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Diazinon Toxicity—Comparative Studies in Dogs and Miniature Swine[1]

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Diazinon Toxicity—Comparative Studies in Dogs and Miniature Swine. EARL, F. L., MELVEGER, BARBARA E., REINWALL, JOHN E., BIERBOWER, GEORGE W., and CURTIS, JACK M. (1971). *Toxicol. Appl. Pharmacol.* **18**, 285–295. Beagle dogs and Hormel-Hanford swine survived single oral doses of 300 or 500 mg/kg of diazinon. In addition, 4 daily doses of 50 mg/kg/day were tolerated. Dogs survived as long as 15 days and swine up to 30 days at doses of 25 mg/kg/day, although 2 female pigs died after 2 and 3 similar daily doses.

In a chronic study, 30 beagle dogs and 30 Hormel-Hanford swine were distributed into 5 groups of 6 animals each. Diazinon, dissolved in corn oil, was given by capsule to the dogs at doses of 0, 2.5, 5.0, 10.0, and 20 mg/kg/day and to the swine at doses of 0, 1.25, 2.5, 5.0, and 10 mg/kg/day for 8 months.

Consistent cholinergic signs were observed, primarily in the high-dose groups of both species. No marked changes in hematologic values were noted in these animals when measured at monthly intervals. Among the blood chemistry determinations made, increases with respect to control values were observed in the serum levels of amylase, lactic dehydrogenase, and ornithine carbamyl transferase. These responses were not sex-related.

In the dose-range studies, gross pathologic findings included hemorrhages of the heart and congestion or hemorrhage of the gastrointestinal tract, along with marked edematous thickening of the duodenum and jejunum in some animals. In the chronic studies, thickening and occasional rupture of the intestinal wall were observed. In swine, ulcers were found in the duodenum and the livers were firm, gritty, and hard to cut. Histopathologically, slight cirrhotic changes of the liver and hemorrhage of the intestinal tract were seen most frequently. In 2 of the 3 male dogs in the high-dose group, testicular atrophy was observed.

Myeloid:erythroid (M/E) ratios in excess of 100/1 were found in dogs which received 20 mg/kg/day of diazinon and died during the first 30 days of the experiment. Three of four pigs receiving 10 mg/kg/day of diazinon died in the same period and had an M/E ratio of 3.4/1 or more. Reticulocytopenia was found in both species, although classical aplastic anemia was not produced in either species.

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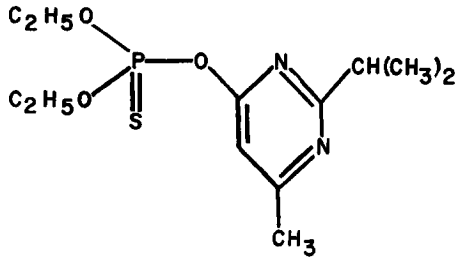
Diazinon (*O*,*O*-diethyl *O*-(2-isopropyl-4-methyl-6-pyrimidinyl)phosphorothioate), shown in Fig. 1, is a broad-spectrum pesticide effective against approximately 120 species of insects and pests. It is metabolized into monothionotetraethyl pyrophosphate, dithionotetraethyl pyrophosphate, and triethylthionophosphate; all these compounds are powerful cholinesterase inhibitors producing nausea, vomiting, fasciculation with muscular twitching, paralysis, and death. Diazinon is toxic not only by ingestion, but also by skin contact.

Diazinon was alleged to have caused aplastic anemia in a child (Private communication, 1965).[5] The unpublished work with dogs as quoted by Lehman (1965) and the work of Williams *et al.* (1959) referred only to the inhibition of cholinesterase activity.

DIAZINON



O, O-diethyl O-(2 isopropyl-4-methyl-6-pyrimidinyl) phosphorothioate

FIG. 1. Structural formula for diazinon.

