pregnancy (Table I). We have no explanation for this.

Summary. By means of countercurrent distribution and paper chromatography, estrone, estradiol and estriol were separated from extracts of one to 2 liters of human pregnancy plasma, quantitated and identified by fluorimetric and isotopic technics. Concentration of estrone and estriol remained more nearly constant (3-9 μg/100 ml of plasma) through second and third trimesters, but the concentration of estradiol varied from 0-8 μg/100 ml.

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Enhancement of Lethal Action of Endotoxin in Mice by Triiodothyronine. (25011)

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Mice survive lethal doses of endotoxin when pre-treated with cortisone(1). Consideration of interrelationship between thyroid hormone and resistance of mice to endotoxin resulted from observations in dogs in that cortisol production and metabolism were significantly increased by analogs of thyroxin, such as triiodothyronine and triiodothyroacetic acid. In our studies for publication it was found that the magnitude of the rise in cortisol production simulated secretory response of the adrenal to exogenous corticotropin. Others(2) previously reported that following injection of l-thyroxine into guinea pigs urinary output of 17 hydroxycorticosteroid was in direct proportion to dose of thyroxine. Because triiodothyronine increased output of cortisol in the intact animal, it was anticipated that its administration to mice would protect them against lethal action of endotoxin, but on the contrary, triiodothyronine enhanced this lethal action.

Methods. Purified endotoxins were prepared from cultures of *Escherichia coli, Salmonella typhosus* and *Brucella abortus,* as described elsewhere(1). Male ABC mice weighing approximatley 20 g were given endotoxin intraperitoneally in dose of 0.2 to 0.4 mg/20 g body weight. The dose of endotoxin selected for each experiment was that amount which did not produce death in normal mice within 48 hours. A solution of 3:5:3′-l-triiodothyronine was prepared to yield a concentration of 40 μg/0.1 ml.* Groups of 10 mice each were used in a series of experiments. In the first experiment, 20 mice were given 40 μg of triiodothyronine intraperitoneally, and 18 hours later this dose was repeated, a total of 80 μg within 24 hours. Preliminary metabolic studies revealed that within 6 to 12 hours after final injection metabolism was accelerated, reaching a peak within 24 hours. Then, 10 mice served as controls, and the 10 remaining animals received intraperitoneally 0.3 mg/20 g of *E. coli* endotoxin. An additional group of 10 mice received only endotoxin. In a second series of experiments the same procedure was carried out except that 0.4 mg/20 g of endotoxin prepared from *Br. abortus* was used. Similarly, in the third series, 0.2 mg/20 g of endotoxin made from *S. typhosus* #360 was injected.

Results. Results of 3 experiments using 3 different preparations of endotoxin are presented in Table I. Triiodothyronine consistently enhanced the lethal action of endotoxin. Deaths were recorded as having taken place within 48 hours after mice had received endo-

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TABLE I. Enhancing Effect of Triiodothyronine on Lethal Action of Endotoxin in Mice in 3 Experiments.

Source of endotoxin	Deaths within 48 hr*	
	Endotoxin	Triiodothyronine and endotoxin
E. coli	1/10	9/10
Br. abortus	0/10	6/10
S. typhosus	1/10	7/9
Total	2/30	22/29

* Numerator signifies No. dead.

toxin. Mice surviving usually do not succumb to lethal effect after this period. The majority of animals in our experiments died within 24 hours after having endotoxin injected into them. None of the control mice receiving only triiodothyronine died. There were no unusual gross lesions of organs in animals dying within 48 hours. The outstanding feature summarized in Table I is that only 2 out of 30 mice receiving endotoxin alone died, whereas 22 of 29 mice receiving both triiodothyronine and endotoxin died.

Comment. Cortisone has consistently protected mice against the lethal effect of endotoxin. Since it had been established in the intact dog that triiodothyronine administration resulted in an increase in cortisol production, it was anticipated that enhancement of metabolism in the mouse by triiodothyronine would afford protection against endotoxin. On the contrary, the lethal action of endotoxin was accelerated shortly after administration of triiodothyronine. The mechanism whereby triiodothyronine enhances lethal action of endotoxin is not understood. It could be postulated that the induced state of hyperthyroidism greatly accentuated inactivation of steroid so that adrenal steroid synthesis was exceeded, and adrenal insufficiency occurred even in presence of maximal adrenal secretory activity. In dogs pretreated with analogs of thyroxine, rate of disappearance of cortisol from plasma is greatly accelerated. Furthermore, in humans with thyrotoxicosis rate of removal of cortisol from plasma is markedly accelerated(3). Removal of cortisol from plasma is a reflection of rate of catabolism of the hormone by the liver. *In vitro* reduction of ring A (inactivation) of cortisone by Δ^4-steroid hydrogenases of rat liver is enhanced by triiodothyronine(4).

A second and more plausible explanation relates to the basic effect of endotoxin on vascular activity. Immediately after administration of endotoxin a severe degree of vasoconstriction takes place. This in turn results in a state of anoxia involving certain organs and tissues. Since we have observed that triiodothyronine markedly increases oxygen consumption of mice, it is most likely that the increased demands for oxygen induced by both endotoxin and by triiodothyronine account for the accelerated death rate.

Summary. Triiodothyronine enhances the lethal action of endotoxin in mice. It is postulated that this enhancement is associated with anoxia, caused by vasoconstriction and subsequent hypotension that is induced by the endotoxin; and the increased demand for oxygen that is produced by triiodothyronine.

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