Scand. J. Lab. Anim. Sci. No. 4 . 1995 . Vol. 25

Effect of mixed administration of selenium and fenitrothion on pathomorphological pattern in internal organs in rats

by *Józef Szarek, Arkadiusz Zasadowski*, Jacek Fabczak & Dariusz Barski**,
Department of Forensic Veterinary Medicine and Veterinary Medicine Administration,
*Department of Veterinary and Environmental Toxicology, Faculty of Veterinary Medicine,
Olsztyn University of Agriculture and Technology, Oczapowskiego Str. 13, 10-719 Olsztyn, Poland.

Introduction

Modern agriculture relies on the use of pesticides (including fenitrothion). Therefore wildlife inhabiting agricultural land can be exposed to a wide range of phosphoroorganic compounds, especially if the animal is highly mobile, as in the case of birds (*Becker et al.* 1992). Birds in particular, have a very limited capacity to degrade organophosphates and are thus extremely vulnerable to poisoning (*Becker et al.* 1992, *Braley et al.* 1980). The potential for multiple exposures raises the question whether the toxic effects of mixtures of chemicals are simple additives or whether there is enhancement of toxicity. Multiple poisoning usually occurs in the environment but it is always consistent with single poisoning (*Ekholm et al.* 1991, *Ernst et al.* 1991, *Hamdy & Gissel-Nielsen* 1976, *Hill & Fleming* 1982).

The toxic effects of more than one compound can be a result of their incidental synergistic or antagonistic activity (*Ernst et al.* 1991).

Selenium is both toxic and essential to the environment in which it is often introduced. The toxicity of fenitrothion has been studied in a lot of experiments (*Hamdy & Gissel-Nielsen* 1976, *Holmes & Boag* 1990) as well as selenium (*Hill & Fleming* 1982, *Itoh* 1991, *Jepson* 1993, *Johnston et al.* 1994, *Kantola & Vartianen* 1991, *Khan et al.* 1993). The influence of both factors has been rarely presented and the objective of our study was to obtain information on the effect of selenium and fenitrothion on other organs than liver and kidney in the rat.

Materials and methods

The experiment was conducted on 240 Wistar female rats (body mass from 180 g to 220 g, Brwinów near Warsow). Rats were fed with granulated feed "Murigram". Drinking water was given *ad libitum*. The animals were kept in cages measuring 45 cm × 35 cm × 20 cm in groups of 5. Wood scrappings were used for bedding. The room temperature was about 22 °C, the relative humidity about 30–40 %, and the daylight regime 12 h. Animals were allocated to six groups (Table 1) each of 40 rats. Sodium selenite (Na_2SeO_3, POCH, Polish Chemical Reagents, Gliwice) was given in drinking water over 12 days. Fenitrothion (IPO, Warsow) 81.4 % solution in cazole oil was administered per os by gavage on the 6-th and 12-th day. Doses and time for administration of these chemicals were adapted from previous experiments (*Szarek et al.* 1994).

Table 1. Outline of the experiment.

GROUP	TREATMENT	
	sodium selenite* (ppm)	fenitrothion** (mg/kg b.m.)
I	0.08	0
II	0.40	0
III	0	40
IV	0.08	40
V	0.40	40
VI (control)	0	0

* - over 12 days given in drinking water.
** - administered per os by a tube inserted into the stomach.

During the experiment the rats were examined twice daily for clinical signs and the consumption of feed and water was monitored daily. Eight animals from each group were weighed and anaesthetized by ethyl ether on the 1st, 3rd, 7th, 14th and 28th day post intoxication (p.i.). Post mortem examination was conducted and sections of stomach, duodenum, ileum, caecum, lien, lung, cardiac muscle and skeletal muscle were collected for histopathological examination. Sections were fixed in 10 % neutralised formalin and stained with hematoxylin and eosin (HE).

Experimental results

Clinical examination of rats from groups III, IV and V indicated an occurrence of periodic depression with a partially decreased feed intake. Decrease of body mass of rats in all treatment groups was observed compared with controls. These results have been described elsewhere (*Szarek et al.* 1994).

