

Preliminary Observations on the Effects on the Organs of Mice of Administering Some Carbamate Pesticides

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Karbatox 75 and Unden, pesticides of the carbamate group, are widely used in Poland. Though they are not usually regarded as very toxic, we thought it expedient to test the effect of these substances on some internal organs of mice.

Karbatox 75 (Carbaryl, Sevin, 1-Naptyl-N-methylcarbamate) is an insecticide frequently used in fruit-farming and agriculture; it is produced by "Azot," Jaworzno. Carbaryl has already been investigated by several authors as to its mutagenic influence on plants; these authors found that it had an effect on the mitoses in the root tips and led to the occurrence of numerous chromosome aberrations (Wuu, 1967; Amer, 1965).

Unden (Propoksur, Baygon, Bay 39007, 2-Isopropoxy-phenyl-N-methyl-carbamate) produced by "Chemagro" is also a well-known insecticide, often used in Poland. It is more toxic than Karbatox.

The pesticides of the carbamate group act by contact or per os. Their biological effect probably consists in inhibiting choline esterase. Intoxications in man pass rapidly, as these compounds are quickly detoxicated (Melnikov, 1971).

Material and Methods

The investigations were made on 50 male mice from the Balb strain, with a mean body weight of 24 g. The animals were daily injected intraperitoneally with a suspension of the pesticide in physiological saline in a dose of 20 mg per kg of body weight. The control animals were injected intraperitoneally with physiological saline alone. The mice were divided into three groups: (1) animals treated with Karbatox, (2) animals treated with Unden, and (3) control animals. After 10 and 20 days the animals were killed by decapitation and after dissection the following organs were taken for histological investigation: intestine, testes, liver, and kidneys. The brain and hypo-

physis were fixed separately. Preparations were additionally made from the spleen and testes for karyological investigation. After dissection the organs were fixed in Bouin's fluid, sections 10 microns thick were stained with haematoxylin and eosin.

For karyometric investigations such sections were stained with chrome haematoxylin-phloxin after Gomori and modified by Bargmann. The measurements were then made as follows: the long and short axes of cell nuclei were measured in neurosecretory neurons of nucleus supraopticus and nucleus paraventricularis, 100 nuclei in each neurosecretory centre of each animal. The cell nucleus volume was calculated from the formula $V = \frac{\pi}{6} LB^2$, where L is the long axis of the nucleus, and B, the short axis of the nucleus. The means for the experimental and control groups were compared statistically by the χ^2 test.

The Gomori-positive glia was studied quantitatively by counting the glia cells with Gomori-positive granules in the cytoplasm. Such cells were counted in nucleus habenulae bilaterally in areas measuring 0.076 mm². In each animal the Gomori-positive glia cells were counted in 100 such areas. The arithmetical means were compared by the χ^2 test.

Results and Discussion

The more important quantitative results are presented in the following table:

Nuclear volume in neurosecretory cells of hypothalamus

	Group I Karbatox	Group II Unden	Group III Controls
Nucleus supraopticus	408.61 μ^3 *	415.19 μ^3 *	362.57 μ^3
Nucleus paraventricularis	359.34 μ^3 *	302.78 μ^3	312.18 μ^3

Number of Gomori-positive glia cells per 0.076 mm² of nucleus habenulae

	Group I	Group II	Group III
	6.88*	7.93*	5.75

*difference statistically significant

No differences were found in the sections of the testicles. In all three groups the testes were normal, the spermatogenesis also being normal. Similarly, in kidney sections there were no deviations from normal. In the intestine, however, there were some differences between the experimental animals and the controls. In animals treated during 20 days with Uden, on the sections through the intestine, the villi show cells of the columnar epithelium nearly twice as long as in the controls. The striated border in these cells is much thicker and the number of mitoses in the Lieberkühn crypts smaller than in the controls. Also in the liver changes were noted, consisting in the occurrence in the control animals of parenchymal cells with large nuclei containing a drop or two of an amorphous lightly staining material. These nuclear inclusions were not observed in the experimental animals or were much rarer. The liver cells in the experimental animals treated with Uden were smaller and more intensely stained than in the controls, being at the same time less vacuolated.

Karyological investigations were made on preparations of the spleen and testicles following conventional karyological methods (Jordan, 1959).

In the karyological preparations no changes were found in the insecticide-treated animals. The number and structure of the chromosomes were apparently quite normal. While we know from the literature about chromosomal aberrations caused by pesticides (Epstein and Legator, 1971; Johnson and Jalal, 1973), and in particular by those of the carbamate group, we believe that in the future more attention should be paid to this problem. These compounds will be further investigated in our laboratories as to their karyological and other effects both in vitro and in vivo on a more varied material.

The increase in number of Gomori-positive glia cells in animals treated with the insecticides indirectly corroborates the hypothesis (Srebro and Rzehak, 1972) ascribing a protective role to these cells for the brain tissue. A similar increase was found as an effect of Togal (salicyl-quinine-lithium) in experiments on rats (Srebro and Szirmai, 1971). Probably the Gomori-positive glia cells intercept xenobiotics which penetrate from the bloodstream into the brain tissue. It is characteristic that the layer of these cells is thickest in places where the blood-brain barrier is lacking (Srebro, 1969).

The authors regard the present paper as an introduction to a more complete cycle of investigations devoted to the problem of pesticide effects.

Summary

Two pesticides, Karbatox 75 and Unden, were used for the investigation, in daily doses of 20 mg per kg body weight, injected intraperitoneally during a period of 20 days. A comparatively low toxicity of these compounds for mice was ascertained. Among morphological changes were found: hypertrophy of the intestinal epithelium, a decrease in size of liver cells and a significant increase in number of Gomori-positive glia cells, as well as an increase in the volume of cell nuclei in the neurosecretory neurocytes of hypothalamus.

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