Bruce *et al.* (1955) reported that the oral LD50 of diazinon for rats is approximately 125 mg/kg. These workers also conducted dog feeding studies in which diazinon 25% wettable powder was given in capsule form 6 days per week. Dosages varied from approximately 4.5 mg/kg/day, given chronically, up to 25 mg/kg/day for a period of 2 weeks. No gross or microscopic abnormalities were observed in the tissues examined, which included bone marrow. Cholinesterase inhibition was the only biochemical abnormality reported.

The purpose of this study was to obtain more information on the pharmacologic and toxicologic effects of diazinon in dogs and miniature swine, and to determine whether anemia could be produced in either species.

MATERIALS AND METHODS

Dose-Range Studies

To establish proper dosage levels for a chronic toxicity study, 16 beagle dogs, 6–19 months of age, were given capsules containing diazinon dissolved in corn oil. The dosage, number of daily doses, and survival or death losses of dogs are shown in Table 1.

[5] Food and Drug Administration (1965). U.S. Department of Health, Education, and Welfare, Washington, D.C. 20204.

TABLE 1

ORAL DOSE-RANGE STUDIES ON DIAZINON IN DOGS

Dog No.	Sex	Age/day	Dose (mg/kg/day)	No. of daily doses	Results
38B	M	575	5	69	Survived, killed
96B	M	339	5	69	Survived, killed
94B	F	339	10	69	Survived, killed
D2	F	194	10	148	Survived, killed
78B	M	342	25	10	Died
41E	F	325	25	15	Killed, moribund
A33	M	353	50	5	Survived
15E	F	477	50	5	Survived
A29	M	281	100	5	Died
81D	F	367	100	5	Survived
37E	M	323	300	1	Died
10F	F	320	300	3	Died
51F	M	220	400	1	Died
84E	F	240	400	1	Died
55F	M	217	500	1	Died
71E	F	421	500	1	Survived

Eighteen Hormel-Hanford miniature swine, 4–14 months of age, were used to determine the lethal and tolerated doses of diazinon dissolved in corn oil. The dosage, number of daily doses, and survival or death losses for swine are shown in Table 2.

TABLE 2

ORAL DOSE-RANGE STUDIES ON DIAZINON IN SWINE

Pig No.	Sex	Age/day	Dose (mg/kg/day)	No. of daily doses	Results
905	M	330	5	69	Survived, killed
925	M	326	5	69	Survived, killed
644	F	408	5	57	Killed, moribund
794	F	362	5	69	Survived, killed
892	M	286	10	9	Died
961	M	271	10	37	Killed, moribund
657	F	368	10	20	Killed, moribund
749	F	390	10	69	Survived, killed
883	M	285	25	30	Killed, moribund
923	M	268	25	21	Killed, moribund
826	F	265	25	3	Killed, moribund
835	F	269	25	2	Died
812	M	261	50	5	Died
855	M	256	100	4	Died
872	M	249	300	3	Died
1122	F	129	300	1	Died
861	M	253	500	1	Survived, killed
922	M	253	500	1	Died

Necropsies were performed on all animals, and histologic sections stained with hematoxylin and eosin were made on 9 of the dogs given 5–100 mg/kg and on 12 of the swine given 5–50 mg/kg that survived the longest period of time.

Chronic Studies

Dogs. Thirty purebred beagle dogs, all previously wormed and immunized against canine distemper, hepatitis, and rabies, were selected from the breeding colony of the Food and Drug Administration. The 15 males and 15 females, 7–9 months old and weighing 5.8–9.4 kg, were distributed in 5 groups for treatment according to littermates, weight, sex, and eating habits. They were housed in a kennel with automatic watering devices and runs for indoor and outdoor exercise.

The dogs were fed Purina Laboratory Chow at 9:00 AM daily and the feed pans were not removed until the following morning; at that time any food remaining was weighed and recorded. The diet was fed at a rate of 35 g/kg/day and the daily amount offered each dog was adjusted weekly according to its weight.