About 30 % of animals in group I showed a degree of hyperaemia of the mucous membrane in the stomach, starting 24 hours to 3 days p.i. These changes were sporadically found in the mucous and submucous membrane of the duodenum up to 7 days p.i. The other organs examined showed a normal structure.

Almost half of the animals in group II had hyperaemic mucous membranes and in some cases hyperaemia of the submucous membrane in the region of the stomach and duodenum. This disorder lasted up to 7 days p.i. In 10 % of the cases there was hyperaemia of the mucous membrane in the ileum, which lasted up to 14 days p.i. An increase in size of reaction centers of the spleen was observed. These changes were seen sporadically up to 14 days p.i.

In over half of the animals from group III, there was a noticeable excessive desquamation of the epithelial membrane of the stomach and duodenum which was most di-

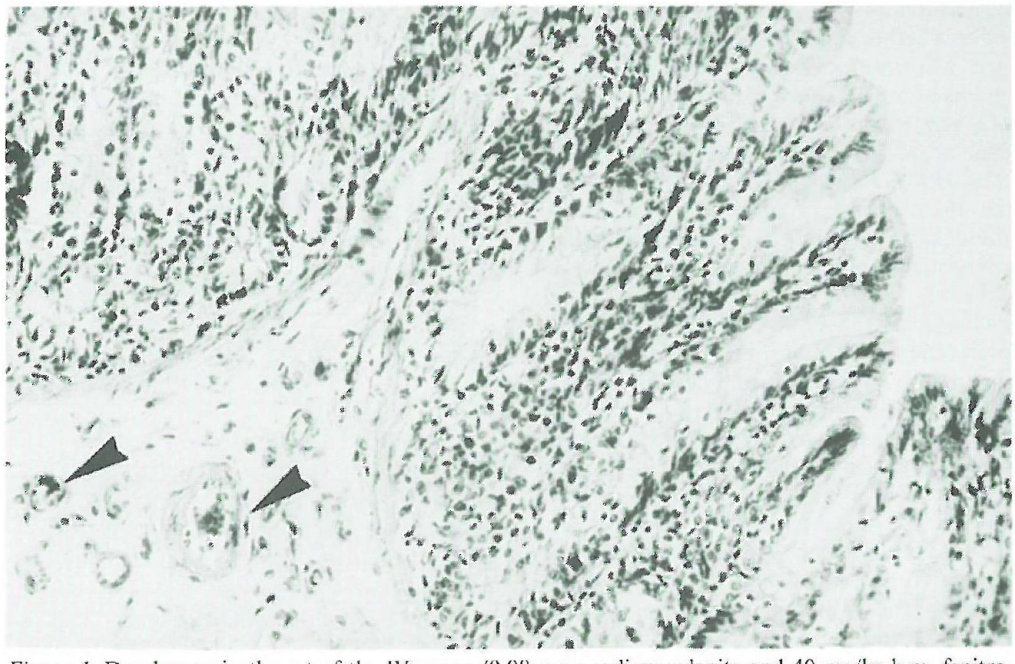

Figure 1. Duodenum in the rat of the IV group (0.08 ppm sodium selenite and 40 mg/kg b.m. fenitrothion), 3 days p.i. – slight hyperaemia (heads of arrows). Staining with HE, magn. × 250.

