the presence of autoclaved chick pancreas (Table II).

From the results of this study maximum liberation of PGA from egg yolk occurs when egg yolk conjugase and chick pancreas conjugase act together.

Summary. Evidence is presented to show that chicken and turkey egg yolk contain a conjugase which liberates PGA from these materials. A synergistic action of egg yolk conjugase and chick pancreas is required in order to obtain the total PGA in egg yolk of chickens and turkeys. The liberation of microbiologically available PGA from egg yolk by the combined action of egg yolk conjugase and chicken pancreas conjugase gives a total PGA value which approaches the value obtained by the chick assay for this vitamin. An inhibitory effect of autoclaved chick pancreas on the conjugase of chicken egg yolk is indicated by the results.

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Effect of Thyroxine on Lymphoid Tissue Mass of Adult Male Mice.* (17326)

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That the secretion of the thyroid gland is intimately related to the physiology of lymphoid tissue has been suggested by both clinical observation and laboratory experiment. Marine and co-workers[1] emphasized that peripheral lymphadenopathy, splenomegaly, and persistence of the thymus gland were detectable in many patients with Graves' disease. Several investigators[2-4] have reported that stimuli which cause accidental involution of the lymphoid tissue of laboratory animals may have a more pronounced or more prolonged effect in the absence of the thyroid gland.

An important link in the chain of evidence —namely that augmented thyroid secretion results in hyperplasia of lymphoid tissue— has previously evaded demonstration despite several experimental trials.[3,5] One possible explanation for this failure lies in the fact that the rat was the experimental animal employed in most of these investigations. Evidence is accumulating that this animal, in the resting state, may be secreting thyroid hormone at a high rate;[6] thus the addition of small doses of exogenous hormone might well be without significant effect. Large doses of hormone may produce relative malnutrition and involution, rather than hyperplasia, of lymphoid tissue.[7] It was, therefore, considered desirable to observe the consequences of thyroxine treatment in the mouse. Profound changes in the lymphoid tissues were observed. In view of the known alterations in adrenal cortical activity following treatment with thyroid hormone,[8,9] a parallel experiment was conducted in adrenalectomized animals to study the possible contribution of this organ

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1 Marine, D., Manley, O. T., and Bauman, E. J., *J. Exp. Med.*, 1924, **40**, 429.

2 Selye, H., *Endocrinol.*, 1937, **21**, 169.

3 Reinhardt, W. O., and Wainman, P., PROC. SOC. EXP. BIOL. AND MED., 1942, **49**, 257.

4 White, A., and Dougherty, T. F., *Endocrinol.*, 1947, **41**, 230.

5 Korenchevsky, V., and Hall, K., *Biochem. J.*, 1941, **35**, 726.

6 Meites, J., and Chandrashaker, B., *Endocrinol.*, 1949, **44**, 368.

7 Andreasen, E., *Acta path. et microbiol. Scand.*, 1939, **15**, 259.

8 Lowenstein, B. E., and Zwemer, R. L., *Endocrinol.*, 1943, **33**, 36.

9 Deane, H. W., and Greep, R. O., *Endocrinol.*, 1947, **41**, 243.

to the changes observed in the intact animal.

Methods. Adult male mice of strain CBA (Strong) were employed. Littermate pairs were assigned to one of two parallel experiments. In the first, which tested the action of thyroxine in intact animals, one member of each pair was assigned at random to the treatment group while its littermate served as a control. In the second experiment, all animals were adrenalectomized, and again the pairs were divided at random into thyroxine-treated and control groups.

Until they reached the age of 11 weeks, the animals were raised on Purina Laboratory Chow and tap water. The diet was then changed to one of bread and milk to which was added whole dried Brewers' Yeast to 5% by dry weight. Both diets were fed *ad libitum*.

Treatment was begun when the animals were 13 weeks of age. The experimental animals received daily intraperitoneal injections of 15 γ of thyroxine‡ in 0.1 ml of slightly alkaline distilled water. Controls received an equal volume of solvent prepared exactly as that employed for the thyroxine. The treatment period was 21 days for the intact animals; 14 days for the operated animals.