Technical grade diazinon[6] stabilized with 5% epoxol, was diluted 1:100 with Mazola corn oil and given daily at 10:00 AM by capsule to 4 groups of 3 male and 3 female dogs at dosages shown in the tabulation below for a period of 8 months. Six dogs were maintained as controls and were given capsules containing a dose of corn oil alone equivalent to that given group D; these dosages were also adjusted weekly according to the dog's weight.

	Diazinon (mg/kg/day)	
Group	Dogs	Swine
A	2.5	1.25
B	5.0	2.5
C	10.0	5.0
D	20.0	10.0

Because of the acute effects, such as failure to eat and early deaths, 3 dogs were added to the experiment to determine the effect of emaciation on the hematological, biochemical, and pathological parameters. These dogs were selected and paired according to eating habits and weight with those 3 dogs dying first in group D. They were killed after the same length of time and had consumed the same daily quantity of diet in grams as those dying from diazinon toxicity.

Swine. For this experiment, 14 male and 16 female Hormel-Hanford miniature swine, 4 months old and weighing 14.0 to 20.4 kg, were selected. The pigs had been wormed and immunized against hog cholera and erysipelas. They were distributed among 5 groups of 6 pigs each according to littermates, weight, and sex, and were fed a stock diet consisting of the following: corn meal, 820 lb; tankage, 40 lb; 44% soybean meal, 50 lb; pulverized oats, 350 lb; fish meal, 150 lb; alfalfa meal, 450 lb; linseed meal, 120 lb; Glauber's salt, 4.6 lb; molasses, 100 lb; and mineral mix, 10 lb. Six pounds of a veterinary antibiotic (Aurofac) were added to each ton of feed. The diet was fed at a rate of 25 g/kg/day and the rate was adjusted weekly according to the weight of the animal.

[6] Technical diazinon (Lot M.S. 402702) was generously supplied by the Geigy Chemical Corporation, Ardsley, New York 10502.

Four groups of 3 males and 3 females were given capsules containing a 1:200 dilution of diazinon in corn oil at the dosages shown in the tabulation for a period of 8 months. The controls, 2 males and 4 females, were given corn oil daily by capsule in an amount equivalent to that given group D. Capsules were administered by the use of jaw spreaders and a small balling gun. Feeding and dosing times as well as maintenance of the swine were identical to those of the dogs.

In addition to daily observations on each animal, all animals of both species were physically examined before the test was started and at regular intervals thereafter.

Twice in the 2 weeks before the study was begun and at monthly intervals thereafter, 45 ml of blood was drawn from the jugular vein of each dog and the anterior vena cava of each pig. Animals which died during the experiment were bled before death whenever possible.

Hematologic determinations performed included: hemoglobin, hematocrit, red blood cell count (RBC), reticulocyte count, white blood cell count, white blood cell differential count, platelet count, prothrombin time, and partial thromboplastin time. The methods used for these determinations have been described in a previous publication (Tegeris *et al.*, 1966).

Bone marrow was taken from the third, fourth, and fifth rib of each dog or pig at the time of death, when possible. The tip end of the ribs above the costochondral junction was removed with a sharp knife and marrow was expressed onto a coverslip. Smears were made and those used for counting were diluted with human diagnostic plasma and stained with Wright-Giemsa stain. A total of 1000 cells were differentially counted, and the M/E ratios were calculated (Melveger *et al.*, 1969).

Serum biochemical determinations included: alkaline phosphatase, blood urea nitrogen (BUN), fasting blood glucose, serum creatinine, serum glutamic oxaloacetic transaminase (SGOT), lactic dehydrogenase (LDH), ornithine carbamyl transferase (OCT), creatinine phosphokinase (CPK), amylase, lipase, and total lipids. Routine clinical methods were used to make the first 7 biochemical determinations; references to their application have previously been published (Tegeris *et al.*, 1966). The method of Oliver (1955) was used to determine creatinine phosphokinase. The procedure for the determination of amylase, as modified by Dade Reagents, Inc., is based on a standard iodometric technique (Smith and Roe, 1949; Caraway, 1959), and the results are reported in Somogyi units. The determination of lipase was based on the method of Vogel and Zieve (1963), while total lipids were determined by the method of Bragdon (1951).

