streptomycin-resistant strain of *Salmonella enteritidis*. In mice treated with streptomycin 24 hours before inoculation, <3 Salmonella sufficed to initiate infection in 50% as compared with approximately 10^5 in untreated controls. 2. This effect of streptomycin decreased as the interval between treatment and inoculation was lengthened but was still detectable on the 5th day. Smaller doses of streptomycin (5-10 mg) resulted in smaller increases in susceptibility. 1 mg was ineffective. 3. Representative numbers of mice killed for culture showed the spleen to be infected in 90% and heart's blood in 72% of those with positive fecal cultures at the time of autopsy. 4. It is believed that this increase in susceptibility following streptomycin treatment resulted from a disturbance of the normal intestinal microflora caused by the antibacterial action of the drug. 5. It is suggested that this method may be applicable to the experimental study of other enteric infections.

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Received April 20, 1954. P.S.E.B.M., 1954, v86.

In vitro Conversion of Thyroxin to Triiodothyronine by Kidney Slices.* (21031)

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(Introduced by O. O. Meyer.)

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Several reported observations suggest that 3-5-3′L-triiodothyronine is derived from thyroxin by deiodination in extrathyroidal tissues (1-3). This concept is supported by the following observations on kidney slices, demonstrating *in vitro* deiodination of thyroxine to triiodothyronine.

Methods. 150-200 g male rats (Sprague-Dawley strain) were used. The animals were sacrificed by decapitation, the kidneys aseptically removed and cut into thin slices. Slices representing approximately one-half of a kidney were placed in each of the Warburg flasks containing 3 ml of Krebs-Ringer phosphate solution supplemented with 0.03 mg glucose. 0.01 μg of chromatographically pure I^{131} labelled thyroxin† were added to each flask. The slices were incubated at 37°C for periods of 0, 3, 6, 9, and 12 hours. Controls were run as follows: To one flask incubated for 12 hours potassium cyanide was added to yield a final concentration of 0.01 M; to a second flask incubated 6 hours potassium iodide was added to yield a final concentration of 0.01 M; in a third flask incubated 12 hours kidney slices which had been boiled for 5 minutes were used; to a fourth flask incubated 12

* This study was supported in part by the Wisconsin Alumni Research Foundation.

† Obtained from Abbott Laboratories.

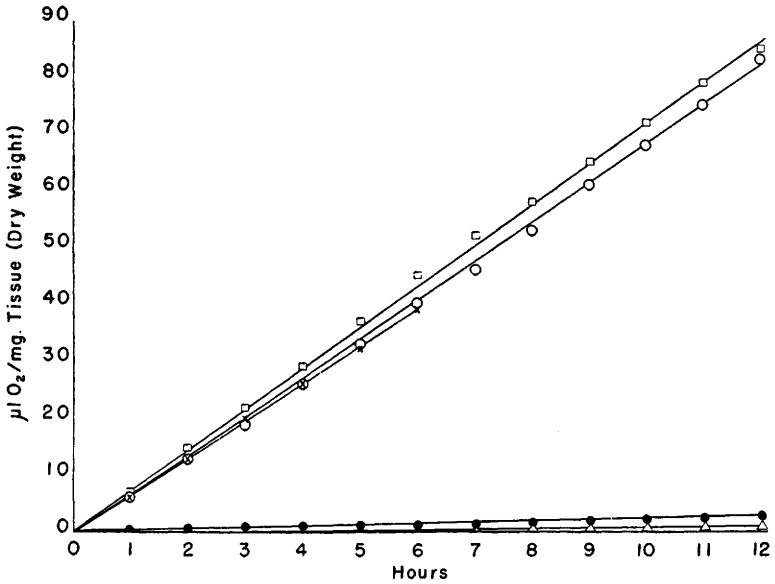

FIG. 1. Oxygen uptake by kidney slices. ○———○ represents untreated tissue; □———□ antibiotic control; ×———× potassium iodide control; ●———● potassium cyanide control; △———△ boiled control.

hours, 1000 units of penicillin G and 100 μg of streptomycin were added. At the end of the incubation period the slices were removed from the media, dried with blotting paper, weighed and washed with water to remove excess radiothyroxin. They were then homogenized in 2 ml of distilled water. The pH of the homogenates was adjusted to 3.0 by the addition of 0.1 N hydrochloric acid. The radioactivity of the entire homogenate was

