism of hexobarbital in all species (mice, rats, rabbits, dogs, and humans) studied. Stevens and Greene [102] were able to show that, despite the apparent interference with the metabolism of hexobarbital by malathion, such effects were not correlated highly with effects of this compound on the oxidation of nicotinamide adenine dinucleotide phosphate (reduced form), reduction of cytochrome C, or reduction of cytochrome P-450 in liver microsomal preparations in vitro.

It is likely, therefore, that at least some of the metabolism of hexobarbital is performed by other than the mixed-function oxidase system.

Certain factors other than drug or chemical interactions have been shown to influence malathion toxicity. Brodeur and DuBois [103] studied the differences between mature and immature rats in malathion toxicity. Twenty 23-day-old weanling (50-60 g) and 18 adult male (200-300 g) Holtzman rats were used. Undiluted malathion was administered to the adults and was dissolved in a mixture of 20% ethanol and 80% propylene glycol for administration to weanlings. The LD50 of malathion was determined to be 340 mg/kg for weanling rats and 750 mg/kg for adults. Death or complete recovery occurred within the first 7 days after doses were administered. In a later study, Brodeur and DuBois [104] further investigated the previously documented [103] observation that weanlings were more susceptible than adult rats to malathion. The metabolism in the liver involves both the conversion of malathion to malaoxon, which is the active metabolite responsible for ChE inhibition, and splitting off by hydrolysis of one of the alcoholic residues in the diethylsuccinate portion of the molecule to produce a molecule that cannot be activated by the replacement of S with O. [74] Brodeur and DuBois [104] concluded that the age difference in susceptibility was due to relatively low levels of malathionase in the liver and other organs (especially, perhaps, brain) in young rats.

Weanling rats (23 days old, 50-60 g) and adult male and female rats (200-300 g and 175-250 g, respectively) were injected ip with malathion dissolved in a mixture of 20% ethanol and 80% propylene glycol. [104] Malathion was administered in amounts equivalent to 0.2% and 0.1% of

weanling and adult body weights, respectively. ChE activity was determined by the method of DuBois and Mangun. [84] The livers of the males were 2.5 times more active than those of the females in hydrolyzing malaoxon. Also, the detoxification process was more active in adults than in weanlings and young rats.

In addition, studies [104] indicated that ChE activity was decreased to 37 or 28% of normal in weanlings by 100 or 150 mg/kg of malathion. In adults, the same degree of inhibition resulted only after doses of 600 mg/kg. The LD50 for young, 12-day-old rats was 125 mg/kg; for adults, it was 900 mg/kg. Eighteen-day-old rats tolerated 100 mg/kg of malathion; the LD50 was 200 mg/kg. Thirty-day-old rats tolerated 400 mg/kg with little ChE depression. The LD50 for the 30-day-old rats was 600 mg/kg, and 800 mg/kg for the 42-day-old rats.

The effect of dietary protein on malathion toxicity has been studied by a number of investigators, among them Boyd et al. [105] At weaning, male albino rats were put on diets containing various amounts of casein for 28 days. Oral LD50's of malathion were then determined for each group as follows: Group I - 51 rats, 0% casein, LD50 539 ± 42 mg/kg; Group II - 110 rats, 3.5% casein, LD50 599 ± 138 mg/kg; Group III - 52 rats, 9% casein, LD50 759 ± 91 mg/kg; Group IV - 108 rats, 26% casein, LD50 1,401 ± 99 mg/kg; Group V - 51 rats, 81% casein, LD50 649 ± 51 mg/kg. Group IV (26% casein) represented normal dietary protein intake. The authors noted that prior to inclusion of malathion in the diet, the highest protein intake (Group V, 81% casein) had the following effects on the rats: diarrhea, diuresis, polydipsia, and other unspecified effects. The highest casein concentration in the diet may have produced an unappetizing diet. The mean

weight of the 28-day-old rats in Group V was only 70% of that of the rats in Group IV. This diet was noted also to cause definite pathognomonic changes, eg, renal capillary congestion and diarrhea, among others. The postulated inappetence and the observed diarrhea both would operate to decrease the absorption of protein into the body from the diet of Group V. Boyd et al [106] concluded that the toxicity of malathion was inversely proportional to dietary protein intake, except at extremely high protein levels.