The following tissues were taken at necropsy from the dogs in group D, the parallel feeding group, and the controls and were fixed in standard formalin solution and stained with hematoxylin and eosin for histopathologic study: brain, spinal cord, pituitary, thymus, thyroid, lungs, heart, liver, gallbladder, pancreas, spleen, kidney, adrenal, urinary bladder, stomach, small intestines, colon, testes or ovary and uterus, sciatic nerve, and skeletal muscle. Tissues from group C examined histopathologically were thyroid, heart, liver, kidney, adrenal, gallbladder, pancreas, spleen, stomach, small intestine, colon, and testes or ovary. The testes were the only tissues examined from groups A and B.

For histopathologic studies in swine, tissues taken from group D and the control group were the same as those taken from dogs in group D and tissues taken from group

C corresponded to those taken from dogs in group C. Liver, kidney, and adrenal were the only tissues examined from group B and only liver was examined from group A.

RESULTS

Clinical Observations

Dogs. In group D (20 mg/kg/day), 1 male dog became moribund and was killed 14 days after start of the test. A female was killed after 19 days and another male died in 24 days. The average feed intake of these 3 dogs was approximately 100 g/day during the survival period rather than the normal intake of 300–350 g. Emesis and fasciculation occurred within 4–14 days after administration of the compound and became progressively worse until the dogs died or were killed. One male dog died 166 days after the test began; it had diarrhea and emesis within 4 days but fasciculation did not occur until 40 days after start of the study. These cholinergic signs persisted until the dog died. Two female dogs were the only survivors of group D. Emesis occurred twice in 1 of these dogs and diarrhea was present on 2 different occasions in both dogs, but no other cholinergic signs were observed.

Of the 6 dogs in group C (10 mg/kg/day), only 1 showed marked cholinergic signs during the first 45 days of the study, but it survived. No cholinergic signs were observed in the remaining dogs of this group or of groups A and B.

Swine. Cholinergic signs occurred within 3–26 days after the study began in group D (10 mg/kg/day), but those pigs showing early signs were not necessarily those which died first from toxicity. Deaths occurred on days 12, 16, 20, 25, and 38; only 1 female of this group survived the study. This pig was unaffected for 4 months after the study began; cholinergic signs then occurred for about 15 days before they subsided.

Only 1 pig in group C (5 mg/kg/day) showed marked cholinergic signs during the first 6 months, but it survived the 8-month study. One pig in group B (2.5 mg/kg/day) developed cholinergic signs after 19 weeks, became moribund the following week, and was killed. Toxic signs were not observed in any of the other pigs.

Hematology

No marked effect was observed in the peripheral blood in the dogs or pigs except that 1 dog in group D showed a marked decrease in the RBC with a corresponding drop in the hematocrit and hemoglobin. This animal had a good appetite until death but steadily lost weight (3.0 kg) over a 12-week period. Three pigs in group C showed a transient drop in the RBC, hemoglobin, and hematocrit at the fifth month. Except for the M/E ratios of the bone marrow (reported under the pathological findings), all other hematologic parameters were within the normal range of biological variation.

Biochemical Findings

Dogs. Markedly elevated values for alkaline phosphatase, SGOT, LDH, OCT, and amylase were found in only 2 male dogs. One dog in group D died after 166 days. The second, the dog in group C which had lost 3.0 kg of weight during the experiment, died 5 days before the study was completed. One other dog in group D and 3 dogs in group C showed a definite sustained increase in serum amylase. The LDH values were erratically

elevated in many of the dogs in all groups. All other biochemical parameters were within the normal limits of biological variation for dogs.

Swine. Erratic, randomly elevated levels for OCT, CPK, and amylase occurred in all groups. Sustained elevations of amylase were observed in 1 pig in each of the 4 test groups. All other biochemical parameters were within normal limits of biological variation for swine.

