# Hepatic Catabolism of Thyroid Hormone Studied by Radioactive Iodine in Splenic Autotransplants. (19598)

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Knowledge concerning the fate of thyroid hormone following its elaboration and discharge into the circulation is limited. Attempts to delineate the role of the liver in the catabolism of this hormone have yielded conflicting results. Early studies suggesting such a role (Asimoff and Estrin(1), Krayer(2)) used relatively large amounts of exogenous hormonal principles. More recently, Gross and Leblond(3,4), using intravenous injection of radioactively labelled thyroxine in animals, found significant localization in liver, gastrointestinal tract and feces. In their studies on the fate of ingested and infused thyroid hormonal principles in man, Albert and Keating (5), and Myant and Pochin(6) noted increased counting rates over the epigastrium, suggesting hepatic concentration of the material.

Gabe and Arvy(7) concluded from histological studies following pre-portal transplantation of the thyroid gland in rats that the liver completely removed thyroid hormone, whereas Bondy(8) concluded on the basis of protein bound stable iodine levels in a small number of similarly prepared experimental animals that the liver did not destroy and excrete the thyroid hormone to a significant extent. This report deals with the study in rats of radioactive iodine ($I^{131}$) uptake in thyroid tissue autotransplanted into the spleen and the subsequent level of plasma protein-bound $I^{131}$ as an indirect measure of the removal by the interposed liver of endogenously labelled thyroid hormone.

*Methods.* Five groups of male albino Sprague-Dawley rats (150-250 g), maintained on stock Purina Laboratory diet were prepared, using intraperitoneal nembutal anesthesia. I. Thyroidectomized; II. Thyroidectomized, with thyroid gland implanted within muscle in the right axilla; III. Thyroidectomized with thyroid gland implanted within a splenic pouch; IV. Sham operated controls. These first 4 groups were studied, as outlined below, 24-39 days post operatively. A fifth group (Group V) of 11 animals, thyroidectomized with splenic implants, was followed for one year before sacrifice. Each animal was injected intraperitoneally at completion of the operative procedure with 10 cc of 1% calcium chloride in isotonic glucose solution. This solution also served as drinking water for 5-7 days, followed by tap water *ad lib.* thereafter. Weights were recorded periodically. Preliminary *in vivo* "scanning" studies were carried out at intervals. For this purpose 5 $\mu$c of carrier free $I^{131}$, in 1 cc of saline or Krebs-Henseleit buffer were injected intravenously (leg vein). At intervals after injection, external gamma counts were obtained by holding the region of the animal to be counted up to a lead collimated G-M tube (6 mm window), connected to a count-rate meter. For final studies of $I^{131}$ uptake and plasma $I^{131}$ levels, about 15 $\mu$c carrier free $I^{131}$ were injected intraperitoneally. Blood was obtained at 2 and 24 hour intervals by direct cardiac puncture in the immobilized, unanesthetized animal. The protein-bound $I^{131}$ level was determined by trichloracetic acid precipitation(9), expressed as $PBI^{131}$ x 100/total plasma $I^{131}$. Uptake and conversion of $I^{131}$ by transplanted tissue were determined by alkaline hydrolysis and fractionation(10). At the time of sacrifice a thorough search was made for residual thyroid tissue using a G-M tube for scanning purposes plus careful dissection; minute residual fragments in the neck could be detected in this way. The presence of adhesions between the spleen and abdominal wall or other organs was noted. The transplant and

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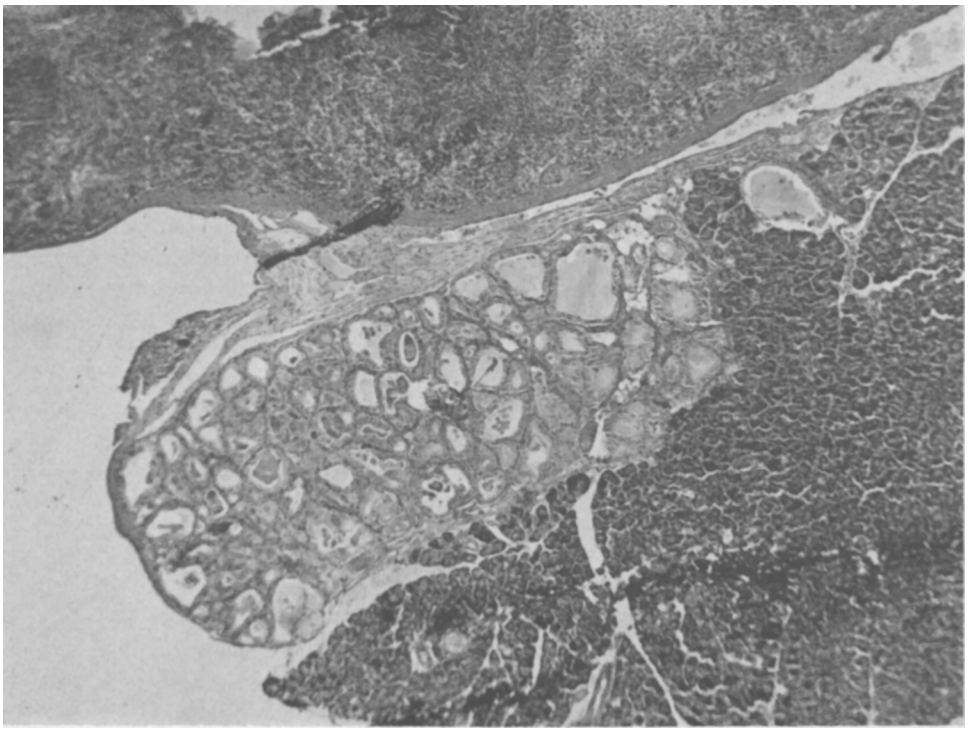