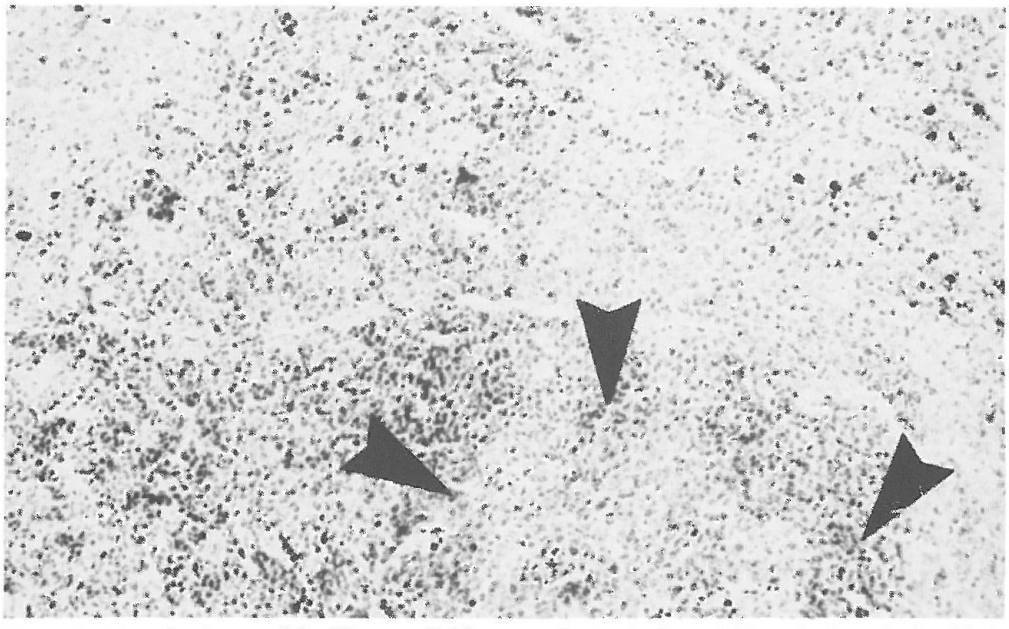

Figure 2. Spleen in the rat of the IV group (0.08 ppm sodium selenite and 40 mg/kg b.m. fenitrothion), 7 days p.i. – magnification of reaction centre (heads of arrows). Staining with HE, magn. × 250.

stinct 24 h p.i. It became less distinct in the later stages. On the 14th day after intoxication only traces were present. In some cases, such changes were accompanied by distinct hyperaemia of the mucous and submucous membrane. The above mentioned disorders were seen sporadically within the walls of the ileum. In some cases hyperaemia of the cardiac muscle was noted up 7 days p.i.

In group IV in almost all cases there was some hyperaemia of the submucous membrane in the stomach and duodenum up to seven days p.i. However, the villi of the small intestines generally did not show any variation from the norm (Figure 1). Excessive desquamation of epithelium was observed up to 7 days p.i. in some cases. About 70 % of the rats had an enlarged reaction center of the spleen up to 7 days p.i. (Figure 2). This particular disorder became less pronounced as time elapsed.

In a few cases rats from group V showed chronic inflammation of the mucous membrane of the duodenum (Figure 3) up to 14 days p.i. Hyperaemia of the mucous and submucous membrane of the stomach, duodenum and ileum was noticed relatively often up to 14 days p.i. There was also excessive desquamation of the epithelial mucous membrane and sometimes a great conglomeration of cells with eosinophilic granules in the cytoplasm. Hyperaemia of the cardiac muscles was observed in about 1/3 of the rats. Germinal centers in spleen were found increased in diameter up to 3 days p.i. Animals from group VI had organs with normal microscopic pattern.

Discussion

The present study has demonstrated that histological changes due to selenium and fenitrothion also occur in organs other than liver and kidneys which are the most often studied in cases of intoxication. Furthermore the post mortem and microscopic examination of rats intoxicated with selenium and