Boyd and Tanikella [106] administered technical malathion (95%) in cottonseed oil by intragastric intubation to young male albino rats to determine the oral LD50. The rats previously had been fed normal (Group II, 26% casein) or low-protein (Group III, 3.5% casein) diets on days 28-56 of age or until they weighed 5-10% more than Group I rats, which were on a lab chow diet. At the end of the modified-diet feeding period, malathion was administered in single oral lethal doses (predetermined in a pilot study) of 700-1,400 mg/kg to Group I; 1,000-2,000 mg/kg to Group II; and 200-1,200 mg/kg to Group III. Each dose was given to 10 rats on the experimental diets, with 15-45 controls receiving cottonseed oil only. LD50's were determined to be 1,090 ± 83 mg/kg for Group I; 1,041 ± 99 mg/kg for Group II; and 599 ± 138 mg/kg for Group III. The authors concluded that the twofold increase in malathion toxicity in Group III rats over that in Group II rats was due to their low-protein diet. The authors speculated that low-protein intake might result in hepatic enzyme activity reduction, thereby altering the animal's ability to detoxify malathion. This was not supported by either experimental data or theoretical considerations.

Marton et al [107] studied the effect of chronic malathion treatment on tolerance to cold. Sixty-eight Wistar rats were randomly divided into four groups of 17 each according to treatment and sex. The control group received Fox Chow. The other groups were given the same chow mixed with 95% technical grade malathion dissolved in corn oil. The concentration of malathion was 4,000 ppm, so that the daily intake of malathion approximated 240 mg/kg of body weight. After 5 months, during which malathion had little effect, the experimental groups were exposed to an environmental temperature of 1.5 ± 1 C. They were given water but no food. Blood ChE activity was determined 2-3 weeks prior to the experiment and again within 15 minutes after death. The rats fed malathion survived the cold for a significantly shorter period than did the control animals (P less than 0.01). ChE activity was also significantly lower in the malathion-fed animals but did not differ significantly between preexposure and cold exposure activities. Malathion-fed rats did not appear to have increased heat loss over the controls. Hence, the authors [107] inferred that malathion decreased the ability of rats to produce a high rate of heat continuously over a prolonged period of time.

Durham et al [108] stated that the muscular weakness seen in the legs of chickens appeared to be the best available index of the possible paralytic effects of organophosphorus compounds in humans. They tested nine organophosphorus compounds, including malathion, for toxicity in atropinized chickens. Immediate muscle weakness appeared in 6 of 10 chickens given one subcutaneous injection of 1,000 mg/kg of malathion. The lowest dose of malathion that produced immediate muscle weakness was 100 mg/kg. Although the weakness was reversible and disappeared completely

within 4-21 days, the authors did not state whether this reversibility was a dose-related effect. Utilizing methods similar to those of Durham et al, [108] Gaines [109] investigated the effects of 9 carbamates and 30 organophosphates (in peanut oil suspension or solution) in an unspecified number of chickens. Within a few hours after the subcutaneous injection of malathion in doses of 100 mg/kg or more, leg weakness was observed and it persisted for 4-14 days. The results of microscopic examinations of tissues were not reported. Frawley et al [110] fed malathion to chickens at levels up to 10,000 ppm for 15 weeks. All of the birds died, but only one exhibited muscle weakness, and none showed microscopic evidence of nerve damage. O'Brien [64] pointed out that, while many known neurotoxic compounds are inhibitors of plasma ChE, most inhibitors of this enzyme do not produce neurotoxic effects. Malathion appears to be a ChE inhibitor that is not neurotoxic and does not induce demyelination of long axons.