Bilateral adrenalectomy was performed in one stage via the lumbar route. Ether and light nembutal anesthesia were employed. For the first 3 post-operative days, 5% glucose and 0.9% sodium chloride were added to the drinking water of the operated animals. For the remainder of the experimental period, the drinking water contained only 0.9% sodium chloride. Operations were performed when the animals were 12 weeks of age; adrenalectomized animals were thus on the experimental diet for one week preoperatively, and were allowed a post-operative interval of one week before thyroxine therapy was instituted.

Eighteen hours after the last injection, the animals were killed by cervical dislocation and complete autopsies carried out. These were performed in a routine manner and included, in addition to the general examination, an inspection of the operative site for post-operative infection or evidence of regenerated adrenal remnants or hypertrophied accessory adrenal tissue. The following organs were dissected free of surrounding tissue and weighed on a precision torsion balance: both adrenals, both kidneys, the spleen, the thymus gland, and a sample of the peripheral lymph nodes which consisted of the two axillary nodes from each side and the two epigastric nodes. Body weights were measured on a double-beam balance at the beginning of therapy and at autopsy.

Statistical analysis involved the application of Fisher's "t-test" for small groups. In this report the word "significant" will be used only to describe differences wherein the analysis has indicated a probability value of the order of 0.01 or less.

Results. The results of the two experiments are collected in Table I. Since body weights do not differ significantly, absolute rather than relative values are given. For the adrenal glands, the range of the weights rather than the standard error is noted as the experimental error is thought to be greater than that due to sampling.

The administration of thyroxine to intact animals resulted in a significant increase in the weights of the spleen, peripheral lymph nodes, and kidneys. A suggestive rise in adrenal weights was observed. The weights of the thymus glands of the treated animals were less than those of their controls. While this difference amounts to almost 20% of the control values, and has a p-value of less than 0.01, the results must be regarded with caution as the sampling error is slightly less than the error generally accepted for the weights of dissected organs ($\pm$ 1 mg).

The reduced tolerance of adrenalectomized animals to thyroid hormone[10] was illustrated by the fact that 7 of 17 animals so treated died within 2 weeks, while all but one of the control group survived. Because of this serious loss, the experiment was terminated after a treatment period of 14 days. The results reported are those obtained for the littermate pairs in which both members survived this

‡ l-thyroxine, Squibb.

[10] Koelsche, G. A., and Kendall, E. C., *Am. J. Physiol.*, 1935, **113**, 335.

TABLE I.
Body Weights and Organ Weights of Adult Male Mice Treated with Thyroxine.

Group	No. of mice	Body wt (g)		Organ wt (mg)				
		Initial	Final	Spleen	Lymph nodes	Thymus	Kidneys	Adrenals
Intact Control	14	28.3 ±0.5	29.7 ±0.5	103 ±3.4	32 ±1.2	27 ±0.9	470 ±9.6	4 (3-5)‡
Intact Thyroxine*	14	28.1 ±0.5	29.5 ±0.5	170 ±8.2	53 ±4.1	22 ±0.9	586 ±13.3	5 (4-6)‡
Adrenalectomized Control	9	27.4 ±0.6	28.3 ±0.6	161 ±5.3	47 ±1.8	45 ±1.2	441 ±10.8	—
Adrenalectomized Thyroxine†	9	27.3 ±0.6	28.2 ±0.7	224 ±5.6	61 ±3.2	53 ±3.5	500 ±27.6	—

* 15 γ per day for 21 days.
† 15 γ per day for 14 days.

‡ Range of values. Other values are means and standard error by $\sqrt{\frac{\Sigma d^2}{n(n-1)}}$

length of time. These results illustrate the well-known effects of adrenalectomy on lymphoid tissues. In addition the weights of the spleen and lymph nodes of the thyroxine-treated animals are significantly higher than those of their controls. The thymus weights showed an increase of limited significance ($p = <0.05$). The shorter course of thyroxine treatment employed for this group did not result in a significant change in kidney weights ($p = >0.05$). Body weight values demonstrate that the dietary regimen adequately maintained the operated animals; and the stability of these values indicates that the animals are well past the period of most active growth (4 to 8 weeks).[11]