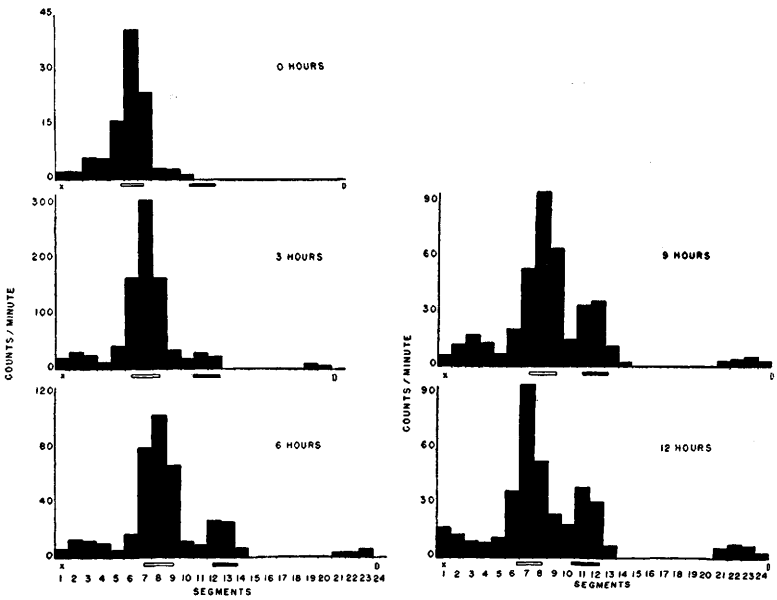

FIG. 2. Distribution of radioactivity in chromatograms following varying periods of incubation. The starting point of chromatography is indicated by ×, the developer front by the vertical bar. The position of thyroxin is shown by the horizontal open bar, that of triiodothyronine by the horizontal closed bar.

determined initially and following each extraction procedure by counting in a well-type scintillation detector. Extraction of the homogenates was carried out with 6 ml portions of water-saturated butanol. Five extractions were necessary for the removal of approximately 95% of the radioactivity. The butanol extracts were pooled and evaporated to dryness at room temperature. The residue was dissolved in 0.1 ml of butanol containing 50 μg of carrier L-triiodothyronine.‡ The final solution was subjected to single dimension chromatography as previously described (4). The chromatograms were cut transversely into 0.5 cm numbered segments and counted in a bell-type end window Geiger counter. After counting, the segments were reassembled and the color of the carrier triiodothyronine was developed as previously reported(4). Coincidence of the radioactivity with the carrier triiodothyronine spot was taken as an indication of conversion of labelled thyroxin to triiodothyronine.

Results. Fig. 1 shows the oxygen uptake of slices, indicating functioning tissue throughout the duration of the experiment. Fig. 2 shows the distribution of radioactivity in the chromatograms following various periods of incubation. The initial peak represents radioactive thyroxin and the second peak corresponds to triiodothyronine. Significant radioactivity begins to appear in the triiodothyronine spot at 3 hours. Increasingly greater concentrations are found at 6, 9, and 12 hours. In Fig. 3 is shown the effect of the addition of potassium iodide, potassium cyanide, antibiotics, and of boiling on the conversion of thyroxin to triiodothyronine. It is clear that the conversion is reduced in the presence of potassium iodide and is abolished in the presence of 0.01 M cyanide or by boiling. The presence of antibiotics did not affect the conversion. The radioactivity localized behind the developer front has not been identified, but is known not to be iodide, which has an Rf value less than that of thyroxin.

Discussion. The demonstration of 3-5-3'L-triiodothyronine in the thyroid gland and in plasma(5-8) and the subsequent observations that it has several times the physiological activity of thyroxin(2,9,10) has led to speculation that it is the peripherally active form of the thyroid hormone. It is evident, however, that thyroxin is present in plasma in