Krause et al [111] administered 40 mg/kg of malathion (purity unstated) in corn oil to male rats on days 4 and 5 of age and 20 mg/kg to another group on days 4-24. Controls and experimental animals received 0.1 ml of corn oil/5 g body weight at each dose. Two rats from each group were killed for microscopic examination of the testes at 6, 12, 18, 26, 34, or 50 days of age. From the results obtained with this rather limited numerical base, the authors concluded that the weight of the testes and the mean diameter of the seminiferous tubules, as well as the number of Leydig cells, were reduced at some time during the experiment. All effects, however, disappeared by the rats' 50th day of life. The lesions in the testes caused by malathion were evaluated as "not very serious," and were limited essentially to the period of time during which the compound was

administered, because of the rapid excretion of the compound. All cell counts were reported to be normal after the 24th day of life.

Mohn [112] applied 0.2 M malathion to Escherichia coli K-12/gal Rs,8, a phenotypic gal-, and incubated it for up to 300 minutes. The resulting cultures were examined for forward mutations to 5-methyl tryptophan (5-MT) resistant colonies. In these experiments, the author reported using a concentration of 20μg 5-MT/ml and an inoculum containing 300,000-500,000 cells/ml. Cultures with spontaneous mutation rates of less than 25/plate were used. The mean number of cell generations was 8-9 during residual growth. According to the author, the results with malathion did not differ significantly from spontaneous values, even when 70% of the cells were inactivated. Malaoxon was not examined.

A study by Huang [113] also failed to reveal a mutagenic potential in malathion. Eighty thousand cells/ml from three different human hematopoietic cell lines were treated with malathion dissolved in dimethyl sulfoxide at concentrations of 50 and 100 μg/ml. Cells were harvested 6, 12, 24, and 50 hours after exposure to malathion. Colcemid at a concentration of 0.04 μg/ml was added to the cultures 2 hours before harvesting for chromosome preparation. [113] Cells were treated with 1% sodium citrate and fixed in acid-methanol (1:3). Flame-dried slides were stained with Giemsa's stain, and 100 metaphase figures were studied with an oil-immersion objective. Clear chromosome lesions (gaps, breaks, exchanges, dicentrics, pulverization, etc) were recorded for each group. Cell growth was inhibited, but returned to normal in cells placed in malathion at a concentration of 50 μg/ml after they were washed with fresh, pesticide-free medium. No resumption of growth was noted in cells placed

in malathion at a concentration of 100 μg/ml. Single gaps and breaks were found in 4% of the controls at 6 hours, 3% at 12 hours, 3% at 24 hours, and 2% at 50 hours. Similar counts had values of 2, 3, 5, and 0% at the same time periods for the 50-μg/ml concentration and of 5, 4, 5, and 5% for the 100-μg/ml concentration. These results, however, are contrary to the findings of Trinh Van Bao et al [54] reported in Effects on Humans.

The use of submammalian test systems and in vitro cell cultures should be regarded only as ancillary procedures to supplement mutagenic studies using intact mammalian test systems, according to a recommendation by a WHO Scientific Group in its 1971 report entitled Evaluation and Testing of Drugs for Mutagenicity: Principles and Problems. [114] NIOSH agrees with the general philosophy of this recommendation.

Kimbrough and Gaines [115] studied the possible teratogenic effects of several organophosphorus pesticides, including malathion, on rat fetuses after a single ip injection into the mother. Six dams were injected ip with 600 mg/kg of malathion and six others with 900 mg/kg on the 11th day of their pregnancy. The fetuses were removed on the 20th day of pregnancy. The higher dose of malathion produced a slight tendency for the weights of the placentas and the fetuses to be reduced and one instance of adactylia among 67 fetuses. The lower dose produced no evident effects. It should be noted that administration of the pesticide in a single dose on day 11 of gestation may have resulted in less teratogenic effect than either repeated or earlier doses.

