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# EFFECT OF VARIATIONS IN ACUTE AND CHRONIC IODINE INTAKE ON THE ACCUMULATION AND METABOLISM OF [$^{35}$S]PROPYLTHIOURACIL BY THE RAT THYROID GLAND*

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**Abstract**—Studies were performed to ascertain the effect of varying levels of chronic iodine intake and varying doses of iodide [0.002–100 μmoles KI/100 g body weight (BW)] given acutely on the rat thyroid metabolism of [$^{35}$S]PTU (8.76 μmoles/kg BW). With variations in both acute and chronic iodine intake, as much as four-fold changes in the thyroid content of total $^{35}$S and unchanged [$^{35}$S]PTU were observed. In the low, normal and high chronic iodine intake groups increasing doses of iodide given acutely produced a biphasic response in the thyroid accumulation of total $^{35}$S and unchanged [$^{35}$S]PTU. In each of the three chronic iodine intake groups, increasing iodide doses up to 0.1 μmoles/100 g BW were associated with an increase in total $^{35}$S and unchanged [$^{35}$S]PTU. At this level of acute iodide dosage, the increase averaged 45% ($P < 0.01$). Following acute iodide doses greater than 0.1 and up to 5.0 μmoles/100 g BW, the mean total $^{35}$S and unchanged [$^{35}$S]PTU decreased to levels significantly ($P < 0.001$) lower than those at the peaks. Greater doses of iodide (up to 100 μmoles/100 g BW) produced no consistent further change in the thyroid content of total $^{35}$S or unchanged [$^{35}$S]PTU. Irrespective of the acute iodide dose, the higher the level of chronic iodide intake, the higher the thyroid accumulation of total $^{35}$S and the lower the overall mean percentage of total $^{35}$S present as unchanged [$^{35}$S]PTU. As would have been expected, increasing acute doses of iodide were associated with progressive decreases in thyroid:serum iodide concentration gradients, and the dose of acute iodide required to induce an acute Wolff–Chaikoff effect was greater the higher the level of chronic iodine intake. No correlation was evident, however, between the effects of acute doses of iodide on aspects of intrathyroid iodine metabolism and their effect on the thyroid metabolism of [$^{35}$S]PTU.

The duration and degree of action of antithyroid drugs is probably related to their concentration in the thyroid [1]. It is likely therefore that factors which influence the ability of the thyroid to accumulate antithyroid drugs are important determinants of antithyroid action, and these include $T_4$, TSH, LATS, iodide and phenobarbital [1–4]. In the rat, iodine deficiency has been shown to affect the thyroid accumulation and oxidation of [$^{35}$S]methimazole [5, 6] and [$^{35}$S]propylthiouracil [6]. The present study describes the separate effects of acute and chronic variations in iodine intake on the accumulation and metabolism of [$^{35}$S]6-*n*-propyl-2-thiouracil ([$^{35}$S]PTU) by the rat thyroid gland and explores the relationship between the effects of varying acute doses of iodide on thyroid [$^{35}$S]PTU metabolism and their effects on thyroid iodine metabolism.

## MATERIALS AND METHODS

[$^{35}$S]PTU, carrier-free [$^{125}$I]- and [$^{131}$I]iodide were obtained from the Radiochemical Centre (Amersham, U.K.). [$^{35}$S]PTU was supplied at sp. acts of 103.2–126.8 mCi/mmole and was shown by TLC in several different solvent systems to be greater than 97% radiochemically pure.

*Chronic iodine intake.* Three hundred and eighteen male Sprague–Dawley rats were used in this study. Their chronic iodine intake was one of the following: (a) low-iodine diet (LID) (mean 0.11 μg I/g diet) for 21 days, (b) normal-iodine diet (NID) (2–3 μg I/g diet), or (c) high-iodine intake (HII) (NID + a daily i.p. injection of 100 μg KI in 0.4 ml 0.9% saline) for 27 days. Tap water containing no detectable iodine was available to all rats *ad lib*.

*Measurement of dietary iodine.* Dietary total iodine was measured by thoroughly digesting a known weight of powdered diet with an acid mixture [perchloric acid/nitric acid, 4/1 (v/v)] and analysing the solution for total iodine using a Technicon Autoanalyser. Acid digestion was found to increase considerably the amount of iodine measured in the diet compared to using a supernatant from diet homogenised in deionized water.

*Acute iodide intake.* Immediately prior to acute iodide administration the mean body weight (BW) of most batches of rats was approximately 250 g.