Gross Pathology: Dose-Range Studies

Dogs. On necropsy of the dogs given 5–100 mg/kg/day of diazinon, the most frequent pathologic findings were hemorrhage on the dura mater and congestion or hemorrhage of the small intestines, ranging from petechia to paint brush hemorrhage of the mucosal and serosal surface to frank hemorrhage of the mucosal surface.

Swine. The most frequent pathologic finding in pigs given 5–50 mg/kg/day of diazinon were the same as those of the dogs, but in addition, hemorrhages of the heart (epicardial, myocardial, endocardial, valvular), large fatty areas in the pancreas and congestion of the stomach were observed. The duodenum and/or jejunum was thickened (5–6 mm) primarily because of edema. The mucosal surface of the affected areas perforated easily, resulting in the death of some pigs from peritonitis.

Gross Pathology: Chronic Studies

Dogs. In group D the most significant lesion was marked edematous thickening (6 mm) of the intestinal wall for a length of up to 18 in. of the duodenum or jejunum in 5 of the 6 dogs, yet it returned to normal size when placed in 10% formalin. A duodenal rupture, 2.5 cm long, occurred with resulting peritonitis in 1 dog, and rupture of the pyloric portion of the stomach occurred in another.

Only the dog that lost weight in group C showed gross pathologic changes. The thyroid glands were very soft. No pericardial fat was present on the heart and the vessels had a cordlike appearance. The liver was yellow and fatty in appearance. The pancreas was atrophied and rough to the touch and the spleen was markedly shrunken and pale. A 4-mm band of congestion was observed at the corticomedullary junction in the kidneys, the testicles were markedly atrophied, and the duodenal wall was thickened (5–6 mm).

Pathologic changes were found in only one remaining dog (group B). These consisted of capsular adhesions of the kidneys and congestion at the corticomedullary junction extending approximately 3 mm into the cortex. The duodenum was slightly congested, with 2 areas showing possible ulcer formation.

Swine. In group D, 4 of the 5 pigs which died had edematous thickening of the wall of the jejunum up to 24 in. long, and 1 pig had a localized mucosal erosion into the muscular layers with marked serosal seepage throughout the intestines. The livers of all of these pigs were very firm and hard to cut; ulcer formation in the duodenum was seen in 3 of the 5.

In group C, edema was present in the wall of the jejunum in 1 pig. Another had a friable liver which was very gritty and had focal subcapsular hemorrhages. Similar findings with serosal seepage were observed in the ileum of 1 pig in group B that died at 141 days; this pig also had abdominal ascites which clotted on exposure to air, and a pale, mottled liver with a whitish sheen to the capsule.

Necropsy findings in the remaining pigs showed no consistent lesions except for occasional mild intestinal congestion in some of the animals.

Histopathology: Dose-Range Study

Dogs. Histopathologic examinations were made on tissues of 9 dogs given 5–100 mg/kg/day of diazinon. Miscellaneous findings were limited to hemorrhage in the colon, patchy areas of parenchymal cells containing a golden brown pigment in the cytoplasm in the livers, and patchy areas of cortical renal epithelium containing a golden brown pigment.

Swine. Tissues of 12 pigs given 5–50 mg/kg/day of diazinon were studied. With the exception of the bone marrow, microscopic changes were inconclusive with respect to toxic effects on specific organs or tissues. Cellular depletion of bone marrow of the rib was seen in 6 of the 8 pigs in which sections of bone were examined; depletion was considered to be moderate to marked in 2, moderate in 2, and slight in 2. Other miscellaneous lesions seen in swine included hemorrhages in the colon, inflammation of the serosal surface of the small intestine, and fat necrosis with peripheral inflammation of the pancreas. Peritonitis was also present in 2 of these. In addition, slight-to-moderate focal dilatation of convoluted tubules with tubular casts were found in the kidney, along with occasional focal vacuolation of renal tubule epithelium. Inflammatory foci up to 100 μ in diameter were found in the portal tracts or interlobular septa in the livers. Areas with a golden brown pigment were noted in Kupffer cells and hepatocytes in the liver of 1 pig. Microscopically, cellular changes were not observed in sections of the intestinal wall which was grossly thickened.