Dobbins [116] conducted a screening study of the teratogenic potential of various malathion doses in 15 pregnant Wistar rats. The drug was dissolved in corn oil and administered by stomach tube on day 9 or 10,

or from days 8 to 12 or 12 to 15, of pregnancy. Seven control rats were untreated. On day 20 of gestation, the dams were killed and the fetuses examined. A suggestion of teratogenic effect on the urinary system was noted, but the data and the experimental detail were confused and insufficient for any conclusion. None of 74 control fetuses was considered to be abnormal; the usual rate of spontaneous abnormalities would be about 1%.

Lillie [117] fed seven groups of 20 Leghorn pullets a breeder diet containing either 0, 250, or 500 ppm of malathion, 250 or 500 ppm of carbaryl, 250 ppm each of malathion and carbaryl, or 500 ppm each of both pesticides. Observations during a 36-week period included changes in body weight, egg production, egg weight and specific gravity, feed consumption, mortality, fertility, hatchability, embryonic abnormalities, and performance of progeny. In the studies of performance by progeny, chicks from hens fed 0 or 500 ppm of malathion or malathion and carbaryl were fed a broiler diet supplemented with 0 or 500 ppm malathion or carbaryl for 4 weeks. The only significant effect was a smaller weight gain in adult birds fed the mixture of carbaryl and malathion. Decreased growth was observed in the progeny fed carbaryl, irrespective of maternal diet, but it was not seen in those fed malathion. In a separate 4-week study, [117] the incorporation of 500 ppm of malathion, of carbaryl, or of both, into the diet of Leghorn cocks produced no significant changes in fertility pattern, production of sperm, or embryonic abnormalities.

Greenberg and LaHam [118] injected malathion at concentrations of 3.99 or 6.42 mg/egg into the yolk sacs of 50 hens' eggs incubated for 4 or 5 days. Twenty-five control eggs were used. The eggs injected with

malathion produced chicks exhibiting sparse plumage, micromelia, overall growth retardation, and beak defects. The authors speculated that the observed abnormalities could be attributed to an in vivo retardation of cell growth and protein synthesis, but they did not provide any experimental support for this contention.

Ho and Gibson [119] injected hen's eggs with 0.1 ml of 2% malathion in corn oil on the 5th day of incubation. The treated embryos showed a generalized reduction in body size, delayed patterns of mineralization in certain endochondral bones, and micromelia. Tibiotarsi consistently exhibited retarded growth, cartilage necroses, and angulation. In another paper, [120] Greenburg and LaHam used the technique of injecting into the yolk sacs of 4- or 5-day incubated hens' eggs 6.42 mg/egg of malathion either alone or with various amino acids, vitamins, and nicotinamide adenine dinucleotide precursors to attempt to find means for preventing the malformations and retardation of growth induced by malathion. Nicotinamide (5 mg), nicotinic acid (5 mg), and quinolinic acid (4.3 mg) prevented the malformations, but growth was still stunted. Tryptophan (5.1 mg) was the only compound of 32 tested that prevented both malformation and growth retardation. Indole, indoleacetic acid, and serotonin had no antagonistic value. Indeed, in two of four experiments, indole had a more marked effect than malathion itself, and indoleacetic acid enhanced the effects of malathion. The authors suggested, therefore, that malathion may have had no direct metabolic effect on the egg but rather may have disturbed the electronic milieu within the egg, and that tryptophan nullified this effect by virtue of its particular ionization potential. Compounds having an ionization potential near that of tryptophan (adenine, guanine, alpha-

naphthol, beta-naphthol, indoleacetic acid, and quinoline) by themselves had no effect on the embryos, but did have a magnifying effect when given with malathion. Imidazole, with an ionization potential quite different from that of tryptophan, was neither therapeutic nor synergistic with malathion. The ionization potential appears, therefore, not to be the basis of either the ovotoxic effect of malathion or the protective action of tryptophan.

Wilson and Walker [121] reported that malathion in concentrations greater than 1 μg/ml was toxic to primary cultures of chick embryo fibroblasts.

