ENERGY METABOLISM AND FUNCTION OF THYROID GLAND IN VITRO

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It has been demonstrated by several workers that the concentrating ability of surviving thyroid slice of inorganic iodide from surrounding media is intimately related to respiratory activity of tissue slice (Schachner et al., 1943, 1944; Slingerland, 1955; Freinkel and Ingbar, 1955a). Furthermore, it has been established that the organic binding of iodine to tyrosine molecules and the condensation of diiodotyrosine to thyroxine stands in close relation to oxidative reaction in biological as well as in chemical processes (Schachner et al., 1944; Harington and Pitt-Rivers, 1945; Weiss, 1952; Taurog et al., 1955). These results might suggest that the energy for hormone synthesis in thyroid tissue is obtained by means of oxidative metabolism through the agency of high energy phosphate bond. Actually, 2, 4-dinitrophenol, an uncoupling reagent of oxidative phosphorylation, has inhibitory effect upon thyroid function (Weiss, 1953; Slingerland, 1955; Freinkel and Ingbar, 1955b). Oxidative phosphorylation is usually measured employing mitochondria preparations and the activity is expressed by the ratio P/O. Though a work was done on phosphorus metabolism of thyroid slice by Schwarz and Morton (1955), suitable indices of oxidative phosphorylation in a functionally organized system, such as tissue slices, had not been established.

Recently, Katsura (1957) in this laboratory, showed that relative P/O ratio of radioactivity of P32-phosphate incorporated to an acid-soluble organic phosphate fraction to oxygen consumption. Since intact architecture of tissue is necessary for performance of thyroid function in vitro (Schachner et al., 1943; Morton and Chaikoff, 1943), the method of the author just cited above is useful for such an experiment employing tissue slices.

Present experiments were undertaken to study the relation between energy metabolism and the process of hormone biosynthesis in thyroid tissue by means of simultaneous observations of oxygen consumption, oxidative phosphorylation and iodine metabolism employing respective inhibitors for each process, the details of which are rendered in the following account.

MATERIALS AND METHODS

Fresh beef thyroid packed in ice was brought back from a slaughterhouse. Slices of thyroid tissue of about 0.5 mm thickness were prepared with a sharp razor blade.

Dog thyroid slices used in the experiment were prepared from a dog sacrificed by electro-shock.

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Iodine metabolism  Beef thyroid slices of 200-300 mg (in the case of dog, 100 mg) were put into a Warburg flask containing Krebs-Ringer phosphate solution (pH 7.4) as suspending medium and air as gas phase and incubated at 37°C for 2 hours. Oscillation frequency was 100 per minute. In the center well, 0.2 ml of 2N NaOH and filter paper were placed. The Krebs-Ringer solution per flask contains 8 μCi carrier-free radioactive iodide (NaI) and total contents of each flask amounted to 3.0 ml which contains inhibitors solution or its solvent as control. The mixtures were placed into the main chamber at the beginning of experiments.* After preincubation for temperature equilibration for 10 minutes, oxygen consumption was determined at 30 minutes intervals. After the end of incubation, the slices were taken out of the flask, and radioactive iodide attached to the surface of them was carefully washed away with about 30 ml of isotonic saline. After washing 3 times, the radioactivity of the washing fluid became negligible. After the removal of the slices, an aliquot of the medium was taken out of each flask, diluted to ten volumes with water; 1 ml of it was placed in a steel cup (2.5 cm in diameter and 0.6 cm in depth) and after alkalization with drops of 10 per cent NaOH, was desiccated under the infra red lamp. Differences in radioactivity between media with and without slices were designated as 1131 uptake.

The measurement of radioactivity of each sample was carried out with a counter (Kobe Kogyo Co., Ltd.) provided with the mica end-window Geiger-Mueller tube.