FIG. 1. Transplanted thyroid gland intimately associated with spleen and pancreas.

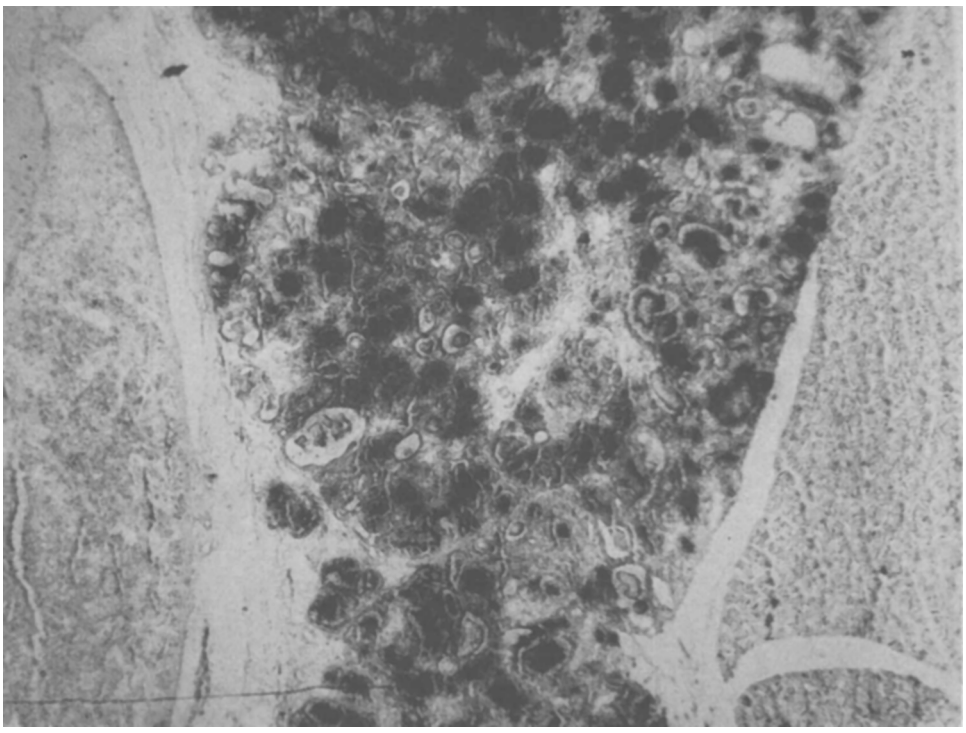

FIG. 2. Autoradiograph of intra-splenic thyroid gland (Eastman NTB plate, 50 $\mu$).

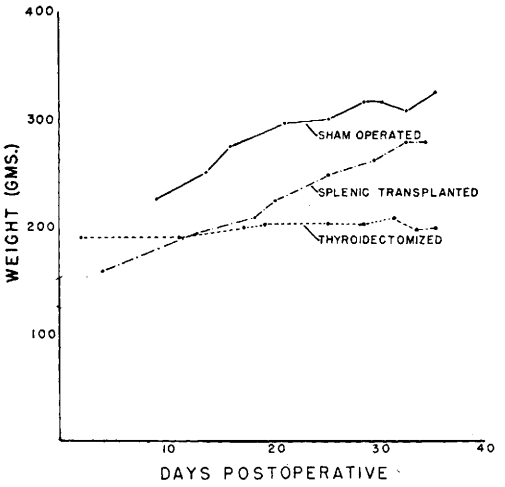


FIG. 3. Effect of thyroidectomy and splenic transplantation on avg wt curves.

residual neck thyroid tissue (if present) were removed for uptake and fractionation determinations and radioautography (Eastman NTB plates, 50 mμ thick). Other organs were fixed in Bouin's fixative for histological studies.

*Results.* Only one of 16 splenic transplanted animals died post-operatively. Viable, $I^{131}$ concentrating tissue was found in 100% of the transplants, including those animals found to have residual thyroid tissue in the neck. There was no evidence of thyroid hyperplasia, hyperfunction or adenoma formation in the transplants (Fig. 1 and 2).