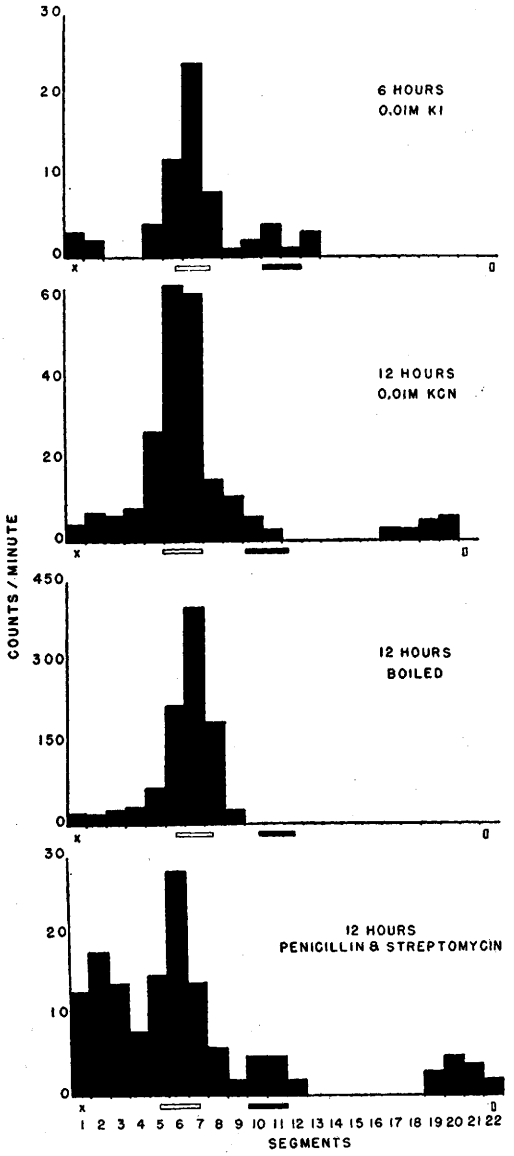

FIG. 3. Distribution of radioactivity in chromatograms of controls. The starting point of chromatography is indicated by ×, the developer front by the vertical bar. The position of thyroxin is shown by the horizontal open bar, that of triiodothyronine by the horizontal closed bar.

‡ Triiodothyronine was kindly supplied by Dr. Arthur Heming of Smith, Kline & French Laboratories.

greater quantities than triiodothyronine(11-13). These observations have led to the suggestion that thyroxin is converted peripherally to triiodothyronine by a process of deiodination. In support of this possibility, Gross and Leblond found a compound, later identified as triiodothyronine, in the organs of thyroidectomized mice following the administration of I^{131} labelled thyroxin(1). These observations have been confirmed recently by Kalant *et al.* in propyl thiouracil-treated animals(3).

The data reported in this paper demonstrate that extrathyroidal conversion of thyroxin to triiodothyronine can occur, at least in the kidney.

Summary and conclusions. 1. Chromatographically pure I^{131} labelled l-thyroxin was added to surviving kidney slices which were incubated in a modified Krebs-Ringer phosphate solution. 2. Following incubation, the slices were homogenized, extracted with butanol and the thyroxin and triiodothyronine separated and identified by paper chromatography. 3. The triiodothyronine was found to be radioactive. This was taken as evidence of deiodination of thyroxin to triiodothyronine. 4. Conversion of thyroxin to triiodothyronine was reduced by excess iodide and abolished by boiling the tissue or by the addition of potassium cyanide to the media.

The authors wish to acknowledge the invaluable assistance of Miss Sue Ames, Medical Technician.

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Received April 15, 1954. P.S.E.B.M., 1954, v86.

Distribution and Properties of an Unidentified Growth Factor for Avian *Lactobacillus bifidus.** (21032)

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Veltre, Shorb, and Pelczar(1) reported that *Lactobacillus bifidus* of avian origin failed to grow in a defined medium unless supplied with a substance(s) found in Phytone, yeast extract, beef extract, or fish solubles. A disaccharide from mucin, galactose acetylglucosamine(2,3), and blood group substances A and B(4) failed to replace the requirement of the avian bifids as these compounds did for the variants of *L. bifidus* from humans. Because large numbers of the *L. bifidus* organisms were associated with a depression of chick growth(5) perhaps by robbing the avian host of some required nutrient, further studies on the distribution and properties of the substance required by these avian bifid organisms have been carried out.

Experimental. An assay procedure has

* Scientific Paper A460 of the Md. Agri. Exp. Station. This study begun under contract between Office of Naval Research, and the University of Maryland. The project has continued under a grant from Merck & Co., Inc., Rahway, N. J. The technical assistance of Maye L. Hansen is gratefully acknowledged.

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