The washed slices were put in a whole glass homogenizer with 1 ml of 0.1 M carbonate buffer pH 9.0 and was kept in a water bath at 100°C for about 5 minutes. After cooling in the room temperature, 1 ml of carbonate buffer at pH 9.0 containing trypsin (Merck) and pancreatin (each of 2 per cent) was added and slices were homogenized. Then homogenate was subjected to digestion in a water bath at 37°C for 48 hours after addition of a drop of toluene and digestion was continued for 24 hours more with addition of 1 ml of the proteolytic enzyme solution. After completion of digestion, inorganic iodide, moniodotyrosine, diiodotyrosine and thyroxine were fractionated from the hydrolysate after the method of Morton and Chaikoff (1943). Inorganic iodide was extracted as iodine with CCl4 and washed away repeatedly to prevent probable contamination of inorganic iodide to another fraction. n-Butanol was used to extract thyroxine and diiodotyrosine fraction. Thyroxine was extracted from alkalized hydrolysate (20 per cent of NaOH) and diiodotyrosine at pH 3.5-4.0 respectively. So-called diiodotyrosine fraction contains almost all parts of organic iodine compounds of the slice and consisted of both mono- and diiodotyrosine. The amount of each collected fraction was 5 ml and 1 ml of it was put in a cup, desiccated after alkalization and β counting was carried out.

In some trials, radiochromatography was performed on a filter paper (Tokyo Roshi No. 50, 2 x 40 cm) by ascending method, employing n-butanol, acetic acid and H2O (78 : 5 : 17) as solvent, and radioinactive iodide (KI), diiodotyrosine and thyroxine as carriers. After development and drying, paper was covered with 1 cm thick lead plate with a slit (2 x 1 cm) placed under the Geiger-Mueller tube and radioactivity was measured as c. p. m. for each 1 cm length. Then color was developed to confirm the positions of diiodotyrosine and thyroxine by spraying ninhydrin and inorganic iodide by 0.1 M CuSO4.

Phosphorus metabolism  Phosphorus metabolism in the thyroid slice was determined after Katsura (1957). All experiments were performed in similar conditions as in the experiments above described except that the phosphate concentration in the Krebs-Ringer phosphate solution was reduced to half to attain elevated incorporation of radiophosphate. Ten to forty μCi of P32 (Na2HPO4) was employed per flask.

The method of determination of radioactivity of acid-soluble organic phosphate fraction of the thyroid slices is outlined as follows: Well washed thyroid slices were homogenized and

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* When radioactive iodine solution was added from the side arm after completion of preincubation, measured data of radioactivity differed from flask to flask.
extracted with 8 per cent trichloroacetic acid and such procedure was repeated twice. After addition of sulfuric acid, 2 per cent ammonium molybdate was added, and inorganic phosphate was changed to phosphomolybdate, which was extracted with iso-butanol. All these procedures were carried out under ice-cold condition. Radioactivity remained after such extraction was measured as acid soluble organic phosphate.

Experiments were made from January to April 1956.

RESULTS

Effects of potassium cyanide

When KCN is added to the thyroid slice, oxygen consumption decreases according to the amount of KCN; phosphorus metabolism and hormone biosynthesis change as shown in Tables 1 and 2. Data listed in these tables represent actually observed values. Figure 1 shows the relations between KCN concentration and iodine and phosphorus metabolisms; numerals attached to ordinate represent per centage as compared with the control experiment. "Iodination" was used in the sense of function of thyroid slices as shown by disappearance of $I^{131}$ from the surrounding medium ($I^{131}$ uptake), inorganic iodide, diiodotyrosine and thyroxine fractions formed in thyroid slices. As shown in Fig. 1 A, with the decrease in oxygen consumption, iodide uptake and subsequent organic binding of iodine decrease. It is of interest to find that the amount of inorganic iodide in the beef thyroid slice is elevated above the level of control at $1 \times 10^{-4}$ M concentration of KCN. Also with dog thyroid slices, the addition of KCN in final concentration of $10^{-5}$-$10^{-4}$ M tends to increase the amount of inorganic iodide fraction.