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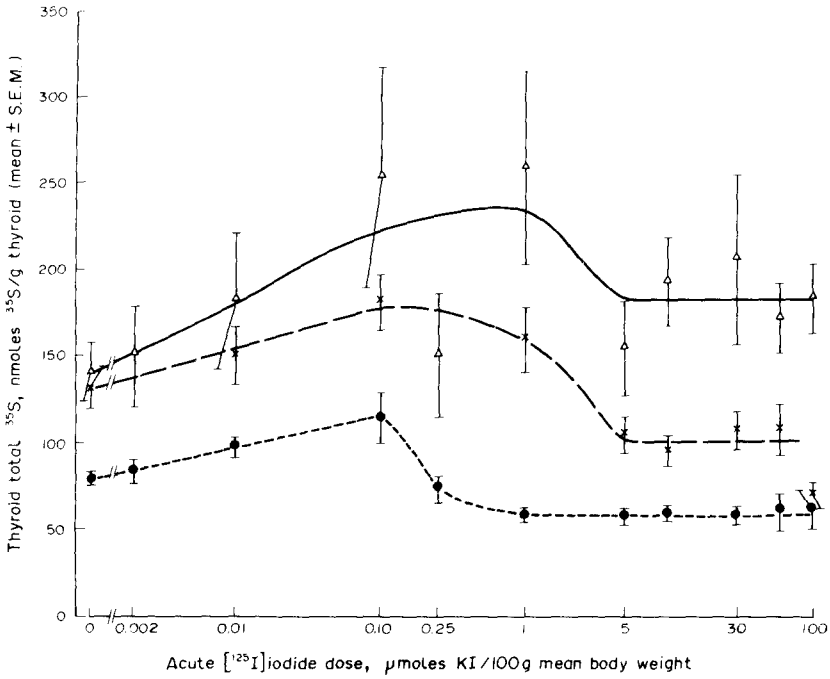


Fig. 1. The effect of chronic iodine intake (●-----●, LID; ×—×, NID; △—△, HII) and acute iodide dose given 1 hr prior to [$^{35}$S]PTU administration on rat thyroid total $^{35}$S level. For LID and HII rats each point is the mean ± S.E.M. of five or six thyroids and for NID rats of 15–18 thyroids. For all of the HII rats and for a third of the NID rats the acute iodide dose was $^{125}$I-labelled. The acute iodide given to LID rats was $^{131}$I-labelled.

Table 1. Effect of chronic and acute iodine intake on thyroid accumulation of unchanged [$^{35}$S]PTU

| Acute iodide (μmoles KI/100 g mean BW) | nmoles unchanged PTU/g thyroid* | | |
|---|---|---|---|
| | LID | NID | HII |
| Control (0.000) | 25.8 ± 3.2 (3) | 54.4 ± 5.8 (17) | 34.8 ± 5.2 (5) |
| 0.002 | | | 44.1 ± 13.3 (6) |
| | | | NS |
| 0.01 | 47.0 (2)† | 52.6 ± 9.3 (17) | 59.3 ± 16.4 (6) |
| | | NS | NS |
| 0.10 | 56.9 ± 8.8 (5) | 78.7 ± 7.5 (16) | 95.6 ± 27.0 (6) |
| | NS | P < 0.02 | NS |
| 0.25 | | | 36.3 ± 11.2 (6) |
| | | | NS |
| 1.0 | 42.3 ± 2.7 (3) | 64.6 ± 10.6 (18) | 107.9 ± 22.2 (6) |
| | P < 0.02 | NS | P < 0.02 |
| 5.0 | 38.0 (2)† | 43.9 ± 5.2 (17) | 55.4 ± 12.4 (6) |
| | | NS | NS |
| 10.0 | 28.7 (2)† | 43.3 ± 4.4 (18) | 64.3 ± 9.3 (6) |
| | | NS | NS |
| 30.0 | 25.6 ± 3.4 (4) | 49.6 ± 5.50 (15) | 71.4 ± 15.5 (5) |
| | NS | NS | NS |
| 60.0 | 32.2 ± 6.8 (5) | 36.7 ± 3.8 (15) | 56.8 ± 4.8 (5) |
| | NS | P < 0.02 | P < 0.02 |
| 100.0 | 33.6 ± 9.9 (3) | 25.9 ± 4.1 (14) | 39.9 ± 7.5 (5) |
| | NS | P < 0.001 | NS |

Figures in parentheses indicate number of animals in each group.
Data expressed as means ± S.E.M.
For all of the HII rats and for a third of the NID rats, the acute iodide dose was $^{125}$I-labelled. The acute iodide given to LID rats was $^{131}$I-labelled.
* P values from Student's *t* quoted relative to control data.
† Mean of two rats.

Each rat received a single i.p. injection of 0.4 ml of either 0.9% saline or potassium iodide (0.002–100 μmoles KI/100 g mean BW). In experiments in which it was desired to correlate the effects of varying acute iodide doses on the thyroid metabolism of [$^{35}$S]PTU with their effects on intrathyroid iodine metabolism, $^{125}$I (the HII rats and a third of the NID rats in Fig. 1 and Table 1) or $^{131}$I (the LID rats in Fig. 1 and Table 1) tracer (0.5–100 μCi) was included.

*Acute [$^{35}$S]PTU administration.* One hour after each acute iodide/saline injection, a single dose of [$^{35}$S]PTU (8.76 μmoles/kg mean BW in 0.4 ml of 0.9% saline) was administered i.p. One hour later, samples of thyroid and blood were obtained.