While many of the basic concepts of teratology were initially investigated using avian eggs, the absence of anatomic and physiologic maternal-fetal relationships during incubation makes it difficult to extrapolate from the effects on the avian egg to teratogenic hazards to humans. The use of mammalian species for evaluation of possible teratogenic hazard is now recommended by the WHO Scientific Group on the Principles for the Testing of Drugs for Teratogenicity. [122] Because of the difference between the conditions within an egg and within the amniotic sac of mammals, the production of terata within eggs indicates a teratogenic potential but does not necessarily mean that mammals will be likely to develop terata on exposure of pregnant females to the same influence.

In a study using malathion administered in the diet at a rate of approximately 240 mg/kg/day, Kalow and Marton [123] found no effects attributable to malathion in the reproductive performance of 18 female and 12 male experimental rats mated after 10 weeks of feeding on the malathion-

containing diet compared with 18 female and 12 male control animals. However, the survival to weaning of the offspring of animals fed malathion was only 31.8% versus 64.7% in the control group. The mean gain in body weight of offspring from the malathion-fed rats was only about 85% of that of control offspring from weaning to 9 weeks of age. The authors suggested that this effect may have been due to a lowered resistance of the treated animals to infection or "other noxious influences" in the colony and not necessarily to ChE inhibition.

The effects of malathion on the reproductive capacity of rats in a three-generation reproduction study have been reported. [124,125] The test material (95% pure) was incorporated into the diets of all three generations of rats in concentrations of 100, 500, or 2,500 ppm (approximately 5, 25, or 125 mg/kg/day). Sixteen pairs of rats in each group were mated, beginning with the original weanling rats, as were an equal number of untreated paired controls. Signs of ChE inhibition were not present. The following effects ascribed to respiratory disease were observed in the 2,500-ppm group: death of 4 of 16 dams; lower ratios of litters to pregnancies and of pups weaned to pups aged 5 days plus lower pup weights in all 3 generations. Both matings of the second generation on the 2,500-ppm diet produced pups of lower body weight and reduced ratios of pups weaned to pups at 5 days, relative to the controls. The authors [124,125] suggested that this may have been due to a change of bedding material which, in turn, induced respiratory distress. The fertility of the rats used in the final mating at the 2,500-ppm level also was reduced, but no malformations in the pups were observed. Microscopic examinations of the pups from the third generation, first mating, of the control and

high-level groups revealed similar minor lesions. The data revealed no adverse effects at the 100- and 500-ppm levels. However, the respiratory distress, reduced fertility, reduced pup weights, and poor pup survival at the 2,500-ppm level may have been due to malathion.

Correlation of Exposure and Effect

A number of biologic effects associated with malathion exposure have been documented in humans and experimental animals. The effects of exposure are the complex group of signs and symptoms which result to an unknown extent from the accumulation of ACh at various neural synapses and motor endplates. These signs and symptoms are discussed in detail in Effects on Humans and are tabulated in Table III-2. [48,49] Death due to malathion intoxication has been attributed to depression of the respiratory center of the brain, [126] presumably as a result of accumulation of ACh.

In humans, ingestion of malathion has been shown to produce all of the signs and symptoms of cholinergic stimulation up to and including death. Nalin [50] reviewed 264 reports of attempted suicide collected over a period of 3 years in Guyana. All patients experienced signs and symptoms consistent with cholinergic stimulation, and 53 individuals (20% of the total) died.

Eight case study reports [19,20,25,39,44-46] found in the literature specified the amount of malathion ingested and the characteristic manifestations of cholinergic stimulation observed. In the first case, [20] a 14-year-old boy weighing 165 lb ingested approximately 4 ounces (approximately 118 ml) of malathion (purity not reported). The authors calculated this to be a dose of approximately 0.4 g/kg of body weight. The