Table 1. Effect of cyanide on phosphorus metabolism of beef thyroid slices

<table>
<thead>
<tr>
<th>Conc. of KCN</th>
<th>Weight* of slices (mg)</th>
<th>O$_2$ uptake (pl)</th>
<th>Radioactivity (c. p. m.)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>* Initial wet weight per flask</td>
<td>** Per cent change of control value calculated from average values</td>
<td>† Total acid-soluble phosphate fraction</td>
</tr>
<tr>
<td>0</td>
<td>{300} 140</td>
<td>5,535 880 335 340</td>
<td></td>
</tr>
<tr>
<td>1 x 10$^{-3}$ M</td>
<td>{# 271 22%**} 75</td>
<td>3,455 631 271</td>
<td></td>
</tr>
<tr>
<td></td>
<td># 271 22%**</td>
<td>6,078 902 36 36</td>
<td></td>
</tr>
<tr>
<td></td>
<td># 5,072 998 84 84</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>{300} 159</td>
<td>10,262 1,763 646</td>
<td></td>
</tr>
<tr>
<td>1 x 10$^{-4}$ M</td>
<td>{# 145 93%} 152</td>
<td>11,979 2,241 630</td>
<td></td>
</tr>
<tr>
<td></td>
<td># 10,637 1,723 715 715</td>
<td>10,710 1,589 592 592</td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>{200} 107</td>
<td>1,721 221 101 101</td>
<td></td>
</tr>
<tr>
<td>5 x 10$^{-5}$ M</td>
<td>{# 91 91%} 106</td>
<td>1,671 257 95 95</td>
<td></td>
</tr>
<tr>
<td></td>
<td># 1,371 151 84 84</td>
<td>1,545 105 88 88</td>
<td></td>
</tr>
</tbody>
</table>

Table 1. Effect of cyanide on phosphorus metabolism of beef thyroid slices
Table 2. Effect of cyanide on iodine metabolism of beef thyroid slices

<table>
<thead>
<tr>
<th>Conc. of KCN</th>
<th>Weight* of slices (mg)</th>
<th>$O_2$ uptake (μl)</th>
<th>Radioactivity (c. p. m.)</th>
<th>Fractions in thyroid slice</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>$^{131}$ I uptake</td>
<td>Inorg. I</td>
</tr>
<tr>
<td>0</td>
<td>1300</td>
<td>151</td>
<td>10,250</td>
<td>4,427</td>
</tr>
<tr>
<td></td>
<td>153</td>
<td></td>
<td>9,132</td>
<td>3,360</td>
</tr>
<tr>
<td>$1 \times 10^{-3}$ M</td>
<td>31% 21%**</td>
<td>33%</td>
<td>4,334</td>
<td>$^{38}$%</td>
</tr>
<tr>
<td>0</td>
<td>300</td>
<td>157</td>
<td>16,362</td>
<td>10,758</td>
</tr>
<tr>
<td></td>
<td>150</td>
<td></td>
<td>14,781</td>
<td>13,155</td>
</tr>
<tr>
<td>$1 \times 10^{-4}$ M</td>
<td>143 90%</td>
<td>134</td>
<td>16,379</td>
<td>$^{104}$%</td>
</tr>
<tr>
<td></td>
<td>134</td>
<td></td>
<td>16,929</td>
<td>18,656</td>
</tr>
<tr>
<td>5 $\times 10^{-5}$ M</td>
<td>110 100%</td>
<td>115</td>
<td>2,703</td>
<td>$^{79}$%</td>
</tr>
</tbody>
</table>

* Initial wet weight per flask
** Per cent change of control value calculated from average values
† "Diiodotyrosine fraction"
‡ "Thyroxine fraction"

Fig. 1. Effect of cyanide on "iodination" (A), "phosphorylation" (B) and oxygen consumption of beef thyroid slices. In A, $I^-$: inorganic iodide fraction, DIT: diiodotyrosine fraction, Tx: thyroxine fraction. In B, organic P: acid soluble organic phosphate fraction.

On the other hand, acid soluble organic phosphate fraction of the slices decreases in accordance with the decrease in oxygen consumption (Fig. 1B). These
data show that the blocking of oxygen consumption results in a similar inhibition of formation of acid soluble organic phosphates, of iodide collection and synthesis of organic iodine compounds. Figure 2 represents the radiochromatograms of hydrolysate of thyroid slices to which KCN was added at concentration of $10^{-3}$ M and control. Compared with control curve, the effect of KCN is evident. After treatment with ninhydrin and CuSO$_4$, spots of inorganic iodide, diiodotyrosine, and

![Image of radiochromatogram](image)

**Fig. 2.** Radiochromatogram of hydrolyzed beef thyroid slices. Effect of KCN ($1 \times 10^{-3}$ M). $\Gamma^-$: inorganic iodide, MIT: monoiodotyrosine, DIT: diiodotyrosine, Tx: thyroxine.