*Counting techniques.* In experiments in which the acute iodide dose given prior to [$^{35}$S]PTU was not labelled, the thyroid was homogenised in 0.4 ml ice-cold deionized water; total $^{35}$S and the level of unchanged [$^{35}$S]PTU in thyroid and serum were determined using a Tri-carb liquid scintillation counter (Packard) and radiochromatogram scanner equipped with an integrating unit (Panax Ltd), as described previously [5, 6] except as follows: serum samples for liquid scintillation counting were decolourised by addition of 0.2 ml 30% (w/v) hydrogen peroxide. In TLC of serum, 10 μl of each sample was applied to cellulose plates and run in an ethanol/1 M ammonium acetate (75/30 v/v) solvent system.

In experiments in which the acute iodide dose given before [$^{35}$S]PTU contained $^{125}$I or $^{131}$I, the thyroid was homogenised in 1.0 ml of ice-cold deionized water and 0.5 ml was transferred to a gamma counting tube containing 2.0 ml of ice-cold 10% (w/v) trichloroacetic acid (TCA) solution containing $10^{-3}$ M methimazole and $10^{-4}$ M KI. Separation of counts from $^{35}$S and $^{125}$I (or $^{131}$I) in liquid scintillation counting samples of thyroid and serum was achieved by the combined use of a liquid scintillation counter and a gamma counter by an accurate method similar to that described previously in detail [1]. An indication of the degree of accuracy and precision of the $^{35}$S measurements obtained using the double-label procedure is given by comparison of the mean level of serum $^{35}$S (± S.D.) at time of death in 61 NID rats which also received $^{125}$I (14.3 ± 1.85 nmoles $^{35}$S/ml) and in 99 NID rats which received no $^{125}$I (15.1 ± 2.23 nmoles $^{35}$S/ml). Such insignificant differences are certainly no greater than are obtained where comparison is made of two batches of animals where no $^{125}$I was involved. Therefore for the NID animals we feel justified in pooling $^{35}$S measurements obtained from single- and double-label experiments (only relevant for NID rats). Separation of $^{35}$S from $^{125}$I emissions on TLC plates was carried out by using a special $^{125}$I detector head (detects only $^{125}$I γ-emissions) in addition to the normal detector head (detects β-emission from both $^{35}$S and $^{125}$I, the latter with low efficiency) with the radiochromatogram scanner. Each TLC plate had an $^{125}$I standard and was scanned under both detectors. A subtraction procedure was used to obtain net $^{35}$S activity detected by the β-detector. Counts detected by the $^{125}$I γ-detector for each area of the plate were subtracted from the total counts detected by the β-detector for the same area, correcting for the difference in percentage efficiency of detection of the $^{125}$I standard by the two detectors. By careful choice of using as little $^{125}$I per rat as would allow accurate measurement of thyroid and serum $^{125}$I by the autogamma counter, the problem of $^{125}$I interference with $^{35}$S detection on the TLC plate by the B-detector was minimised. In many of the thyroid TLC scans negligible $^{125}$I counts were present. When $^{131}$I and $^{35}$S were used (the first double-label experiment, LID rats), the $^{131}$I activity was allowed to decay to background activity before the plates were scanned for $^{35}$S activity. However, a long delay before scanning for $^{35}$S, in combination with the lower sp. act. of [$^{35}$S]PTU available for use in this experiment, resulted in many of the thyroid TLC strips having insufficient $^{35}$S activity to allow accurate quantitation of components. This accounts for the much lower number of determinations of thyroid unchanged [$^{35}$S]PTU (Table 1) than of thyroid total $^{35}$S (Fig. 1) among LID animals for each acute iodide dosage group. In subsequent double-label experiments $^{125}$I was used in preference to $^{131}$I. The total $^{125}$I (or $^{131}$I) in each thyroid or serum sample was counted in a Packard Autogamma Counter with appropriate standards. Tubes containing samples of thyroid were then centrifuged at 3000 rpm at 0° for 10 min, the supernatant was discarded and the TCA-precipitated–protein-bound iodine (PBI) was washed with 2.5 ml of cold TCA solution (this washing was found to remove any remaining free iodine), resuspended in 4 ml water and recounted.

The known sp. act. of the stable iodide in each acute iodide dose was used to calculate thyroid and serum levels of acutely administered iodide and the quantity of administered acute iodide organified in the thyroid. An approximate thyroid/serum ratio of inorganic radioiodide (T/S*$I^-$) was calculated only for acute [$^{131}$I]iodide doses which almost completely inhibited organification of thyroid [$^{131}$I]iodine (Wolff–Chaikoff effect). This approximate T/S*$I^-$ ratio was considered only as an index of the activity of the thyroid iodide transport system and a convenient parameter for illustrating saturation of $I^-$ trapping at very high acute $I^-$ doses. The calculation was based on two assumptions: (a) all serum radioiodine was considered as iodide, and, (b) all TCA-soluble thyroid radioiodine was considered as iodide, where the organification of iodide was inhibited (Wolff–Chaikoff effect, 1–100 μmoles KI/100 g BW).