Histopathology: Chronic Studies

Dogs. Liver changes found in 3 of 6 dogs in group D consisted of moderate cirrhosis, or changes related to cirrhosis ranging from focal necrosis to fibrous infiltration. Viable hepatic cells appeared dissociated. Marked testicular atrophy was present in 2 dogs; spermatogonia and sertoli cells were present in a third dog dying in this group but development was arrested beyond this stage. Localized chronic nephritis was observed in the kidneys of 1 dog in each of groups C and D, characterized by atrophied and degenerating tubules and glomeruli with fibrous infiltration.

The liver of the emaciated dog in group C which died after 232 days exhibited parenchymal atrophy and hepatic cell dissociation. Moderate atrophy was observed in the splenic pulp, as well as atrophic changes in the pancreatic acinar cells and interstitial fibrosis. Spermatogenesis was completely arrested. There were no histopathologic changes in the other dogs which could be attributed to diazinon or starvation.

Swine. Histopathologic changes were seen most frequently in the liver and intestines of the swine. Liver changes consisted of thickening of the interlobular connective tissue in 3 of 6 pigs in group D and in 1 of 6 in group B. Congestion of lobules at the lobe border was present in 1 of 6 in group C and in 1 of 6 in Group B. Hemorrhagic areas were present in the liver of 1 of 6 pigs in group C. There was a slight inflammatory reaction in the interlobular septa of 1 pig in group A, and an occasional lobule with congestion and degenerative hepatic cells in the central area. Slight thickening of the serosa, occasional focal hyperemia, and hemorrhage of the outer muscle layer were observed in the intes-

tines of pigs from groups C and D. Remarkable histopathologic changes were not observed in the other swine examined.

Myeloid-Erythroid (M/E) Ratios

Bone marrow smears were obtained from 5 of the 6 dogs in group D. The hematocrits, red blood cell counts, reticulocyte counts, and the M/E ratios were obtained from the dogs and, along with the average for the control dogs, are shown in Table 3. All 3 dogs dying within 14–24 days showed an increase in the myeloid elements of the bone marrow with slight to moderate hypocellularity, and 2 of the dogs from which blood was obtained showed a reticulocytopenia. As can be seen from Table 3, no peripheral anemia was present in any of these dogs. The M/E ratios of the dogs in the other groups were within normal limits, with the highest being 2.0/1.

TABLE 3

TERMINAL HEMATOLOGIC FINDINGS IN DOGS GIVEN DIAZINON (20 MG/KG/DAY)

Dog No.	Sex	Days on test	Hemoglobin (%)	Hematocrit (%)	RBC $\times 10^6$	Reticulocyte (%)	M/E Ratio
48G	F	19	22.3	65.0	9.8	0.0	143.0/1
54G	F	237	16.8	50.0	6.9	1.8	1.6/1
77G	F	237	15.8	47.5	5.8	2.4	1.1/1
89F	M	24	—	—	—	—	114.0/1
18H	M	14	18.8	56.5	6.4	0.0	183.0/1
Control							
Average	—	—	15.9	47.5	6.1	0.9	1.4/1
Range			(14.7–16.8)	(45.0–49.5)	(5.9–6.3)	(0.4–1.8)	(1.1/1–1.9/1)

Bone marrow smears were obtained from 5 of 6 pigs in group D. Three of the M/E ratios were considered to be elevated (Table 4). All pigs examined in this group showed a reticulocytopenia. One pig in group C which died after 141 days had an M/E ratio of

TABLE 4

TERMINAL HEMATOLOGIC FINDINGS IN SWINE GIVEN DIAZINON (10 MG/KG/DAY)