victim exhibited a wide range of severe manifestations consistent with cholinergic stimulation. He was comatose and recovered only after intensive therapy. In another case, a 45-year-old man ingested 50-90 ml of 50% malathion in a petroleum hydrocarbon base. Although the patient's actual weight was not reported, assuming one of 70 kg, a maximum dose of approximately 0.8 g/kg can be calculated. The patient experienced the full range of cholinergic symptoms, including marked respiratory insufficiency requiring tracheostomy and mechanically assisted ventilation, cardiac arrhythmia, and unconsciousness. He survived only with intensive and prolonged therapy. In a third incident, [39] a 42-year-old woman ingested 120 ml of 50% malathion garden spray solution. Goldin et al [39] estimated the maximum intake at 1.0 g/kg. The victim displayed signs consistent with cholinergic stimulation, including coma and profound shock. After intensive therapy, she survived without sequelae. In five similar cases, [25,44-46] the estimated doses consumed, which resulted in severe poisoning that would have been fatal but for intensive therapy, were in the order of 0.5, 0.9, 0.7, 0.5, and 0.6 mg/kg. From the extremely grave clinical picture exhibited by these patients, it is clear that all had absorbed doses of malathion which would have been lethal without intensive and prolonged therapy. Based on these cases, each of which was stipulated by the authors to have been nearly fatal, it appears that without treatment, the acute oral lethal dose of malathion in humans would be somewhat below 1.0 g/kg.

In a human experimental study, [21] five men ingested 16 mg of malathion/day for 47 days. The malathion was administered as a corn oil solution in gelatin capsules. The subjects exhibited no clinical effects

and reported no subjective complaints during or after the study. There was no drop in blood ChE activity. If an average weight of 70 kg is assumed, the approximate daily dose for these men was 0.22 mg/kg.

In a similar study, [21] the same authors administered malathion at a daily oral dose of 24 mg to five men for 56 days. This dose would amount to 0.34 mg/kg, assuming an average weight of 70 kg. Although no clinical signs or symptoms of poisoning were observed, a statistically significant drop in plasma ChE activity was noted after 2 weeks. The maximum depression of 25% occurred after 3 weeks. However, the activity returned to normal throughout the remaining 5 weeks of the experiment. At 7 weeks, there was also a drop in erythrocyte ChE activity. This reached a maximum 2 weeks after completion of administration of malathion.

There are very few human data available that give a quantitative relationship between respiratory exposure to malathion and biologic effects. In Golz's study, [56] four volunteers were exposed to malathion in a room in which an aerosol was dispersed at the beginning of a 1-hour exposure period. The maximum theoretical air concentration at the beginning of the exposure period was approximately 2.4 g/1,000 cu ft, or approximately 85 mg/cu m of malathion. The true concentration to which the men were exposed probably was smaller than this since the aerosolized formulation may have settled out within a short time following its generation. The subjects were examined weekly during the 42-day period of twice-daily exposures, and no signs or symptoms consistent with excess ACh accumulation were observed. Several studies [35-37] have been performed in which respiratory exposure to malathion during actual application was estimated. In cases in which workers were exposed to malathion during its

application in field use, the respiratory rate of exposure ranged from approximately 0.01 to 1.23 mg/hour. The skin exposure rate was 10-100 times the respiratory rate, that is, the amount of malathion which could have been deposited was 10-100 times the amount they inhaled. In none of these studies [35-37] did the investigators look for signs or symptoms of poisoning related to the exposure, but it is reasonable to assume that, had there been any significant effects, they would have been reported.

Signs and symptoms of cholinergic stimulation attributed to absorption of malathion through the skin have been reported. [24] An 8-year-old girl developed signs and symptoms consistent with ACh accumulation after having her hair washed with a 50% malathion-xylene solution. She died 5 days after the hairwashing. No quantitative estimate of the malathion exposure can be made in this case. In addition, Quinby and Lemmon [127] reported a case study of clinical poisoning in a group of field workers who were exposed to malathion residues on crop foliage. Again, no quantitative estimate of the exposure is possible.