Discussion. While quantitative histologic studies were not performed, microscopic examination suggested that much of the increase in lymphoid tissue mass was due to hyperplasia of these organs. In response to either thyroxine or adrenalectomy, the spleen and peripheral nodes registered an increase in weight of over 50%. The two treatments appeared to be additive and together resulted in a doubling of the weights of these organs. That this increase following thyroxine treatment occurred in animals showing essentially stable body weights indicates that the effect of this treatment was quite distinct from any that might result from a change in rate of growth.[12] Furthermore, exhaustion of the adrenal cortex[9] does not appear to have contributed to this effect, since treatment of adrenalectomized animals with thyroxine resulted in an increased weight of the spleen and peripheral nodes over and above that following adrenalectomy alone. If anything, the data for the adrenal and thymic weights of the first experiment suggest augmented adrenal cortical activity during the period studied. The slight decrease in thymic weights of intact animals treated with thyroxine appears not to have occurred in the adrenalectomized animals. This behavior of the thymus, in contrast to the other lymphoid tissues studied, is consistent with its greater sensitivity to the action of adrenotrophic hormone.[13,14]

An increase in kidney weight has been previously shown to follow thyroid treatment in rats[15] and testosterone treatment in mice.[16] In both instances, the renal hypertrophy was

11 Morris, H. P., *J. Nat. Cancer Inst.*, 1944, **5**, 115.

12 Koger, M., Hurst, V., and Turner, C. W., *Endocrinol.*, 1942, **32**, 237.

13 Dougherty, T. F., and White, A., PROC. SOC. EXP. BIOL. AND MED., 1943, **53**, 132.

14 Simpson, M. E., Li, C. H., Reinhardt, W. O., and Evans, H. M., PROC. SOC. EXP. BIOL. AND MED., 1943, **54**, 135.

15 MacKay, E. M., and MacKay, L. L., *J. Nutrition*, 1931, **4**, 33.

related to an increased rate of protein metabolism. It is perhaps significant that adrenal cortical hypertrophy, well known to occur under thyroid treatment, is also thought to be a response to an increased rate of protein utilization.[17] In addition, evidence has recently been presented that lymphoid tissue may represent an important source of metabolic protein[4]—a source limited in volume, but of extreme lability. While the present experiments record only gross structural alterations, and the possible contribution of these changes to the physiology of the organism must remain in the realm of speculation, it is suggested that an increased rate of nitrogen metabolism may well represent the common denominator underlying the many changes noted in this investigation.

Summary. The administration of thyroxine to adult male mice resulted in an increase in the organ weights of the kidneys, spleen, and peripheral lymph nodes. Similar treatment of adrenalectomized animals resulted in an increase in the weights of the spleen and peripheral lymph nodes over and above that which followed adrenalectomy alone. It is concluded that thyroxine produces an increase of lymphoid tissue mass which is independent of the rate of body growth and of the level of adrenal cortical activity.

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[16] Kochakian, D. C., in Recent Progress in Hormone Research, Vol. 1, New York, Academic Press, Inc., 1947, p. 177.

[17] Tepperman, J., Engel, F. L., and Long, C. N. H., *Endocrinol.*, 1943, **32**, 373.

Failure of Aureomycin in the Treatment of Experimental Tuberculosis. (17327)

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The failure of streptomycin, p-aminosalicylic acid and the sulfone compounds to prove ideal agents for the treatment of tuberculosis has stimulated the testing of many new agents. Because of the remarkably wide anti-microbial spectrum of aureomycin, it seemed worthwhile to study its effect upon experimental tuberculosis.

Guinea pigs weighing 400 to 500 G each were inoculated subcutaneously with 0.02 mg tubercle bacilli from a recently isolated, virulent, streptomycin-sensitive human strain. After 28 days, the animals were divided into two groups. The 12 animals in Group A were treated with daily subcutaneous injection of 5 mg aureomycin hydrochloride, 6 days a week for 6 weeks. The 11 animals in Group B served as infected, untreated controls.

A third group, Group C, consisted of 6 uninfected guinea pigs given aureomycin in the same dosage as Group A, in order to observe possible toxic effects of the drug.

Animals were autopsied as they died, and those surviving after 6 weeks of treatment or 10 weeks after infection were sacrificed and autopsied.

In Table I, it can be seen that mortality was higher in the treated than in the untreated group. Both treated infected and uninfected groups showed marked weight loss, and one of the uninfected animals died during treatment.

All the infected animals showed widespread tuberculous involvement of the regional lymph nodes surrounding the injection site, the liver, spleen, and lungs. Gross and microscopic examination failed to show any appreciable difference between the treated and untreated animals. Acid fast bacilli were demonstrated

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