Blood Cholinesterases, Serum Parathion Concentrations and Urine p-Nitrophenol Concentrations in Exposed Individuals[1]

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Depressions of plasma and/or erythrocyte cholinesterases have been regarded as confirming contact with, and providing absorption indices for, cholinesterase inhibiting pesticides such as the organic phosphates and carbamates. (1) Although depressions of such enzymes can also be produced by certain drugs, and in the case of the plasma enzyme may also reflect liver disease such situations are, according to Hayes (2) not consistent with active work programs.

From the general acceptance of assays of blood cholinesterases as indices of absorption for this class of pesticides it seems reasonable to assume that the concentrations of such substances in the blood should be relatable to depressed enzyme activities. Feeding experiments with parathion such as those referred to by Ganelin (3) and Edson (4), have

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[1] Contribution No. 1469, Department of Entomology, Arizona Agricultural Experiment Station. This research was supported under contract no. PH-86-65-84 by the Pesticide Program, U. S. Department of Health, Education and Welfare.

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Bulletin of Environmental Contamination & Toxicology,
Vol. 4, No. 6, 1969, published by Springer-Verlag New York Inc.

produced marked cholinesterase depressions in experimental animals. Edson (4) indicates that oral intake of parathion in man has the greatest effect on the plasma enzyme. Recent investigation of two episodes of acute intoxications in man (5) have indicated that marked depressions of blood cholinesterases were associated with the detection of cholinesterase inhibiting pesticides in the patient's serum. Nabb, et al. (6) in investigating dermal exposures of human volunteers found considerable amounts of p-nitrophenol excreted via the urine but did not detect any significant cholinesterase depressions. Funckes, et al. (7) in similar studies, using respiratory exposures, found evidence of parathion absorption from the excretion of urinary p-nitrophenol and accompanying depressions of blood cholinesterases.

In the course of an intensive cholinesterase surveillance program of a number of cooperating aerial applicators in 1968 we noticed that several individuals with acceptable levels of erythrocyte and/or plasma cholinesterase had concentrations of methyl and ethyl parathion in their blood that appeared inconsistent with the observed cholinesterase values. In a period of two months during the 1968 agricultural season we were able to collect appropriate samples from cooperating aerial applicators (pilots, loaders and flaggers) to investigate this matter further.

Materials and Methods

Blood samples from cooperating aerial applicators were collected

via venipuncture during the most active spray season. Urine sample collections, while not initially a part of this program, were initiated for p-nitrophenol determinations.

Plasma and erythrocyte cholinesterase activities were determined by the method of Michel. (8) The urine p-nitrophenol concentrations were determined colorimetrically by the method of Elliot, et al. (9).

Serum samples were extracted by the Dale-Cueto (10) method and analyzed by gas liquid chromatographic (GLC) techniques employing both electron capture (EC) and flame photometric detectors (FPD). Details of the GLC procedures and operating parameters were as follows:

Equipment Microtek MT 220: Electron capture detector, 200 mc tritium foil: voltage 15 to 22.5 VDC. Carrier gas-nitrogen 60 ml/min. Inlet temp. 230, col. temp. 205, det. temp. 210. Column 6' x 1/4"; column packing 1.5% OV-17, 2% QF-1 on ABS chromosorb "W" 100/200 mesh.

Flame Photometric Detector. Hydrogen 140 ml/min, oxygen 20 ml/min. Carrier gas-nitrogen 80 ml/min. Column 8' x 1/4"; column packing 1.5% OV-17, 2% QF-1 on ABS chromosorb "W" 100/200 mesh.

On column injection was employed for these tests.

Initial detection and quantification was accomplished via the EC detector with confirmation by the FPD operating in the phosphorus mode and also on repeat analyses with the FPD in the sulfur mode.

Results and Discussion

Results of the analyses of blood and urine samples collected from 23 individuals are presented in Table 1. Examination of these data reveals that pesticide concentration is well correlated with urine p-nitrophenol concentrations (r-0.56). This finding reassures us that our analysis of serum parathions is indeed qualitatively and quantitatively correct. No significant correlation exists between serum parathions and cholinesterase levels or between cholinesterase levels and urinary paranitrophenol excretion.

Because of the variation of cholinesterase activities among individuals and within individuals we have employed a critical value for the plasma enzyme of 0.40 Δ pH units as the point at which we advise applicator personnel to review their safety programs. Using this criteria we would have detected only 52% of the apparent failures of safety programs by relying on plasma cholinesterase values. These data suggest that erythrocyte and/or plasma cholinesterase values are not as sensitive as indices of ethyl or methyl parathion absorption as are the concentrations of these compounds in serum. Studies by Elliot et al. (9) suggest that assay of urinary p-nitrophenol is more sensitive as an absorption index than blood cholinesterase. The present data indicate that assay of serum samples via GLC are even more sensitive than urine p-nitrophenol measurements.