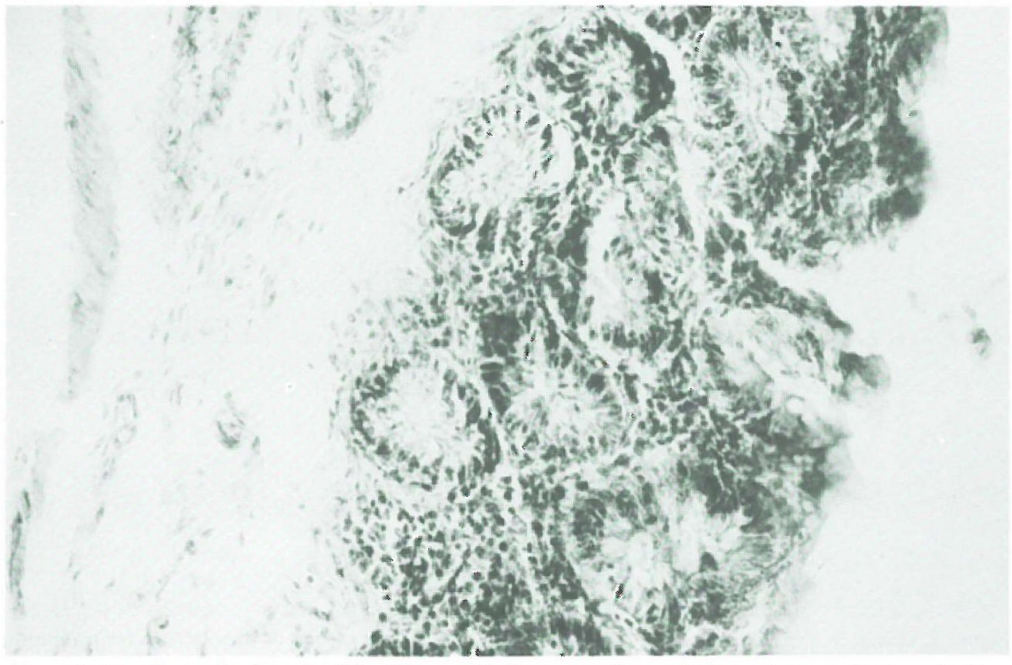

Figure 3. Duodenum in the rat of the V group (0.40 ppm sodium selenite and 40 mg/kg b.m. fenitrothion), 7 days p.i. – duodenitis catarrhalis. Staining with HE, magn. × 250.

fenitrothion in the present study suggest a variation in the pattern and severity of changes in rats of different groups. The highest relative deviation from the norm occurred after application of a mixture of selenium and fenitrothion at a dose of 0.4 ppm and 2 × 40 mg per kg body mass, respectively. Changes occurred over a very extensive area. When a lower dose of selenium (0.08 ppm) with fenitrothion (2 × 40 mg/body mass) was applied, there was a marked decrease in the disorders of organs tested. In this case, the intensity of changes was lower than in cases when selenium (0.4 ppm) and fenitrothion were used separately. It is important to note that the changes in the examined organs were different when mixed intoxication was applied from changes when only a single toxicant was used. This was also the case for liver and kidneys, but these differences were less distinct (*Szarek et al.* 1994).

Comparison of the effect of the same dose of fenitrothion (40 mg/kg i.m.) administered with (group IV) or without (group III) sodium selenite indicated a protective effect of sodium selenite at dose level of 0.08 ppm against the fenitrothion. However a mixed poison at a dosage of 0.4 ppm caused an increased deviation from the norm. Such a diversity of selenium activity in relation to other compounds has been noted in the literature (*Jepson* 1993, *Khan et al.* 1993, *Szarek & Khan* 1993, *Szarek et al.* 1992).

Moreover, our own observation has proved the importance of having a good knowledge of the impact of selenium in the environment (*Johnston et al.* 1994, *Kantola & Vartianen* 1991, *Koivistoinen & Huttunen* 1986, *Lindberg & Bingerfors* 1970, *Zasadowski & Buszko* 1988). Such knowledge allows for the right prediction of the behaviour of organisms when compounds (as for example

fenitrothion) are introduced to the environment by man (*Itoh* 1991, *Kantola & Vartianen* 1991, *Lari et al.* 1994, *Nakonieczny* 1993, *Walker et al.* 1993).

Summary
Mixed intoxication in rats with sodium selenite (0.40 ppm) and fenitrothion (40 mg/kg b.m.) resulted in pathomorphological changes in stomach, intestine, spleen and cardiac muscle which were more intense than in cases when the mentioned compounds were administered singly. When a lower dose of selenium (0.08 ppm) together with fenitrothion (40 mg/kg b.m.) was used there was a decrease of disorders in examined organs.
Moreover, the changes were even less intense than when selenium (0.40 ppm) or fenitrothion (40 mg/kg b.m.) were administered as a single toxicant.
Thus, it was concluded that selenium at a dose of 0.08 ppm protected the rats by limiting the toxic activity of fenitrothion.

References

Becker D, Z Romic, H Krsnjavi & Z Zima: A contribution to the world selenium map. Biol. Trace Elem. Res. 1992, *33,* 43–49.

