

EFFECT OF COMPLEX COMPOUNDS OF COBALT  
AND IRON ON REGENERATION OF THE LIVER  
IN IRRADIATED ANIMALS

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Complex compounds of cobalt (coamide,  $\text{Co}^{30}$ , and  $\text{Co}^{35}$ ) and iron (ferrant) accelerate regeneration of the liver in rats after irradiation and partial hepatectomy.

Previous investigations by the writers [1-3] showed that certain complex compounds of cobalt and iron stimulate regeneration of tissues and organs, increase the number of blood cells, restore normal blood clotting, increase protein synthesis, and stimulate natural and artificial factors of immunity in irradiated and unirradiated animals.

The object of the present investigation was to study the effect of complex compounds of cobalt (coamide,  $\text{Co}^{30}$ , and  $\text{Co}^{35}$  - compounds synthesized under the direction of Professor M. A. Azizov) and iron

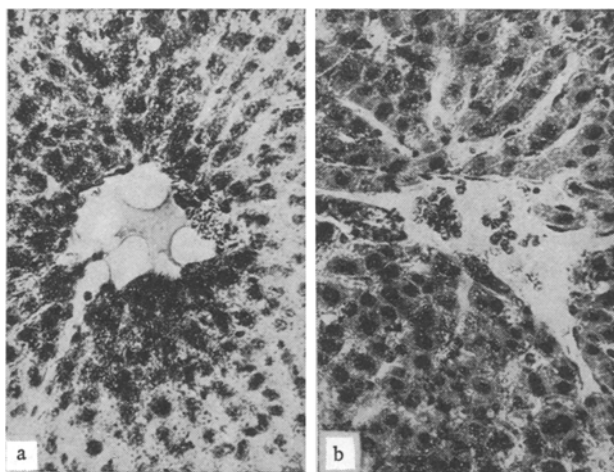


Fig. 1

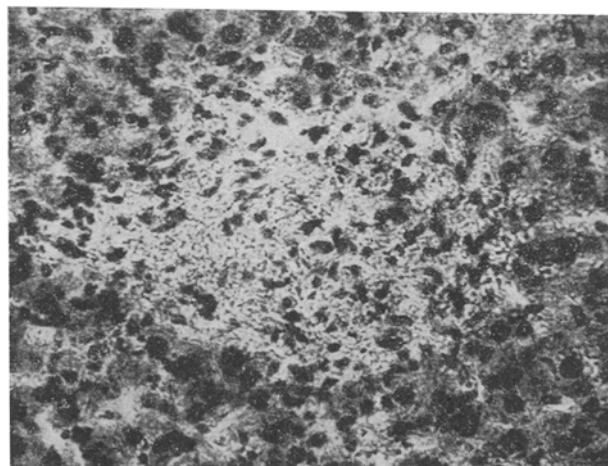


Fig. 2

Fig. 1. Liver in irradiated rats on the 4th day after partial hepatectomy: a) control; b) treated with  $\text{Co}^{35}$ . Dilated central veins, and hypertrophied trabeculae and liver cells can be seen, 200  $\times$ .

Fig. 2. Liver of irradiated, untreated rat on 14th day after partial hepatectomy. Necrosis of the parenchyma of the organ can be seen, 200  $\times$ .

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TABLE 1. Effect of Complex Compounds of Cobalt and Iron on Regeneration of the Liver in Irradiated and Partially Hepatectomized Rats ( $M \pm m$ )