![Image of graph](image)

**Fig. 3.** Effect of 2,4-dinitrophenol on "iodination" (A), "phosphorylation" (B) and oxygen consumption of beef thyroid slices. Symbols are same as in Fig. 1.
thyroxine are revealed in the mentioned order beginning from the origin, but radioactivity at thyroxide spot is actually negligible, indicating that thyroxine synthesis does not almost occur in in vitro conditions. Consequently, the thyroxin fraction obtained by extraction is not so significant. It is also found that the portion of monoiodotyrosine is more abundant than that of diiodotyrosine. At the solvent front on the paper, a radioactive spot is found. Since it is identified with the spot revealed by Sudan black lipid staining, it is assumed to be lipid iodinated with radioactive iodine.

Effects of 2, 4-dinitrophenol

As shown in the results described above, hormone biosynthesis declines with the decrease in oxygen consumption accompanied with inhibition of formation of acid-soluble organic phosphates. On the basis of such data, it is of interest to test the effect of 2, 4-dinitrophenol (DNP), an uncoupling reagent of oxidative phosphorylation. Figure 3 A and B represent relations between concentrations of added DNP and iodine (A) and phosphorus metabolism (B) of beef thyroid slice. The addition of $1 \times 10^{-5}$ M DNP increases oxygen consumption about 30 per cent. Despite such stimulation of oxygen consumption, both $I^{131}$ uptake and organic binding of radioactive iodine decrease. Only thyroxine fraction shows an insignificant increase, the ratio of radioactivity (c. p. m.) of thyroxine fraction to the whole being 2 per cent in the control, and 3 per cent in the DNP-added slice. At $5 \times 10^{-5}$ M and $1 \times 10^{-4}$ M, DNP decreases both oxygen consumption and each of iodine fractions. On the other hand, radioactivity of acid-soluble organic phosphate fraction decreases in accordance with the increase of DNP concentration (Fig. 3 B). It is of interest to note that, as shown in Fig. 3 A, the decrease in organic binding and uptake of $I^{131}$ are of the similar grade. This fact seems to indicate that DNP blocks the process of iodine and uptake organic binding is affected secondarily. Figure 4 represents radiochromatograms showing changes in iodine fractions caused by addition of DNP in $5 \times 10^{-5}$ M. Peaks corresponding the spots

![Graph](image_url)
of diiodotyrosine and thyroxine are not revealed, and that of monoiodotyrosine is remarkably low.

Effects of methylthiouracil

In the next trial, methylthiouracil (MTU), a thiourea derivative which is recognized to block the process of organic binding of iodine in thyroid tissue, is tested. It is shown in Fig. 5 A that MTU exerts no measurable effect on oxygen consumption but completely blocks organic binding (at $1 \times 10^{-4}$-10^{-3} M). Figure 6 re-

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Fig. 5. Effect of methylthiouracil on "iodination" (A), "phosphorylation" (B) and oxygen consumption of beef thyroid slices. All symbols are same as in Fig. 1.

Fig. 6. Radiochromatogram of hydrolyzed beef thyroid slices. Effect of methylthiouracil ($1 \times 10^{-4}$ M). Symbols are same as in Fig. 2.
presents a radiochromatogram in the case of addition of MTU in concentration of $1 \times 10^{-4}$ M. Complete blockade of organic binding by MTU is confirmed in this figure. On the other hand, radioactivity of acid-soluble organic phosphate is affected as follows. It rather tends to be increased by MTU at $1 \times 10^{-4}$ M, in which concentration complete blockade of organic binding of iodine occurs. In contrast, with concentration of $10^{-3}$ M MTU the organic phosphate formation is inhibited in some measure. It is therefore known to have no connection with oxidative phosphorylation.

Effects of substrates

In the experiments mentioned above, no substrate is added to the tissue slices. For comparison of the metabolic activity and function of the thyroid tissue, effects of some substrates are tested in the following experiments. In this trial, dog thyroid slices are employed. Figure 7 shows iodine fractions in the case of addition of glucose, sodium succinate and sodium glutamate respectively. The final concentrations of added substrates are 100 mg/dl for glucose and 0.01 M for the two others.

In this figure, the abscissa indicates oxygen consumption and the ordinate
each iodine fraction in percentage of the control values. For the sake of simplicity, the former is named as "oxidation", and the latter "iodination". If iodination is proportional to oxidation, calculated value will be plotted on an inclined straight line connecting 0 and 100 per cent. Comparing with per cent oxidation the value will be plotted either above or below this straight line. Therefore, such figure is able to show the oxidative iodination, in the sense as oxidative phosphorylation.

