INCREASED CELL MEMBRANE PERMEABILITY TO Na^+ AND K^+ INDUCED BY THYROID HORMONE IN RAT SKELETAL MUSCLE

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Thyroid hormone(T_3) increased Na^+ dependent respiration accompanied by an increase in NaK-ATPase activity. Administration of T_3 increased intracellular K^+ concentration and Na/K ratio in thyroidectomized rats, and the Na^+ efflux rate constant incubated in oxygenized Na^+,K^+-Ringers in euthyroid rats. However, the magnitude of the changes in intracellular K^+ concentration was modest or invisible in comparison to the changes in $QO_2(t)$ and NaK-ATPase activity. The Na^+ and K^+ efflux rate constants in K^+-free +ouabain Ringers were increased by T_3 in both thyroidectomized and euthyroid rats. Thus, thyroid hormone stimulates not only Na pump but also the permeability of cell membrane to Na^+ and K^+. The both effects might contribute to the thyroid thermogenesis.

Evidence has been presented that increased energy expenditure for transmembrane active Na^+ transport mediates a significant fraction of the thermogenic response to thyroid hormone(1-4). The proposal that Na^+ transport is a significant metabolic pacemaker in thyroid thermogenesis is based in part on the inference that mitochondrial oxidation remains coupled to phosphorylation in various thyroid states. In earlier studies, thyroid hormones increased Na^+ dependent respiration accompanied by an increase in NaK-ATPase activity in rat liver, kidney, heart and skeletal muscle(1-4). In this report, we de-

scribe the effects of thyroid status on Na^+ transport dependent QO_2, NaK-ATPase activity, and the cell membrane permeability to Na^+ and K^+ in rat skeletal muscle, and discuss their possible contributions to thyroid thermogenesis.

Na^+-dependent respiration and NaK-ATPase activity

Since skeletal muscle is the most abundant tissue in mammals, the thermogenic response of skeletal muscle is one of the important determinants of the respiratory response of the whole animal to thyroid hormone. The available evidence suggests that modulations in active Na^+ transport also contribute significantly to thyroid thermogenesis in this tissue(1,2). In T_3^*-treated, thyroidectomized rats the increase in $QO_2(t)^*$ accounted for 47% of the increase in QO_2 of diaphragm, and in euthyrid rats it accounted for 84% of this increase(1). Administration of T_3 to thyroidectomized and euthyroid rats decreased intracellular Na^+ and increased intracellular K^+ concentrations without a discernible effect on serum Na^+,K^+, or Cl^- concentrations *in vivo*(4). Further studies revealed that thyroidectomy lowered specific NaK-ATPase activity of the microsomal fraction 32%; injections of T_3 in-

TABLE 1. $Na^+$and K^+ contents of diaphragm from thyroidectomized rats ($\pm T_3$). mEq/L (mean ± s.e.m.) N = 8

Incubation		intracellular concentrations			
		$-T_3$	$+T_3$	Δ	p
Na^+,K^+-Ringers	Na	54 ± 4	53 ± 5	-1	n.s.
	K	143 ± 6	156 ± 5	+13	<0.01
$-K^+$ +ouabain Ringers	Na	151 ± 8	173 ± 7	+22	<0.005
	K	27 ± 3	14 ± 1	-13	<0.005

* Abbreviations: T_3, L-3,5,3'-triiodothyronine; $QO_2(t)$, Na^+-transport-dependent oxygen consumption.

TABLE 2. Na^+ and K^+ contents of diaphragm from euthyroid rats ($\pm T_3$). mEq/L (mean ± s.e.m.) N = 7 in each group. (from ref.1).

Incubation		intracellular concentrations			
		$-T_3$	$+T_3$	Δ	p
Na^+,K^+-Ringers	Na	63 ± 3	66 ± 4	+3	n.s.
	K	136 ± 5	136 ± 4	0	n.s.
$-K^+$ +ouabain Ringers	Na	153 ± 4	157 ± 5	+4	n.s.
	K	15 ± 1	11 ± 2	-4	<0.05

creased this activity 75% in initially hypothyroid rats and 26% in initially euthyroid rats(1). All of these results imply the inference of thyroid hormone stimulation of Na^+ pump.