*Effect of the administered [$^{35}$S]PTU on the level of newly organified iodine.* An additional experiment was performed to determine whether the quantity of [$^{35}$S]PTU given under the conditions of these studies demonstrated a significant antithyroid effect. Nineteen LID rats (given tracer $^{125}$I in saline or in an acute iodide dose of 0.01–1 μmoles KI/100 g) received saline instead of the [$^{35}$S]PTU given to the LID rats whose thyroid $^{35}$S data was shown in Fig. 1 and Table 1. The thyroid was homogenised in 1 ml of ice-cold TCA solution and transferred to a gamma counting tube containing 1.5 ml of the cold TCA solution. A comparison was made of the percentage of thyroid total radioiodine which was TCA-soluble when [$^{35}$S]PTU was administered.

Statistical analyses were performed by the Student's *t*-test and the non-parametric Mann–Whitney

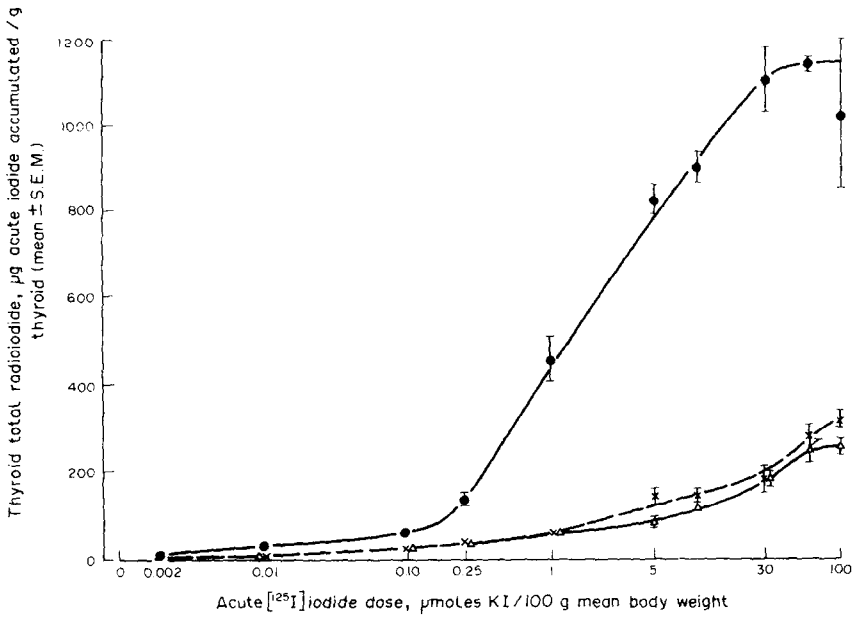


Fig. 2. The effect of chronic iodine intake (●-----●, LID; ×—×, NID △——△, HII) on rat thyroid uptake of acute radioiodide administered 1 hr prior to [35S]PTU. The thyroid 35S data for these animals is shown in Fig. 1 and Table 1. Each point is the mean ± S.E.M. of five or six thyroids. NID and HII rats received [125I]iodide whereas LID rats received [131I]iodide. Error bars not shown are smaller than the size of the point.

*U*-test. The level of statistical significance chosen was $P \leqslant 0.02$ due to the large number of *t*-tests performed and the disparate sizes of groups compared.

## RESULTS

Owing to the large number of rats required, difficulty was encountered in supplying animals. Only data for batches of rats whose mean BW deviated from 250 g by less than 20% are shown, although experiments in which a greater deviation was present gave similar results. Only 63 of the 167 NID rats received $^{125}I$ tracer but as discussed in Materials and Methods pooled $^{35}S$ data for all the NID rats is presented. Fig. 1 and Table 1 show thyroid $^{35}S$ data for LID, NID and HII rats. All the LID ($^{131}I$) and HII ($^{125}I$) rats received radiolabelled acute iodide doses. The thyroid radioiodine data for these LID, NID and HII rats which received radiolabelled acute iodide is shown in Figs 2–4.