It is shown that neither glucose nor glutamate produces change in oxygen consumption, while succinate causes about 80 per cent increase. Iodine fraction, however, is lower than the control values in all cases. Especially in the case of succinate, iodine fraction decreases remarkably despite the significant increase of oxygen consumption resulting in marked decrease of oxidative iodination.

In such a sense, succinate is considered to be an uncoupling agent of oxidative iodination. The thyroid slices seems to be in lower state of physiological activity after addition of exogenous substrates employed in this trial.

Effects of substrates on acid-soluble organic phosphate are shown in Fig. 8. In this figure, the abscissa indicates the "oxidation" as in Fig. 7 and the ordinate
shows the percentage of radioactivity of acid-soluble organic phosphates fraction to the control, which is designated as "phosphorylation". The figure is considered to be a suitable manner to show oxidative phosphorylation. As presented in Fig. 8, neither glutamate nor succinate causes change in phosphorylation. By addition of glucose, phosphorylation is markedly increased. It is shown clearly that, though phosphorylation is not blocked, iodination decreases slightly. In contrast to the case of glucose, succinate causes no measurable change in phosphorylation but marked decrease in oxidative phosphorylation on account of increased oxidation uncoupled with phosphorylation.

**DISCUSSION**

Effects of inhibitors upon oxidation, phosphorylation, iodination as well as oxidative phosphorylation and oxidative iodination are summarized in Figs. 9–11. Representation employed in these figures is the same as in Figs. 7 and 8.

Figure 9 shows marked inhibitory effect of KCN in a high concentration ($10^{-3}$ M) upon oxidation, phosphorylation and iodination. However, at all concentrations employed, neither oxidative phosphorylation nor oxidative iodination differ so much from those of control. These facts appear to show that oxidation, phosphorylation and iodination are affected to similar degrees by addition of KCN. Such a seemingly same extent of inhibition appears to show that KCN acts initially upon oxidative process, and phosphorylation and iodination are subsequently affected. There, results of this experiment show a close relation between energy metabolism and hormone biosynthesis in thyroid tissue. Freinkel and Ingbar (1955a) and Weiss (1955) have shown that the collection of inorganic iodide and also organic binding of iodine in thyroid tissue are markedly depressed under anaerobic condition. It can be assumed that the inhibition of collection of iodide is the primary action of KCN and the organic binding is inhibited secondarily.

Figure 10 shows that 2,4-dinitrophenol (DNP) in its higher concentrations ($5 \times$
Fig. 10. Effect of 2,4-dinitrophenol on "oxidative iodination" (A) and "oxidative phosphorylation" (B) of beef thyroid slices. Symbols are the same as in Figs. 7 and 8.

Fig. 11. Effect of methylthiouracil on "oxidative iodination" (A) and "oxidative phosphorylation" (B) of beef thyroid slices. Symbols are the same as in Figs. 7 and 8.

$10^{-8} - 1 \times 10^{-4}$ M) acts as KCN. While in lower concentration ($1 \times 10^{-8}$ M), oxidation is considerably augmented, and phosphorylation and iodination, on the contrary are considerably inhibited. Thus, oxidative phosphorylation and oxidative iodination are depressed in similar grade. Such effect seems to demonstrate that DNP has a dissociating effect on coupling mechanism of phosphorylation and iodination with oxidation, and that a more intimate relationship exists between phosphorylation and iodination that between oxidation in the process of hormone biosynthesis. Freinkel and Ingbar (1955b) have shown that active transport of iodide is affected in inhibitory way by DNP its derivatives, with the use of sheep thyroid slice in medium containing 2-mercapto-imidazole, a potent inhibitor of organic binding of iodine. They showed that the collection of iodide is inhibited
by DNP in rather lower concentration despite the stimulation of oxidation, which fact agrees with the results of the present trial. On the basis of these results, it is most plausible that phosphorylative process in thyroid tissue is more closely related with that of inorganic iodide collection rather than with organic binding of iodine. Weiss (1953) suggested that the inhibitory effect of DNP upon organic binding of iodine might be competitive with organic binding of iodine into tyrosine. Following his suggestion, a spot of iodinated DNP could be found on radiochromatogram. However, Fig. 4 demonstrates that radiochromatogram of hydrolysate of thyroid slice added with DNP has no radioactive spot other than those of inorganic iodide and monoiodotyrosine. Therefore, competitive inhibition of DNP does not seem probable.

