tive abdominal adhesions in many of the animals receiving the intravenous injection of plasminogen activators probably indicates a lysis of the traumatic inflammatory exudate. It is highly unlikely that the effects of the plasminogen activators can be entirely attributed to a lack of formation of a fibrinous exudate since therapy was not instituted for a full day following trauma. The presence of increased capillary permeability in the traumatic area may account for the appearance of plasmin or of SK at the site of exudation. Although the streptokinase preparation contained streptodornase and streptococcal hyaluronidase, both of which may contribute to the lysis and resorption of the exudate, we have stressed the fibrinolytic system since fibrin is the major insoluble component of the experimental exudate in this study.

The differences observed with the 2 types of activators used in this study, and the special fibrinolytic activity seen after *in vivo* injections of SK alone, have been previously noted (2). Recent studies indicate that the mixture of SK plus the human plasminogen preparation supplies the activator for the animal plasminogen(5,9). The special fibrinolytic effects seen with large doses of SK alone are related to the adsorption of SK on fibrin, and the ability of SK to suppress antiplasmin activity(10). The resultant of these latter phenomena is to provide an "inhibitor free" fibrin for the action of activated plasmin(10).

Conclusion. In dogs with an experimental traumatic peritonitis, the intravenous injection of streptokinase alone in large doses, or of a mixture of smaller amounts of SK plus a partially purified human plasminogen preparation, prevented or modified the development of postoperative adhesions.

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Electrophoretic Distribution of Hexosamine in Plasma Proteins of the Rat Following Thyroidectomy.* (21922)

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Plasma hexosamine levels increase in the rat following either thyroidectomy or hypophysectomy(1). The cause appears to be a deficiency of thyroid hormone in both instances. Hexosamine is present in all plasma protein fractions but is richest in the globulins(2). Since globulins have been reported to be elevated in rats following thyroidectomy (3), electrophoretic studies were carried out to establish which protein fractions were responsible for the plasma hexosamine changes observed in the rat following thyroidectomy.

Methods. Male Sprague-Dawley rats, weighing 55-65 g, were individually caged, fed a high calcium diet(4), and permitted to drink water *ad libitum*. Thyroidectomy was per-

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TABLE I. Electrophoretic Distribution of Plasma Proteins and Hexosamine in Plasma Proteins of Normal and Thyroidectomized Rats.*

Group	Albumin	Globulins α-1	Globulins α-2	Globulins β	Globulins γ	Total
			Protein (% of total)			
Control	31.8 ± 0.2	18.1 ± .7	14.0 ± .3	17.5 ± .7	18.6 ± 1.3	—
Thyroidectomy	29.7 ± 1.0	16.3 ± .8	12.7 ± .6	18.8 ± 1.1	22.6 ± 1.0	—
Significance (P)	<.10	<.15	<.10	<.40	<.03	
			Hexosamine (mg/100 ml of plasma)			
Control	5.7 ± 0.8	48.2 ± 3.1	21.4 ± .7	19.6 ± 1.3	9.7 ± 1.4	104 ± 4.8
Thyroidectomy	8.8 ± 1.1	50.6 ± 2.4	21.3 ± .7	28.6 ± 2.1	18.8 ± 1.5	128 ± 4.6
Significance (P)	<.05	>.50	>.50	<.01	<.01	<.01
			Hexosamine (% of protein)			
Control	.30	4.42	2.55	1.87	0.87	1.73

* Mean ± stand. error. Each group contained 6 rats.

formed under ether anesthesia. All rats were weighed on the day of operation and at 12 and 35 days following the day of operation. The thyroidectomized rats used for study were selected on the basis of plateaued growth curves, a measure of complete removal of the thyroid gland. Blood samples were obtained 35 days after the day of operation from the external jugular vein under light ether anesthesia, using oxalate as an anticoagulant. Hematocrit and hexosamine determinations were carried out as previously described, with plasma hexosamine levels being corrected for hematocrit changes(4,5). Electrophoretic separation of the plasma proteins was carried out on filter paper in an apparatus similar to that used by Durrum(6). Whatman No. 1 paper sheets or strips with a resistance area of approximately 500 cm^2 were used. Electrophoretic separations were carried out at room temperature, at constant current (10mA) in pH 8.6 veronal buffer (ionic strength 0.06) for 11-16 hours. The voltage varied with conditions of evaporation in the system. For protein studies 0.01-0.02 ml of plasma were applied on a 25 mm line. For hexosamine studies 0.1 ml was applied to a 150 mm line. At the completion of a run the paper was dried in an oven at 110°C for 20 minutes, stained for 7 minutes in an alcoholic solution of 0.1% bromphenol blue saturated with mercuric chloride(6), rinsed in tap water and dried again in an oven at 110°C. For protein studies the strip was read at 600 mμ in a Beckman spectrophotometer (model DU) by means of a specially constructed strip holder. A protein distribution curve was obtained by charting optical density (ordinate) against distance migrated (abscissa). The protein distribution was measured by cutting out the charted electrophoretic curve, weighing it and relating the weight of each component to the total weight.‡ The electrophoretic distribution of hexosamines was determined by cutting the paper according to the known protein bands. Each band of paper was hydrolyzed in 3 ml of 1N hydrochloric acid for 15 hours and after isolation on Dowex-50, hexosamine was determined by a modification of the method of Elson and Morgan(5).

Results. The distribution of plasma proteins following thyroidectomy is shown in the upper part of Table I. There was a significant increase in the relative amount of gamma globulin. The beta fraction was unaltered but the albumin and alpha fractions were relatively reduced. An example of the changes seen following thyroidectomy is shown in Fig. 1.