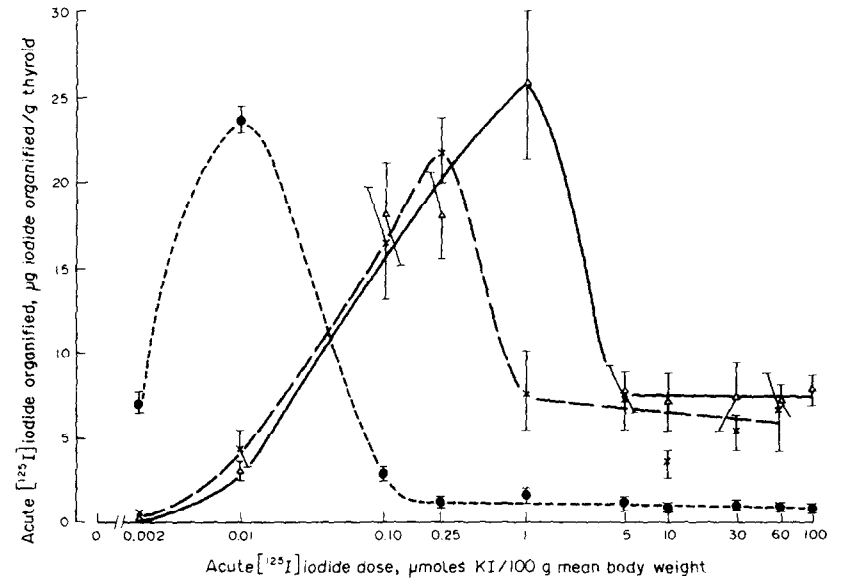


Fig. 3. The effect of chronic iodine intake (●-----●, LID; ×—×, NID; △——△, HII) on rat thyroid organification of acute radioiodide administered 1 hr prior to [35S]PTU (thyroid 35S data in Fig. 1 and Table 1). LID rats actually received [131I]iodide. Each point is the mean ± S.E.M. of five or six thyroids. Error bars not shown are smaller than the size of the point.

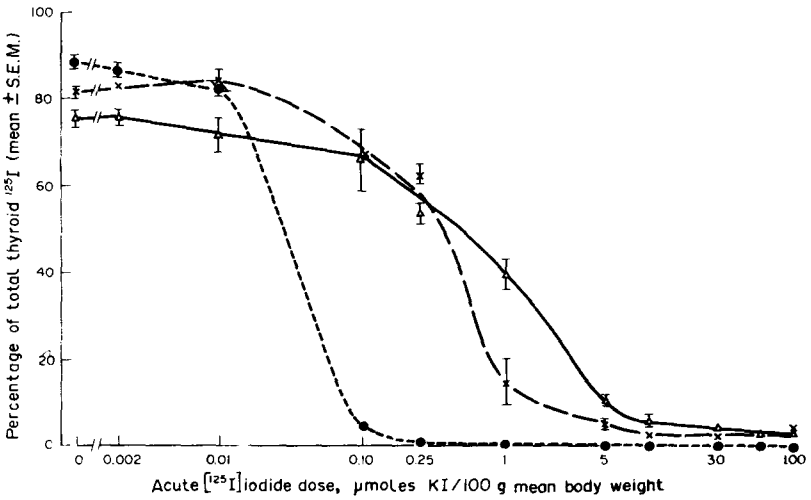


Fig. 4. The effect of chronic iodine intake (●-----●, LID; ×—×, NID; △——△, HII) and acute radioiodide dose (given 1 hr prior to [$^{35}S$]PTU; see Fig. 1 and Table 1 for thyroid $^{35}S$ data) on the percentage of total rat thyroid radioiodine which is organified. Each point is the mean ± S.E.M. of five or six thyroids and error bars not shown are smaller than the size of the point. LID rats actually received acute [$^{131}I$]iodide.

*Thyroid total $^{35}S$ accumulation*

As mentioned earlier only pooled $^{35}S$ data are presented for NID rats. This is the result of three NID experiments but the same doses of acute iodide and [$^{35}S$]PTU were administered in each. It was decided that the NID rat $^{35}S$ data would be best presented as pooled values of all three NID experiments.

Variations in acute and chronic iodine intake among the several groups were associated with thyroid total $^{35}S$ levels that lay between 58.2 ± 3.1 (mean ± S.E.M.) and 261 ± 57 nmoles/g thyroid (Fig. 1).

*Effect of chronic iodine intake.* Irrespective of acute iodide administration, the higher the chronic iodine intake, the higher was the thyroid total $^{35}S$ level. Comparison was by the Mann–Whitney *U*-test, using all of the $^{35}S$ levels from all of the samples for each different chronic iodine intake. Highly significant differences ($P < 0.00006$) in thyroid total $^{35}S$ levels were found between LID, NID and HII rats.

*Effect of acute iodide.* Irrespective of the level of chronic iodine intake, with increased acute KI pretreatment, there was a trend toward increased thyroid accumulaiton of total $^{35}S$ which became statistically significant at the acute KI dose of 0.1 μmoles/100 g BW (overall average of 47% increase from control when LID, NID and HII data taken together, $P < 0.01$). Between 0.1 and 5 μmoles KI/100 g BW, thyroid accumulation of total $^{35}S$ decreased to a significantly lower level ($P < 0.001$) which was unchanged by higher iodide doses up to 100 μmoles KI/100 g. A similar situation was found when LID, NID and HII rats were considered separately (Fig. 1) although the trends observed in HII rats fell short of statistical significance.

*Effect of iodine intake on thyroid metabolism of [$^{35}S$]PTU*

In TLC of thyroid homogenates on cellulose plates in an ethanol/1 M ammonium acetate solvent system (75/30 v/v), four $^{35}S$ peaks were consistently found: protein-bound $^{35}S$ activity, inorganic sulphate, an unknown metabolite X (probably the sulphinic acid of PTU [7, 8]) and unchanged PTU, as previously reported by Marchant *et al.* [1].