Experimental conditions	Wt. of liver before operation (in g)	Wt. of remaining lobes of liver (in g)	Mean weight of regenerating organ (in g)					
			day of experiment					
			2nd	4th	7th	14th	30th	45th 60th
Administration of coamide	5,6±0,2	2,1±0,15	3,2±0,17	4,3±0,21	5,0±0,23	5,4±0,19	5,9±0,18	6,5±0,21
» Co <sup>30</sup>	5,6±0,21	2,1±0,13	3,1±0,15	4,0±0,19	4,8±0,18	5,1±0,21	5,5±0,18	6,2±0,19
» Co <sup>35</sup>	6,4±0,28	2,2±0,2	5,1±0,25	5,1±0,25	5,5±0,24	5,6±0,19	6,2±0,23	7,7±0,2
» ferrant	5,7±0,22	2,1±0,18	3,9±0,23	4,7±0,18	5,0±0,22	6,2±0,25	6,9±0,15	7,8±0,18
Control (irradiated rats)	5,5±0,18	2,1±0,15	2,7±0,14	3,5±0,17	3,6±0,16	3,9±0,27	4,4±0,28	5,3±0,33
Control (unirradiated rats)	5,6±0,16	2,0±0,17	3,5±0,22	4,3±0,20	5,4±0,18	6,0±0,19	6,5±0,2	7,0±0,2

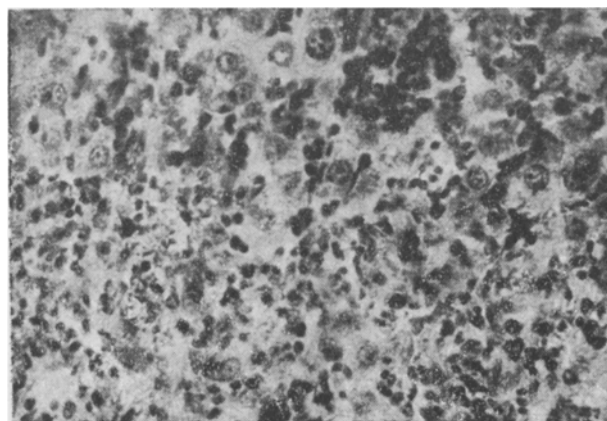


Fig. 3. Liver of an irradiated rat, treated with Co<sup>35</sup>, on the 30th day after partial hepatectomy. Replacement of foci of round-cell infiltration by young liver cells, 200 ×.

(ferrant - a mixture of iron with typhoid antigen suggested by Professor A. I. Nikolaev) on regeneration of the liver in animals with radiation sickness.

#### EXPERIMENTAL METHOD

Experiments were carried out on 245 male albino rats weighing 150-160 g. Of these animals, 210 were subjected to single whole-body irradiation on an RUM-11 x-ray therapy apparatus in a dose of 600 R under the following conditions: voltage 180 kV, current 10 mA, filter 0.5 cm Cu, dose rate 18.5 R/min. On the day after irradiation, hepatectomy was performed on the rats by the method of Higgins and Andersen (about 65-70% of its weight). The animals were divided into 5 groups: 1) control (70 rats); 2) receiving coamide (35 rats); 3) receiving Co<sup>30</sup> (35 animals); 4) receiving Co<sup>35</sup> (35 rats); 5) receiving ferrant (35 rats).

The compounds for testing were injected intramuscularly daily for 25-30 days after the operation as aqueous solutions (coamide and ferrant 1%, Co<sup>35</sup> 0.4%, and Co<sup>30</sup> 0.1%), in a dose of 2-3 mg/kg body weight. The control rats received the same volume of physiological saline by intramuscular injection. An additional control consisted of 35 hepatectomized, unirradiated rats. On the 2nd, 4th, 7th, 14th, 30th, 45th, and 60th days after operation, batches of 3-5 rats were decapitated, the liver was removed and weighed, and pieces of parenchyma were taken from it for histological study. The tissue was fixed in 12% neutral formalin. Celloidin sections, 6-8 μ in thickness, were stained with hematoxylin-eosin and by Van Gieson's method. Before the experiment began, and before sacrifice of the rats, the animals were weighed and a full blood count taken. Regeneration of the liver was studied by determining the times of recovery of its original weight, the degree of normalization of morphological structures of the organ, and mitotic activity of the hepatocytes.