After iv injection of a dose of 14C-malathion into an unspecified number of normal male volunteers, [128] 90.2% of the label was recovered in the urine with a half-time of excretion of 3 hours. Using these figures to correct measured absorption from the skin, Feldmann and Maibach [128] found that 12 subjects absorbed an average of 6.8 ± 2.3% of malathion applied to the skin of the forearm during a period of 5 days after its application. In another study, [129] the skin of the axilla absorbed 4.2 times as much malathion as that of the forearm — the highest absorption measured with malathion. (Scrotal skin was not used.) Therefore, the maximum absorption (outside of the scrotum) was 28.5% of that applied to the skin.

The effects of malathion on laboratory animals are similar to those in man. Acute doses of malathion produce signs of anticholinesterase effects in dogs, rats, mice, and rabbits, [10,33,83] indicating a common analogous mechanism of action in these species.

Laboratory mammals can tolerate large quantities of malathion when it is absorbed at a moderate rate. [124,125] During the three-generation study, [124,125] rats survived average daily intakes of 125 mg/kg/day or 45 g/kg each year.

Animal experiments indicate that the dermal application of malathion can cause cholinergic effects and death. The transdermal LD50 of malathion in rats has been reported as greater than 4,400 mg/kg. [109] This dose is approximately 3-6 times larger than the oral LD50's in this species.

The study by Weeks et al [83] showed malathion aerosols to have no effect on blood ChE activity in concentrations of 6-8 mg/cu m in either rabbits or quail, and no effects at up to about 30 mg/cu m in rabbits or lasting effects (beyond 24 hours) in quail. The formulations tested were technical malathion (95% pure) and 6% malathion in No. 2 fuel oil. Particle size had no effect on toxicity, and the fuel oil component was found to be of greater toxic potential than malathion in the 6% formulation.

There are several experimental investigations whose results suggest a level of dermal exposure to malathion in humans below which no cholinergic effects can be detected. In three separate studies, [35-37] the dermal exposure of individuals during malathion application was estimated by using absorbent pads taped to various areas of the skin or clothing. Estimated dermal exposures ranged from 0.25 mg/hour to an extreme high of 194

mg/hour. These individuals were also exposed via the respiratory route. In none of the subjects were signs and symptoms of cholinergic stimulation looked for, but it may be assumed that any significant complaints would have been noted by the investigators. In another group of studies, [32,129,130] volunteers were dusted daily with powdered formulations containing various concentrations of malathion. Hayes et al [32] reported no significant changes in ChE activities in 60 male volunteers exposed to dermal doses of malathion contained in talc dusting powder during an 8- to 16-week period at levels sufficient to lead to urinary excretion of malathion-derived materials equivalent to a minimum of about 70 mg of malathion/day. Using Feldmann and Maibach's [128] figure for recovery of 14C administered as malathion, the gross values from this study indicate that about 78 mg of malathion were absorbed from the application of 2.8 g of malathion powder to the skin. This absorbed quantity of malathion may be regarded, therefore, as a no-effect or safe dose.

In summary, malathion can cause signs and symptoms in humans resulting presumably from accumulation of ACh at various effector sites after absorption. The approximate single oral lethal dose in humans is probably somewhat below 1.0 g/kg. Although respiratory and dermal exposures to malathion have been reported to cause clinical effects consistent with pseudocholinergic (anticholinesterase) activity, no clear-cut cases of fatal poisoning by these routes of entry have been reported, and no quantitative relationship between exposure and cholinergic effect via these routes has been established. It is evident that a brief exposure to atmospheric concentrations as high as 85 mg/cu m [56] and dermal exposure [55] sufficient to lead to urinary excretion of malathion-derived

materials equivalent to a minimum of about 48 mg of malathion/day, as determined by ether extraction (78 mg/day total), caused no significant depression of ChE activity.

No findings of carcinogenesis by malathion have been reported. There is some information on its possible teratogenic and mutagenic activities. [17,54,111-113,115-119,123-125] Because some of the papers on teratogenic activity are derived from studies on hens' eggs and some of those on mutagenesis relate to studies with unicellular organisms, little information applicable directly to mammals is available in this general field. The data are judged to be insufficient to establish the existence of significant risk of the occurrence of these effects in human populations exposed to malathion.