Increasing doses of iodide acutely caused no consistent change in thyroid metabolism of [$^{35}S$]PTU, i.e. there was no trend in the relative proportions of the four peaks of $^{35}S$ activity found in TLC of thyroid homogenate (data not shown). Considering chronic iodine intake, however, comparison by the Mann–Whitney U-test of the percentage of thyroid total $^{35}S$ occurring as unchanged [$^{35}S$]PTU has shown that (although not true when comparison is only made of LID, NID and HII control animals not given acute iodide) overall values in LID rats were significantly higher than those in NID or HII rats ($P < 0.008$ and $< 0.0006$ respectively). Values in NID rats were very significantly higher than in HII rats ($P < 0.00006$). The overall mean percentage (± S.E.M.) of thyroid total $^{35}S$ occurring as unchanged [$^{35}S$]PTU for LID, NID and HII rats was 50.4 ± 2.55, 42.1 ± 1.06 and 31.1 ± 1.30% respectively.

Variations in acute and chronic iodine intake yielded thyroid unchanged [$^{35}S$]PTU levels between 25.6 ± 3.4 and 107.9 ± 22.2 nmoles/g thyroid (Table 1). Irrespective of chronic iodine intake, with increasing acute iodide doses, the thyroid level of unchanged [$^{35}S$]PTU increased significantly ($P < 0.001$) the maximum being reached at doses between 0.1 and 1 μmoles KI/100 g (greater than 60% increase from control). Between 1 and 5 μmoles KI/100 g, thyroid unchanged [$^{35}S$]PTU fell significantly to a lower level ($P < 0.005$) which was unchanged by larger doses of iodide up to 30 μmoles KI/100 g. When rats receiving different chronic iodine intakes are considered separately (Table 1) a similar situation is seen. Comparison by the Mann–Whitney *U*-test showed that thyroid unchanged [$^{35}S$]PTU levels for LID rats were significantly lower than for NID

($P < 0.018$) or HII rats ($p < 0.0024$); values in NID rats were not statistically significantly lower than in HII rats.

*Serum $^{35}S$ data (results not detailed in tables or figures)*

Neither total $^{35}S$ levels in serum nor the percentage of total $^{35}S$ occurring as unchanged [$^{35}S$]PTU changed significantly from control values throughout the range of acute iodide dosage in LID, NID or HII rats. Overall mean $^{35}S$ levels (± S.E.M.) for LID, NID and HII rats were 18.5 ± 0.22, 14.8 ± 0.18 and 16.0 ± 0.20 nmoles/ml serum respectively. Corresponding values for the overall mean percentage of serum total $^{35}S$ occurring as unchanged [$^{35}S$]PTU were 81.5 ± 0.60, 72.3 ± 0.84 and 81.5 ± 0.90%.

*Thyroid accumulation of acute radioiodine*

LID rats accumulated a much larger amount of each dose of acutely administered iodine than NID or HII rats. At acute KI doses above 1 μmoles/100 g BW, HII rats appeared to accumulate less iodine than did NID rats. NID and HII rats accumulated progressively more iodine with increasing KI doses throughout the acute KI dose range. LID rats displayed rapidly increasing thyroid accumulation of iodine at doses between 0.1 and 30 μmoles KI/100 g. At the latter dose iodine accumulation appeared to level off, indicating that the thyroid had reached a maximum iodine capacity (Fig. 2). For LID rats the approximate $T/S^*I^-$ values were calculated for iodide doses where the Wolff–Chaikoff effect was operating (1–100 μmoles KI/100 g BW) and are shown with the thyroid $^{35}S$ levels in Fig. 5. As the acute iodide dose was increased from 1 up to 100 μmoles KI/100 g BW the $T/S^*I^-$ decreased rapidly towards unity but the thyroid total $^{35}S$ level was unaffected.

*Thyroid organification of acute radioiodine*

In LID, NID and HII rats, the thyroid level of organified radioiodine peaked at different acute iodide doses, 0.01, 0.25 and 1 μmoles KI/100 g BW respectively. Irrespective of chronic iodine intake, as the acute iodide dose was increased, the thyroid level of organified radioiodine increased until the Wolff–Chaikoff effect occurred, i.e. with higher acute iodide doses, the thyroid level of organified radioiodine fell to a minimal value (Fig. 3). To facilitate comparison, thyroid accumulation of $^{35}S$ and organification of acute [$^{131}I$]iodide in the same LID rats are shown together in Fig. 6. Fig. 4 shows for LID, NID and HII rats how the percentage of total thyroid radioiodine which is protein-bound varies with the acute radioiodide dose (administered 1 hr prior to [$^{35}S$]PTU).

