

# EFFECT OF THYROID EXTRACT AND TRIIODOTHYRONINE, WITH OR WITHOUT THE ADDITION OF THEOPHYLLINE, ON REGENERATION OF THE LIVER IN RATS

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UDC 612.35-612.6.03]014.46:615:357.441

Starting from the day after operation, partly hepatectomized rats received thyroid or triiodothyronine separately or in conjunction with theophylline, by mouth. Thyroid and the thyroid hormone increased the oxygen consumption but considerably reduced the glycogen content of the liver. No statistically significant increase in the degree of regeneration hypertrophy, studied on the 7th day after the operation, was produced by these preparations. In conjunction with theophylline they increased the absolute and relative weight of the regenerating liver. The absolute weight of the liver was increased by 19-36%. The DNA content, the number of nuclei, and the number of binuclear cells were indistinguishable from the control.

**KEY WORDS:** thyroid extract; triiodothyronine; theophylline; regeneration of the liver.

Administration of thyroxine sharply reduces the liver glycogen content [7, 8, 11, 12]. Considering that glycogen breakdown is the source of energy in the regenerating liver it might be expected the hyperthyroidization would stimulate regeneration of the liver. This has in fact been demonstrated [3, 4, 9]. However, Drabkin [6] concluded that thyroxine inhibits the restoration of the mass of the liver after its resection. According to Peters [10], thyroxine stimulates regeneration of the liver only if thyroid function is reduced. There is thus no general agreement regarding the character of the effect of thyroid hormones on regeneration of the liver.

The problem at issue was whether the beneficial action of thyroid preparations on regeneration of the liver can be stabilized by the use of moderate doses combined with theophylline. Theophylline facilitates the preservation and accumulation of cyclic AMP, a substance with an important role in glycogenolysis. It was expected that the breakdown of glycogen would be increased by the action of thyroid preparations and this, in turn, would stimulate regeneration of the liver. The author has previously shown that theophylline, in conjunction with glycogenolytic hormones such as adrenalin and glucagon, stimulates regeneration of the liver [1].

## EXPERIMENTAL METHOD

Experiments were carried out on adult male albino rats. In one of the three series of experiments the animals were given thyroid extract, as an aqueous suspension, in a dose of 10 mg/100 g body weight twice a day by gastric tube. In the other two series of experiments the rats received triiodothyronine by the same route in increasing doses - from 10 to 25  $\mu$ g/100 g body weight twice a day. Theophylline was given internally in a total daily dose of 15 mg/100 g body weight, twice a day. Treatment with these preparations began on the day after partial hepatectomy and continued for 5 days. The animals were decapitated on the 7th day after the operation. In the experiments of series II and III, the oxygen consumption was measured, on the day before the rats were sacrificed, by Kalabukhov's method [2]. The weight of the liver and adrenals and the glycogen content in the liver were determined. The liver samples weighing 0.5 g were defatted in

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Department of Physiology of Animals, N. G. Chernyshevskii Saratov University. Central Scientific-Research Laboratory, Saratov Medical Institute. (Presented by Academician of the Academy of Medical Sciences of the USSR A. P. Avtsyn.) Translated from *Byulleten' Éksperimental'noi Biologii i Meditsiny*, Vol. 78, No. 8, pp. 88-90, August, 1974. Original article submitted October 15, 1973.

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TABLE 1. Effect of Thyroid Extract and Triiodothyronine, with or without Theophylline, on Regeneration of the Liver

Series	Treatment	Body wt. (g)	wt. of liver (mg)	Wt. of liver/100 g body wt.		Wt. of adrenals, in mg/100 g body wt.	Glycogen (mg/g)	Oxygen consump. (ml)
				(in mg)	(in %)			
I	Control	210 (11)	6 305±157	3000,2±89	100,0	—	—	—
	Thyroid extract	204 (11)	6 481±204	3177±164	105,8	—	—	—
	Thyroid extract + theophylline <i>P</i>	192 (7)	7 301±363 <0,005	3802±88 <0,001	126,6	—	—	—
II	Control	203 (8)	6 231±239	3069±116	100,0	20,3±4,3	32±3,7	193±11,3
	Triiodothyronine <i>P</i>	213 (8)	6 894±365 >0,05	3236±132 >0,05	105,4	22,5±4,5	8,8±0,7 <0,001	290,3±9,7 <0,001
	Triiodothyronine + Theophylline <i>P</i>	199 (8)	7 444±298 <0,02	3741±335 >0,05	121,6	24,0±3,2	5,3±0,76 <0,001	332±12,4 <0,001
III	Control	333 (9)	8 005±595	2404±174	100,0	17,5±0,86	28,8±3,8	103±4,3
	Triiodothyronine <i>P</i>	309 (9)	8 783±1 130 >0,05	2842±280,8 >0,05	118,2	22,4±0,88	6,7±0,8	161±10,8 <0,001
	Theophylline	312 (8)	8 100±399	2596±100	107,9	22,4±1,1	27,4±3,6	118±8,5
	Triiodothyronine	311 (8)	10 944±977 <0,05	3519±174 <0,001	146,3	25,3±2,1 <0,01	5,3±0,7 <0,001	171±9,4 <0,001

Note. Number of animals is given in parentheses.

anhydrous acetone and dehydrated in absolute ethanol. The degree of decrease in weight of these pieces of liver was calculated. The content of DNA in 50 mg of tissue treated in this way was determined by the diphenylamine method [5]. In addition, the number of nuclei in 50 fields of vision of the microscope and the number of binuclear cells in 100 fields of vision were determined in paraffin sections through the liver under an immersion objective.

The structure and results of the experiments are given in Table 1.

## EXPERIMENTAL RESULTS

As Table 1 shows, thyroid extract did not affect the weight of the regenerating liver. In conjunction with theophylline, however, it led to a substantial increase in both the absolute and the relative weight of the liver. Triiodothyronine led to a small increase in the absolute and relative weight of the regenerating liver, which was not statistically significant; it also caused a significant decrease in the glycogen content and an increase in the oxygen consumption. In conjunction with theophylline, however, thyroid hormone caused a significant increase in the absolute weight of the liver in the experiments of series II and III and a statistically significant increase in the relative weight of the liver in series III. In both series of experiments in which triiodothyronine was given, especially in conjunction with theophylline, the weight of the adrenals was increased.

Histological treatment revealed no significant difference in the number of nuclei or of binuclear cells in the liver of the rats of the various experimental groups. The DNA content in the various groups also was closely similar.

The increase in weight of the regenerating liver in the rats receiving triiodothyronine in conjunction with theophylline could not be attributed to the accumulation of lipids or water in the organ, for the degree of decrease in weight of the pieces of liver after defatting and dehydration was very similar in all groups. By itself theophylline did not affect the weight of the regenerating liver. Since theophylline potentiated the effect of thyroid hormone on regeneration of the liver, it can be postulated that this effect is based on an increase in the synthesis of cyclic AMP.

Why is it that neither thyroid extract nor triiodothyronine, in the doses used, had any significant effect on restoration of the weight of the liver? Presumably in the present experiments the synthesis of cyclic AMP was stimulated by thyroid extract or thyroid hormone, but in an amount sufficient only to mobilize the glycogen. If theophylline was given additionally, the quantity of cyclic AMP was increased. It became sufficient not only for the mobilization of glycogen, but also for the activation of the hepatocytes, with the consequent stimulation of regeneration of the liver.

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