Figure 11 shows effect of methylthiouracil (MTU). MTU shows no effect upon oxidation, in concentrations employed here (10^{-4} to 10^{-3} M). However, organic binding of iodine is completely blocked by MTU even in lower concentration. Thus, oxidative iodination shows extremely low value, which appears to indicate that no correlation exists between oxidative process and hormone biosynthesis in this case. On the other hand, phosphorylation is slightly inhibited in higher concentration, and inorganic iodide fraction also decreases in similar degree at the same concentration of MTU. This fact is considered to show another evidence of the correlation of phosphorylation with the process of collection of iodide. In the lower concentration of MTU, phosphorylation is seemingly increased notwithstanding complete inhibition of organic binding of iodine. Therefore, it may be clear that the mechanism of blocking action of MTU on organic binding of iodide has nothing to do with phosphorylative processes, and is due to another mode of affection.

Concerning the effect of some enzyme substrates, concentrations of succinate employed in the present study are far higher than in physiological condition. Each substrate affects oxidative iodination in inhibitory way, especially succinate causes profound depression. On the other hand, glutamate has no effect on oxidative phosphorylation, while succinate causes uncoupling effect, the degree of which is comparable with the case of oxidative iodination. By addition of glucose, in rather physiological concentration, “oxidative phosphorylation” is increased. One of possible assumption for this mechanism is that glucose and its decomposed products, such as triose, may act as acceptor of newly incorporated radioactive phosphate. Mechanism of rather depressant effects of substrates on oxidative iodination is still not clear. Probably it may be correlated with the disturbance of active transport of iodide through cell membrane, caused by unphysiological conditions.

On the basis of results and considerations of the present study, the author is of opinion that “iodination” of thyroid tissue, one of the most important function of the gland, is closely related to high energy phosphate bonds generated from oxidative processes.

**SUMMARY**

To obtain the information concerning the relation between the hormone synthetic function and metabolisms of thyroid gland, effects of some metabolic in-
hibitors upon oxidative, iodine and phosphorus metabolism of surviving beef and
dog thyroid slices were studied with the uses of radioactive iodide (I\textsuperscript{131}) and
orthophosphate (P\textsuperscript{32}). The main results obtained are as follows:

1. Potassium cyanide (1×10\textsuperscript{-3}M) shows remarkable inhibiting effect, in the same
grade, upon oxygen consumption (“oxidation”), I\textsuperscript{131} uptake, iodine metabolism
(“iodination”) and incorporation of radioactive phosphate into acid-soluble organic
phosphate fraction (“phosphorylation”) of beef thyroid slice.

2. 2, 4-Dinitrophenol (DNP) in the concentrations of 1×10\textsuperscript{-5} M, augments
“oxidation”, while it inhibits “iodination” and “phosphorylation” of beef thyroid
slice to a similar extent, showing an intimate relation between phosphorylation
and iodination. In higher concentrations (5×10\textsuperscript{-5} to 1×10\textsuperscript{-4} M) DNP decreases
“oxidation” as well as “iodination” and “phosphorylation”.

3. Methylthiouracil (MTU) in concentrations of 1×10\textsuperscript{-4} to 1×10\textsuperscript{-3} M, has
no effect on “oxidation” while it shows completely blocking effect on organic
binding of iodine. However, “phosphorylation” is slightly inhibited by MTU
(1×10\textsuperscript{-4} M) and is inconsiderably increased in lower concentration (1×10\textsuperscript{-4} M).
In accordance with decrease of phosphorylation, iodination is also inhibited by
MTU (1×10\textsuperscript{-3} M).

4. Glucose (100 mg/dl), succinate (0.01M) and glutamate (0.01 M) decrease
“iodination” of dog thyroid slice despite increase of “oxidation” in the case of succinate.
“Phosphorylation” of beef thyroid slice is not affected by the additions
of glutamate and succinate. Glucose exerts an increasing effect of “phosphory-
lation”.

5. The significance of the present results on the relation between energy
metabolism and function of thyroid gland was discussed.

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REFERENCES