*Effect of the administered [$^{35}S$]PTU on the level of newly organified iodine (data not shown in tables or figures)*

In LID rats given saline in place of [$^{35}S$]PTU, 99.3 ± 0.3% (mean ± S.E.M.) of total thyroid radioiodine was in the organic form when only $^{125}I$ tracer was given. When 0.01 μmoles KI/100 g BW was administered, 94.3 ± 2.3% of thyroid radioiodine was organified. In rats given [$^{35}S$]PTU, the values corresponding were 88.6 ± 1.5 and 82.0 ± 1.5% respectively. The minor, but significant ($P < 0.002$), decrease in the percentage of thyroid radioiodine organified when this low dose of [$^{35}S$]PTU was given, was observed with acute iodide doses up to and including 0.01 μmoles/100 g BW; no significant difference was seen at higher iodide doses.

## DISCUSSION

With variations in both chronic and acute iodine intake, as much as a four-fold change was observed in the level of total $^{35}S$ and unchanged [$^{35}S$]PTU in the thyroid of rats given [$^{35}S$]PTU 1 hr after the acute iodide dose. Generally, the higher the chronic iodine intake, the higher the thyroid accumulation of total $^{35}S$ and unchanged [$^{35}S$]PTU and the higher was the acute iodide dose required to induce the Wolff–Chaikoff effect, but the lower was the percentage of thyroid total $^{35}S$ occurring as unchanged [$^{35}S$]PTU and the lower was the thyroid accumulation of acute radioiodine.

The inter-animal variation in thyroid $^{35}S$ level was found to increase markedly with increasing chronic iodine intake as reflected in the S.E.M. (Fig. 1 and Table 1). This variation was not associated with a difference in thyroid size and was restricted to thyroid $^{35}S$ data since, in the same thyroid glands, radioiodine data showed no large inter-animal variation.

Thyroid metabolism of [$^{35}S$]PTU was affected by chronic iodine intake but appeared independent of

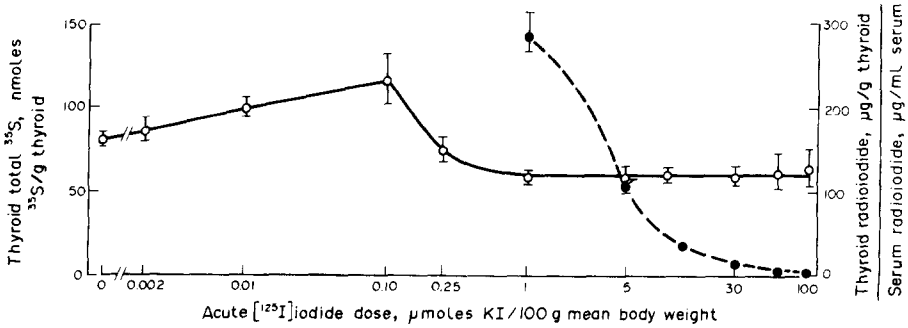


Fig. 5. The effect of a wide range of acute [$^{131}I$]iodide doses given in the same LID rats as in Fig. 4 1 hr prior to [$^{35}S$]PTU on thyroid accumulation of total $^{35}S$ (○——○) and inorganic radioiodide (●-----●, expressed as an approximate thyroid/serum ratio). Each point is the mean ± S.E.M. of five or six animals and error bars not shown are smaller than the size of the point.

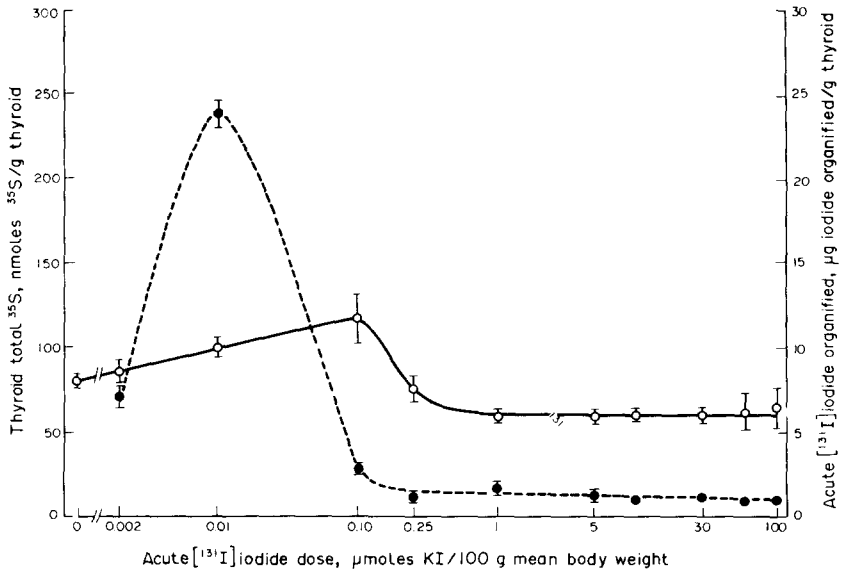


Fig. 6. The effect of a wide range of acute [$^{131}$I]iodide doses administered in the same LID rats as in Fig. 3 1 hr prior to [$^{35}$S]PTU on thyroid accumulation of total [$^{35}$S] (○——○) and organification of the acute [$^{131}$I]iodide (●-----●). Each point is the mean ± S.E.M. for five or six thyroids. Error bars not shown are smaller than the size of the point.