The concentration of total plasma hexosamine increased following thyroidectomy (middle part of Table I). This change could

‡ The dye-binding capacities of albumin and globulins differ from one another(7) so that the protein values are not those one would get with moving boundary electrophoresis, where albumin values run higher and globulins lower. It was shown in these experiments that the measurements used followed the Beer-Lambert law. This was established by scanning stained strips prepared by electrophoretically separating serum proteins of known concentration. It was shown that the areas (as determined by O. D. readings) under each band were proportional to the original concentrations of protein.

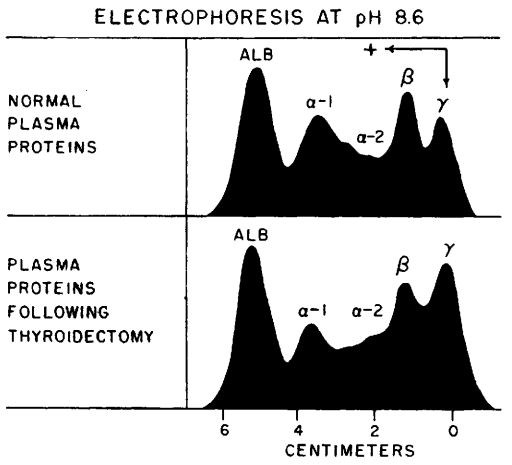

FIG. 1. Effect of thyroidectomy on plasma proteins of the rat.

be accounted for almost entirely by a significant increase in the hexosamine content of both the beta and gamma globulin fractions. The albumin hexosamine was also increased but this contributed little toward the total plasma hexosamine change. The alpha globulins were unaffected. With the known normal distribution of plasma proteins and hexosamine in plasma proteins (Table I), the concentration of hexosamine in each fraction can be estimated (assume a normal plasma protein concentration of 6%). As can be seen in the lower part of Table I, the alpha globulins were the richest hexosamine-containing proteins with albumin and gamma globulin having the lowest concentrations.

Discussion. Normal rat plasma contains all the major protein components seen in human plasma. Whereas human plasma normally contains more alpha-2 globulin, the reverse appears to be true in rat plasma. Many workers have reported difficulty or inability to demonstrate alpha globulins in normal rat serum with the use of moving boundary electrophoresis(8-13). Experience in our laboratory has shown that the separation of the alpha globulins by zone electrophoresis, although much slower with rat than with human plasma, will result with more time or higher voltage. It is likely that the reported absence of alpha globulins by Moore *et al.*(8,9) was in actuality a failure to separate albumin from alpha globulins. That rat plasma normally contains alpha globulins has clearly been demonstrated here and elsewhere(12).

The distribution of hexosamine in normal rat plasma proteins is similar to that seen in human serum(2), being richest in the alpha globulins(14,15).

The plasma mucoprotein fraction described by Winzler(16) is rich in hexosamine and migrates electrophoretically at pH 8.4 with alpha globulins(17). Mancini, Garberi, and de la Balze(18) have reported that in severe myxedema the mucoproteins, as determined chemically by Winzler's(16) method, are increased. On the other hand, Lewis and McCullagh(19) reported that the alpha-globulins are increased in hyperthyroidism and decreased in hypothyroidism. In support of this is the observation that plasma mucoproteins, as measured electrophoretically at pH 4.5, are increased in hyperthyroidism and decreased in hypothyroidism(20). These divergent observations are difficult to resolve without further study.

In the rat the alpha globulins do not appear to be altered following thyroidectomy. This is contrary to the observations of Moore, Levin, Smelser(9), who found that alpha globulins were absent in normal rats but appeared following thyroidectomy (see above), but is in agreement with the earlier findings of Levin and Leathem(3) that serum globulins in the rat, as measured chemically, increase following thyroidectomy. The findings here indicate that the greatest protein increase is in the gamma globulin fraction and that both the beta and gamma globulin fractions account for the increase in plasma hexosamine seen following thyroidectomy(1).

Since thyroidectomized rats do not develop the alpha-globulin changes which are seen in humans with hypothyroidism this probably represents a species difference.

Plasma fibrinogen is closely associated with, but poorly separated from, beta globulins(11) in the rat. It could not be clearly distinguished in the studies reported here.

Summary. Following thyroidectomy the rat develops a relative increase in gamma globulin. The increase in total plasma hexosamine levels following thyroidectomy can be

almost entirely accounted for by the increase in the hexosamine content of the beta and gamma globulin fractions. The normal distribution of hexosamine in rat plasma is as follows: Albumin 5%, alpha globulins 67%, beta globulins 19%, and gamma globulin 9%. The richest fraction is alpha-1 globulin which contains approximately 4.4% hexosamine. The other globulins contain less and albumin contains only 0.30%.

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Stimulation of Adenine Uptake into Mouse Lung and Liver *in vitro* By Tumor Extracts.* (21923)

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Kelly and Jones(1) observed that repeated injections of tissue mashes into normal animals bring about changes in the nucleic acid turnover of the spleen and liver. Our studies (2) have indicated that single injections of a tumor homogenate into normal mice alter the incorporation of adenine-8-carbon^{14} (C^{14}) into the desoxyribonucleic acid (DNA) of several tissues. Fractions obtained from tumor homogenates by centrifugation or pH adjustment were tested for the presence of the factor(s) responsible for this effect by injection into normal mice simultaneously with adenine-8-C^{14} solution. Increase of lung DNA C^{14} activity in animals which had received the tumor preparations was taken as an indication that the factor(s) was present in the preparation. Goldwasser(3) reported that an *in vitro* system consisting of rat liver slices or of cell-free homogenates of pigeon liver incorporated adenine into ribonucleic acid but not into DNA. In simplifying the assay procedure for the tumor factor(s), the authors have developed an *in vitro* system in which significant increases in DNA C^{14} radioactivity

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