Braley CJ, CH Walker & BC Baldwin: A-esterase activities in relation to the differential toxicity of primiphos-methyl to birds and mammals. Pest. Sci. 1980, *11,* 546–554.

Ekholm P, M Ylinen, M Eurola, P Koivistoinen & P Varo: Effects of general soil fertilization with sodium selenite in Finland on the selenium content of milk, cheese and eggs. Milchwissenschaft 1991, *46,* 547–550.

Ernst W, G Julien & P Hemsigar: Contamination of ponds by fenitrothion during forest spraying. Bull. Environ. Contam. Toxicol. 1991, *46,* 815–821.

Hamdy AA & G Gissel-Nielsen: Relationship between soil factors and selenium content in Danish soils and plants. Risø Rapport 1976, *349,* 1–16.

Hill EF & WJ Fleming: Anticholinesterase poisoning in birds: field monitoring and diagnosis of acute poisoning. Environ. Toxicol. Chem. 1982, *1,* 27–38.

Holmes SB & PT Boag: Effects of the organophosphorus pesticide fenitrothion on behaviour and reproduction in zebra finches. Environ. Res. 1990, *53,* 62–75.

Itoh K: Metabolism of fenitrothion, parathion and cyanophos by isolated salition – degrading bacteria from soil. Journ. Pest. Sc. 1991, *16,* 97–100.

Jepson PC: Ecological insights into risk analysis: the side-effects of pesticides as a case study. Sc. Total Environ. 1993, suppl. part 2, 1547–1566.

Johnston G, CH Walker & A Dawson: Interactive effects between ebi fungicides (prochloraz, propiconazole and penconazole) and op insecticides (dimethoate, chlorpyrifos, diazinon and malathion) in the hybrid red-legged partridge. Environ. Toxicol. Chem. 1994, *13,* 615–620.

Kantola M & T Vartianen: Selenium content of breast milk in Finland, after fertilization of soil with selenium. J. Trace Elem. Electrolytes Health Dis. 1991, *5,* 283–284.

Khan MZ, J Szarek, M Saeed, A Koncicki & A Krasnodebska-Depta: Effects of concurrent oral administration of monensin and selenium on some haematological and biochemical parameters in broiler chickens. J. Vet. Med. B. 1993, *40,* 667–675.

Koivistoinen P & JK Huttunen: Selenium in food and nutrition in Finland. An overview on research and action. Am. Clin. Res. 1986, *18,* 13–17.

Lari L, A Massi, MC Fossi, S Casini, C Leonzio & S Focardi: Evaluation of toxic effects of the organophosphorus insecticide azinphosmathyl in experimentally and naturally exposed birds. Arch. Environ. Contam. Toxicol. 1994, *26,* 234–239.

Lindberg P & S Bingerfors: Selenium levels in forages and soils in different regions of Sweden. Acta Agric. Scand. 1970, *20,* 133–136.

Nakonieczny M: Functional aspects of cadmium and selenium interactions in insect digestive tract. Enzyme studies. Sc. Total Environ. 1993, *40,* 667–675.

Szarek J, A Zasadowski, J Fabczak & A Spodniewska: [Effect of sodium selenite and fenitrothion on the pathomorphological pattern of liver and kidney in rats]. Zesz. Nauk. 1994, *9,* 149–156. (Polish).

Szarek J & MZ Khan: Concurrent exposure to lead selenium or monensin effects on hepatic porphyrin levels in broiler chickens during acute toxicosis. Scand. J. Lab. Anim. Sci. 1993, *20,* 231–235.

Szarek J, T Rotkiewicz & J Fabczak: [Effect of cadmium and selenium on the histhopathological pattern of internal organs in broiler chicken]. Pat. Pol. 1992, *43,* 49. (Polish).

Walker CH, GO Johnston & A Dawson: Enhancement of toxicity due to the interaction of pollutans at the toxicokinetic level in birds. Sc. Total Environ. 1993, suppl. part 1, 525–531.

Zasadowski A & M Buszko: [Role of active biochemical elements in the animal organisms]. Acta Acad. Agricult. Tech. Olst. 1988, *17,* 167–181. (Polish).