the occurrence of the Wolff–Chaikoff effect and saturation of thyroid iodide transport. What little other data is available is generally consistent with the results of the present study. Thus Marchant *et al.* [1] found in NID rats that 15 μmoles KI/100 g BW given prior to [$^{35}$S]PTU caused a 42% decrease in thyroid total $^{35}$S. Marchant [9] reported that a LID also reduced thyroid total $^{35}$S and that 2.4 μmoles KI/100 g prior to [$^{35}$S]PTU caused no change in the thyroid metabolism of [$^{35}$S]PTU. Using [$^{14}$C]PTU, Lindsay *et al.* [10] in an abstract, reported that thyroid $^{14}$C in rats was decreased by 50% when an LID was fed and accumulation of $^{14}$C increased with increasing acute iodide dose from 0.012–0.1 μmoles KI/100 g BW but decreased progressively with higher iodide doses until essentially no effect was seen with 12.5 μmoles KI/100 g. Nakashima *et al.* [6] administering 11.8 μmoles [$^{35}$S]PTU/kg BW to rats and removing the thyroid 6 hr later found that the thyroid total $^{35}$S level was reduced by 80% when rats were fed an LID alone (0.015–0.018 μg I/g diet for 4–5 months) compared to rats on the same diet but given supplemental KI in their drinking water (0.5 μg I/ml). In the rats with the very low iodine intake, 92% of total $^{35}$S was unchanged drug compared to 23% in the higher iodine intake group. In consequence there was only a 20% decrease seen in the thyroid level of unchanged drug due to low iodine intake. Aungst *et al.* [11] found that 30 μmoles KI/100 g BW administered i.p. to NID rats 1 hr prior to [$^{35}$S]PTU (27.7 μmoles/kg i.p.) had no significant effect on the T/S ratio of [$^{35}$S]PTU. However, three days treatment of 2 × 5 mg KI i.p. prior to [$^{35}$S]PTU increased the T/S ratio of PTU at least three-fold from 10 hr post-dose. The adaptation of the rat thyroid to iodide excess and depletion has been described [12]. In rats chronically exposed to a low iodine diet the iodide trap will accumulate a greater proportion of plasma iodide and in those chronically exposed to a high iodine diet the trap will accumulate a lesser proportion of plasma iodide. These findings were observed in our study. Above a critical intrathyroidal concentration of inorganic iodide, organification of iodide is inhibited—the Wolff–Chaikoff effect. Since the iodide trap activity varies inversely with chronic iodine intake, one would expect that the Wolff–Chaikoff effect would appear at different acute iodide doses for LID, NID and HII rats, as was observed in this study. In LID rats, the dose of [$^{35}$S]PTU administered appeared to have had only a minor inhibitory effect on organification of the administered acute iodide. This appeared true for NID and HII rats also since more than 75% of thyroid radioiodine was organified at low acute iodide doses (Fig. 6). However, this does not necessarily imply that the dose of [$^{35}$S]PTU administered is insufficient to block the organification of iodide in the rat thyroid at any time or to block the thyroid at the more sensitive step of coupling of iodotyrosines. Since the acute iodide was administered 1 hr prior to [$^{35}$S]PTU and the rat killed 1 hr after [$^{35}$S]PTU then two considerations are relevant: (a) much of the radioiodine in the thyroid is likely to have been accumulated and organified prior to significant quantities of [$^{35}$S]PTU entering the thyroid, and (b) it is known that in the rat the maximum thyroid level of [$^{35}$S]PTU is not achieved until at least 8 hr after i.p. administration [1, 2].

Thyroid uptake of [$^{35}$S]PTU appeared independent of the occurrence of the Wolff–Chaikoff effect as is clearly shown in Fig. 6 for the same LID rats.

Fig. 5 shows in the same LID rats that over an increasing acute iodide dose range between 1 and 100 μmoles/100 g BW, the T/S*$I^-$ fell sharply from about 290 to a value approaching unity, indicating rapidly increasing saturation of the iodide-trapping mechanism; nevertheless, thyroid accumulation of $^{35}$S in this iodide dose range was unaffected. A similar

situation was found for NID and HII rats. Therefore thyroid uptake and metabolism of [$^{35}$S]PTU appeared to be independent of the occurrence both of the Wolff–Chaikoff effect and of the saturation of the thyroid iodide transport mechanism.

Further work is required to find whether iodine intake similarly affects $^{35}$S and unchanged [$^{35}$S]PTU accumulation in the human thyroid. If so, the effect may be of clinical importance in treating hyperthyroid individuals whose chronic iodine intake was different and when both PTU and iodide are being administered, e.g. prior to subtotal thyroidectomy, treatment of thyroid crisis or hyperthyroidism [13–16].

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