The thyroidectomized animals showed the classical "flattening" of the weight curve post-operatively whereas the curves for those with the splenic transplants showed a progressive weight gain comparable to those of the normal, sham-operated controls (Fig. 3). The splenic transplanted animals of Group V showed a progressive weight gain to values of 520-650 g at the end of one year.

Scanning of body areas by the use of a collimated G.M. tube and count-rate meter made it possible to confirm and follow at intervals in the same animal the presence of a functioning transplant and residual cervical thyroid tissue, if present. In normal animals there was a gradual, progressive rise in counts over the thyroid area for the first 24-48 hours; a comparable rise over the splenic area (attributed to contiguous structures, chiefly kidney) occurred during the first 4-6 hours, followed by a progressive decrease. In adequately thyroidectomized animals, the count over the thyroid area showed a sharp, progressive drop after 4 hours; residual tissue was suggested by a persistently high count beyond this time interval and confirmed, in each instance, by post-mortem examination. Similarly a persistently high level of radioactivity over the splenic area beyond 6 hours suggested a functioning transplant, confirmed at the time of examinations.

The percentage of $I^{131}$ organically bound by transplanted thyroid 24 hours following uptake in 5 animals averaged 72.6 ± 10.9, a value in general agreement with those found in normal rat thyroids(10). The plasma $PBI^{131}$ values at 24 hours following $I^{131}$ injection are presented in Table I.

Residual cervical thyroid fragments were found in 5 operated animals. In 4 of these, the $PBI^{131}$ values equalled or approached those of the controls (avg = 20.9 ± 5.9).

*Discussion.* The data presented are consistent with the interpretation that under the conditions described the liver removes or inactivates a fraction of the thyroid hormone delivered to it.

Failure to effect an athyreotic weight curve, to find evidence for compensatory thyroid hyperplasia or hyperfunction, or to obtain the low $PBI^{131}$ values of thyroidectomized animals indicates that the liver does not completely remove the hormone. This contrasts with the interpretation of Gabe and Arvy(7) who based their conclusions of complete hepatic inactivation of thyroid hormone on histological studies of the pituitary, liver and adrenals of similar experimental animals but published no further data relative to the weight curves or metabolic status of their animals. Bondy (8) concluded that there was no significant hepatic destruction or excretion from data in 3 surviving animals with splenic transplants. He based this conclusion on control values of serum protein-bound stable iodine, but the possibility of residual thyroid tissue or adhesions by-passing the hepatic circulation was not ruled out. In agreement with the finding by Reinhardt(11) that about 50% of rats

TABLE I. Plasma $PBI^{131}$ Levels 24 Hr Following Injection of $I^{131}$.

| Group | No. animals | 24 hr $PBI^{131}$ |
|---|---|---|
| SHAM | 4 | 29.9 ± 8.2 |
| AXILLARY TRANSPL.* | 3 | 23.2 ± 2.9 |
| THYROIDECTOMIZED* | 6 | 3.82 ± 1.6 |
| SPLENIC TRANSPL.* | 7 | 12.2 ± 5.4 |

* Without demonstrable residual cervical thyroid tissue.

subjected to surgical thyroidectomy still contain iodine-concentrating tissue, we found that even minute residual fragments resulted in $PBI^{131}$ values at or approaching control levels.

In evaluating the data presented by Bondy it is further to be noted that since the PBI level is the resultant of 2 processes, *i.e.*, delivery to and removal from the vascular compartment, the interpretation of hormonal delivery on the basis of such levels rests on the assumption that the "peripheral utilization" of the hormone is not significantly altered. Knowledge concerning this latter parameter of thyroid function is limited. The data presented herein might thus be interpreted as representing an increased rate of hormonal utilization despite an unaltered delivery from the thyroid, through the hepatic circulation, to the systemic circulation. Further studies are necessary for a more complete clarification of the qualitative and quantitative role of the liver in the catabolism of thyroid hormone.

*Summary.* The role of the liver in the metabolism of endogenous thyroid hormone, labelled by $I^{131}$ in splenic autotransplants has been studied in rats. Serial observations on the "functional take" of the transplants have been carried out. Data obtained by study of uptake and conversion of $I^{131}$ and the resultant plasma levels of labelled protein-bound iodine indicate a partial removal or alteration of thyroid hormone by the liver under these experimental conditions.

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## Composition of Urinary Calculi from Mink.* (19599)

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The occurrence of urinary calculi in mink is a problem frequently encountered on mink ranches. The incidence of urinary calculi is largely confined to 2 distinct periods of the year. In the spring calculi are found primarily in female mink during pregnancy or soon after whelping. The development of calculi before whelping is generally coincident with the presence of a large number of unborn kits. In the summer months calculi occur mainly in young male kits. In this country the occurrence of urinary calculi in mink appears to be limited primarily to the north central states. This regional difference in calculi formation may be related to the diet. In

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