Pig No.	Sex	Days on test	Hemoglobin (%)	Hematocrit (%)	RBC $\times 10^6$	Reticulocyte (%)	M/E Ratio
1424	M	16	—	46.0	—	0.1	3.9/1
1516	M	20	—	48.0	—	0.4	4.5/1
1456	F	25	17.3	50.5	9.8	0.2	3.4/1
1487	F	12	—	51.0	—	0.2	2.4/1
1497	F	238	16.2	48.0	7.9	1.0	1.3/1
Control							
Average	—	—	15.4	46.4	7.4	1.0	1.8/1
Range			(14.0–17.0)	(44.0–51.0)	(6.3–8.3)	(0.6–1.5)	(1.3/1–2.2/1)

9.2/1 and the marrow was hypocellular. Although this pig had shown a reticulocytopenia early in the study, the count was normal at the time of death, as were the hematocrit, hemoglobin, and RBC. Peripheral anemia was not present in any of these animals in which an increase of myeloid elements of the bone marrow was demonstrated.

DISCUSSION

The dose-range studies revealed a wide variation in the toxic dose in both species of animals. In dogs, the females seemed to tolerate higher doses than did the males, whereas the reverse was true in swine. These observations were somewhat substantiated in the chronic study in that 2 female dogs survived the study and the males died earlier than females. The first pig to die was a male followed by alternating deaths by sex thereafter. Vascular lesions observed in the small intestines were similar to those reported in man by Limaye (1966). Marked resistance was shown in the 1 female surviving the study. However, a female littermate to this female also showed marked resistance to diazinon in another study. Whether this difference is caused by individual variability, hepatic microsomal enzyme activity, hormonal influence, or is possibly influenced by fat deposition in the body is not known at this time.

No overt toxicologic signs were seen in other studies when 10 and/or 20 mg/kg/day of diazinon was added to the diet of swine and dogs for 8 weeks. However, in the dose-range and chronic studies with the same dosages of diazinon administered in oil, death occurred in a much shorter period of time. Apparently, absorption of diazinon is enhanced by the oil but the compound is poorly absorbed when added to the diet or possibly absorbed over a longer period of time.

Marked serum amylase elevations that recurred in certain animals cannot be explained at this time as there was no correlation between the elevated amylase values and pathological findings.

Starvation of the dogs on parallel feeding studies did not influence the hematologic or biochemical parameters that were measured.

The outstanding necropsy finding in both dogs and swine was the marked thickening (up to 6 mm) of the small intestine. This thickening was edematous in nature and was not accompanied by any other significant pathologic changes. This finding, along with rupture of the duodenum or jejunum, has been observed previously in a toxicity study conducted in this laboratory in which parathion was used.

Histopathologic changes occurred primarily in the high-dose groups, but there was little uniformity of lesions. An effect was seen in the livers of both species, but the lesions were not similar and the changes seen in swine livers were not striking in character. Diazinon probably has an adverse affect on the liver when enhanced by the stress of inadequate food consumption. In group D, 2 of the 3 male dogs had moderate to marked testicular atrophy, but the swine testes all appeared normal. It is possible that the added stress of malnutrition may have contributed to this testicular atrophy. Food consumption of animals which received high doses of the pesticide dropped drastically very early in the experiment. However, the dogs placed on parallel diets with the three anorectic dogs did not have similar lesions.

The pancreatic changes seen in a male dog in group C appeared to be spontaneous chronic pancreatitis and were probably not the direct effect of diazinon.

The increase in the M/E ratio without anemia in both dogs and pigs was an unusual finding. Length of survival and individual susceptibility appear to play an important role in the increase of this ratio. An exception to this was the 1 pig which received 2.5 mg/kg/day and survived 141 days. The mechanism of action of diazinon on the bone marrow appears to be associated with the production of erythrocytes, since the observed hypocellularity is in the erythrocytic and not in the myeloid series. It might be postulated that some mechanism within the animal triggers this bone marrow change before death and, if survival of the animal could be prolonged, anemia would result.

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