Intracellular concentrations of Na^+ and K^+: in vitro study

Euthyroid or thyroidectomized rats received three injections of T_3(50μg/100g b.w.) or diluent on alternate days, whose schedule was the same as that of the earlier experiments. The muscle segments of the diaphragms were rapidly dissected and half segments were transfered into the flaskes including either Na^+,K^+-Ringers of K^+-free + ouabain Ringers with ^{14}C-inulin. After one hour incubation of tissues in a Warburg respirometer, the segments were analysed for the determination of intracellular concentrations of Na^+ and K^+. In thyroidectomized rats, administration of T_3 increased intracellular K^+ concentration and decreased Na/K ratio in this tissue with the incubation in Na^+,K^+-Ringers (Table 1). T_3 treatments, however, decreased intracellular K^+ and increased Na^+ concentration in K^+-free +ouabain Ringers. The former result exibits the existence of thyroid stimulation of active Na^+ transport and the latter the existence of thy-

roid hormone induced increase of cell membrane permeability to Na^+ and K^+. In euthyroid rats, however, only intracellular K^+ concentration incubated in K^+-free+ouabain Ringers was significantly decreased by T_3 (Table 2).

Na^+ and K^+ efflux rate constant

In the euthyroid diaphragm, the stimulation of active Na^+ pump by T_3 was not observed by the measurements of Na^+ and K^+ concentrations in vitro. Then, the efflux of ^{22}Na and ^{42}K from the I.C.F.* was measured in various thyroid states to detect the movement of Na^+ and K^+ across the cell membrane by the method of Kleinzeller et al (5). The Na^+ efflux rate constant in diaphragm incubated in Na^+,K^+-Ringers was significantly increased by T_3 in eu-

* Abbreviation : I.C.F., intracellular fluid.

TABLE 3. Effect of T_3 on ^{22}Na efflux rate constants in rat diaphragm incubated in Na^+,K^+-Ringers(unpublished observations). Mean ± s.e.m. k = rate constant.

Pairs of rats	status	k (min^{-1})	Δ	p
8	Euthyroid	0.128 ± 0.005	0.041	<0.005
	Euthyroid +T_3	0.169 ± 0.007		

TABLE 4. Effect of T_3 on ^{22}Na and ^{42}K efflux rate constants in rat diaphragm incubated in K^+-free +ouabain Ringers(unpublished observations). Mean ± s.e.m.

Pairs of rats	status	k (min^{-1}) ^{22}Na	k (min^{-1}) ^{42}K	Δ	p
7	Euthyroid	0.076 ± 0.003		0.011	<0.05
	Euthyroid +T_3	0.087 ± 0.004			
6	Euthyroid		0.052 ± 0.003	0.013	<0.05
	Euthyroid +T_3		0.065 ± 0.004		
6	Hypothyroid		0.044 ± 0.003	0.015	<0.01
	Hypothyroid +T_3		0.059 ± 0.003		

thyroid rats(Table 3). The result reveals that T_3 stimulated the active Na^+transport in diaphragm of euthyroid rats in vitro. The $Na^+$and K^+efflux rate constants were increased by T_3 in euthyroid rats incubated in K^+-free + ouabain Ringers, and the K^+efflux rate constant in hypothyroid rats(Table 4). These findings show that T_3 increased the cell membrane permeability to Na^+ and K^+.

Conclusion

Information is now available on the thyroid-dependent changes in rat skeletal muscle $QO_2(t)$, NaK-ATPase activity, $Na^+$and K^+ contents in various medium, and the Na^+, K^+efflux rate constants. These results revealed that T_3 stimulates not only Na pump but also permeability of cell membrane to Na^+ and K^+. Both effects might contribute to the thyroid thermogenesis. (Supported in part by a Grant from the Japanese Ministry of Education, Science and Culture [No. S49-977024].)

References

1. Asano, Y., U. A. Liberman, and I. S. Edelman: Thyroid thermogenesis: relationships between Na^+-dependent respiration and Na^++K^+-adenosine triphosphatase activity in rat skeletal muscle, J. Clin. Invest. 57(1976), 368-379.
2. Ismail-Beigi, F., and I. S. Edelman: Mechanism of thyroid calorigenesis: role of active sodium transport, Proc. Natl. Acad. Sci. U. S. A. 67(1970), 1071-1078.
3. Ismail-Beigi, F., and I. S. Edelman: The mechanism of the calorigenic action of thyroid hormone: stimulation of Na^++K^+-activated adenosine triphosphatase activity, J. Gen. Physiol. 57(1971), 710-722.
4. Ismail-Beigi, F., and I. S. Edelman: Effects of thyroid status on electrolyte distribution in rat tissues, Am. J. Physiol. 225(1973), 1172-1177.
5. Kleinzeller, A., K. Janáček, and A. Knotková: A simple method for measuring steady-state ion compartments and rate constants of ion fluxes in tissue slices, Biochim. Biophys. Acta. 59(1962), 239-241.

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