Four individuals, two agricultural pilots and two loaders, working

TABLE 1

Blood Cholinesterase Values, Serum Ethyl and Methyl Parathion Concentrations and Urine p-Nitrophenol Concentrations

Cholinesterase pH		Serum Ethyl and Methyl Parathion	Urine p-Nitrophenol
RBC	Plasma	ppb	ppm
.54	.34	12.9	1.1
.75	.39	21.9	1.0
.54	.30	12.4	1.2
.40	.39	15.3	2.2
.65	.28	35.7	0.9
.67	.24	54.7	3.9
.92	.35	20.3	1.1
.99	.29	15.1	1.8
.57	.17	199.5	2.0
.79	.39	16.9	2.2
.98	.36	16.9	0.7
.67	.18	33.4	2.2
.73	.40	84.7	3.6
.56	.53	58.8	12.3
.62	.57	52.4	2.8
.70	.60	64.3	8.5
.59	.47	11.4	0.9
.78	.64	145.3	13.2
.57	.52	106.1	11.9
.64	.45	6.9	0.4
.71	.66	4.0	0.5
.71	.59	2.6	0.5
.34	.52	21.2	3.8

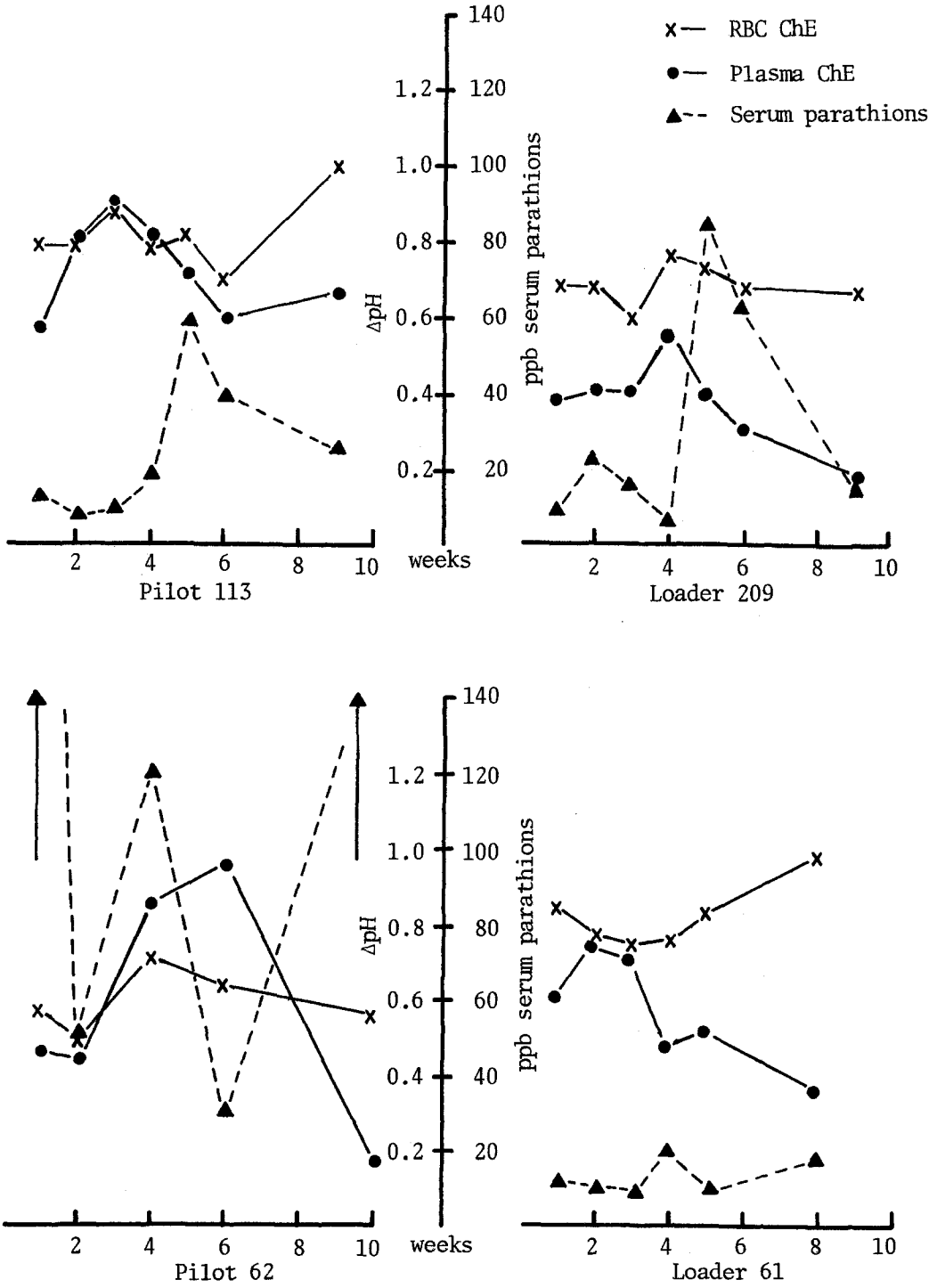

Figure 1 Comparisons of Blood Cholinesterases and Serum Parathion Concentrations

with these pilots are of particular interest. (Figure 1) These individuals have been employed in the aerial agricultural chemical business from 5 to 20 years. During the period of the present study all were without signs or symptoms of intoxication despite the relatively high concentrations of parathions in their serum. One loader, in addition, showed a very low plasma cholinesterase activity. The extreme variations in cholinesterase activity among individuals and with time in an individual are apparent in Figure 1.

Pilot 62 is unique in the extreme variability of ethyl and methyl parathion concentrations in his serum and the occasionally very high concentrations of these pesticides.

Although the serum concentrations of ethyl and methyl parathion in three of these individuals are equal to or higher than that observed in at least one recent case of acute intoxication investigated by our laboratories they are not inconsistent with the known work practices of these individuals. We suggest, therefore, that there are extreme differences in individual susceptibility to the actions of organic phosphates. From a practical standpoint, this compromises the utility of cholinesterase determinations as indices of absorption for evaluation of safe work practices.

We did not detect any of the oxygen analogs in samples from these cooperators, although we have, from time to time, detected paraoxon in other serum samples. We feel that the extraction procedures employed

are not adequate for the isolation of the oxygen analogs or other more polar organic phosphorus compounds that are being more extensively used.

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