#### EXPERIMENTAL RESULTS

Irradiation produced moderately severe and severe degrees of radiation sickness in the rats, while the operation shortened the time elapsing before its appearance and aggravated the severity of the disease. The mortality among animals of the control group reached 35%, and among those treated with Co<sup>30</sup> 12%, coamide 10%, ferrant 9%, and Co<sup>35</sup> 8.6%. On the 14th day of irradiation the body weight of the control rats was lowered

TABLE 2. Changes in Mitotic Activity of Liver Cells in Irradiated Rats and Rats Treated with Complex Compounds of Cobalt and Iron

Experimental conditions	Mean weight of regenerating organ (in g)			
	day of experiment			
	2-й	4-й	7-й	14-й
Control (irradiated rats)	220±5,75	70±3,58	18±2,1	4±0,8
Control (unirradiated rats)	81±5,4	30±2,8	15±0,77	8±1,1
Administration of coamide	128±1,72	51±1,43	23±1,09	11±0,82
» Co <sup>30</sup>	118±2,62	48±1,2	19±1,27	9±0,81
» Co <sup>35</sup>	126±4,7	55±3,2	23±4,2	14±1,62
» ferrant	128±1,9	47±1,9	17±1,3	9±0,77

on the average by 24-28%, and it returned to its initial level on the 45th day. In the group of treated animals, the weight on the 7th day was reduced by 10-12%, and on the 14th day of the experiment it was restored completely. On the 7th day of irradiation, the leukocyte count in most rats of the control group was lowered to 850, returning to its initial level on the 30th day. In the group of treated animals the leukocyte count fell only to 3500, and its recovery was complete on the 14th day of the experiment. The increase in mass of the partially resected liver took place much more rapidly in the groups of treated rats than in the control (Table 1).

It is clear from Table 1 that by the end of the observations on the irradiated control rats, the weight of the liver had not regained its original value. Starting from the 2nd month of the experiment, the weight of the liver in animals treated with ferrant gradually fell, and by the end of the observation it had reached its initial level. Meanwhile, in the unirradiated rats and rats treated with Co<sup>35</sup>, the mass of the liver increased progressively, while in groups treated with coamide and Co<sup>30</sup>, no great fluctuations in weight of the liver took place during this period. Consequently, in rats treated with ferrant, the increase in size of the liver took place more rapidly, and the time required for restoration of the normal morphological structure of the organ was shortened.

A study of the histological sections showed that on the 2nd day after the operation there were no differences in liver structure between the treated and untreated rats, apart from differences in the number of mitoses, which were much more numerous in the experimental animals than in the control, irradiated rats (Table 2). On the 4th day the structure of the liver in the treated rats differed sharply from that of the untreated animals (Fig. 1). In the control rats, the trabecular structure of the hepatic lobules was disturbed, extensive foci of breakdown of the normal pattern of the lobules were observed, hepatocytes were distributed chaotically, their cytoplasm was swollen and pale and their nuclei polymorphic, and small, hyperchromic cells were predominant. On the 14th day, foci of hemorrhage and necrotic areas of different sizes appeared in the substance of the lobules (Fig. 2). The number of Kupffer cells was reduced. At the height of radiation sickness, signs of destruction of the morphological elements of the organ continued to progress, and not until the 30th-45th day of the experiment was a gradual but irregular return to normal begun. However, even on the 60th day after the operation, complete recovery of the normal liver structure had not taken place in the control group.

In the groups of treated rats, throughout the experiment the trabecular structure of the liver lobules was preserved, with only slight differences depending on the compounds used. The hepatocytes and their nuclei were enlarged. The cytoplasm of the cells was filled with fine basophilic granules, and vacuolation was observed only on the 4th and 7th days of the investigation. The mitotic activity of the liver cells was considerably higher in the treated animals, and on the 7th-14th day it was higher than in the unirradiated rats. Observations showed that in irradiated and treated rats, the structure of the liver was completely restored on the 14th-30th day of the experiment. Foci of lymphoid infiltration in the substance of the parenchyma and of extramedullary hematopoiesis, observed in the groups of rats treated with coamide and Co<sup>35</sup>, by the 30th day of the experiment were replaced by young hepatocytes with pale cytoplasm and large nuclei (Fig. 3).

Ferrant, Co<sup>35</sup>, and coamide thus stimulate regeneration of the liver in rats with combined radiation injuries, and improve their general condition, the state of their peripheral blood, and their survival rate.

#### LITERATURE CITED

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