0013-7227/89/1245-2443$02.00/0
Endocrinology

Vol. 124, No. 5
Printed in U.S.A.

Carrier-Mediated [^{125}I]-T_3 Uptake by Mouse Thymocytes*

MARCO CENTANNI, GIANCARLO MANCINI, AND MARIO ANDREOLI

Second Chair of Endocrinology, Department of Experimental Medicine, University La Sapienza (M.C., M.A.), and the Department of Experimental Medicine and Biochemical Sciences, University Tor Vergata (G.M.), Rome, Italy

ABSTRACT. Thyroid hormone entry into the thymocyte, a thyroid hormone target, was investigated by incubating the cells with tracer amounts of [^{125}I]L-T_3. At 37 C T_3 uptake was linear with time up to 2 min, and then approached a plateau. The specific T_3 uptake, obtained by subtracting the uptake in the presence of excess unlabeled T_3, represented 48 ± 6% of the total at equilibrium. Unlabeled L-T_4, D-T_3, and triiodothyroacetic acid were less effective than L-T_3 in reducing [^{125}I]T_3 uptake. Kinetic studies on the initial rate of T_3 uptake indicated, for the saturable process, a maximum velocity of approximately 1 pmol/10^6 cells·min and a K_m of approximately 0.8 nM. Lowering incubation temperature to 4 C resulted in a two thirds reduction of the total T_3 uptake. Washout experiments indicated a different hormone release, being more rapid for cells incubated at 4 C than at 37 C; at 30 min 70% of labeled T_3 was released when incubation was carried out at 4 C compared to only 35% after incubation at 37 C, indicating the major intracellular location of the hormone at the latter temperature. An energy requirement of T_3 uptake in thymocytes was shown by sensitivity to oligomycin; the effect was dose dependent, showing a maximal decrease in specific uptake of 85%. The involvement of cation movement in the entry process of T_3 was indicated by the sensitivity to ouabain. These results indicate the existence of a stereospecific, energy-dependent, saturable process for T_3 entry in thymocytes. (*Endocrinology* **124:** 2443–2448, 1989)

SEVERAL studies suggested that T_3 entry in target tissue is a carrier-mediated process, either active or facilitated (1–12). Some reports also indicated the involvement of sodium in T_3 entry into cells (1, 4, 13). More contradictory results have been obtained for cellular T_4 uptake (4, 6, 14). The mode of entry into cells may play a role in determining the ultimate intracellular bioavailability of the most powerful thyroid hormone, T_3. This T_3 availability comes from the intracellular 5′-deiodination of T_4 and from the circulating T_3 pool; the relative contribution of the two sources strongly depends on the tissue (15–17). Therefore, it is conceivable that in those tissues in which the contribution to intracellular T_3 by T_4 biotransformation is minor or negligible, the plasma membrane is an important regulator of T_3 entry and metabolic availability. Thymocytes represent such a tissue; they are target cells for thyroid hormones in which more than 80% of intracellular T_3 derives from circulating T_3 (15). In these cells plasma membrane high affinity binding sites for T_3 were identified (18) and administration of T_3 *in vitro* increases 2-deoxy-D-glucose uptake (19–21) as well as amino acid uptake (22). Although these effects may be derived from hormone localized to the plasma membrane, evidence has also been presented that thyroid hormones enter the thymocytes (6, 15). The mechanism of entry, however, is as yet poorly defined. Since these cells also possess a well characterized ionic pump activity on the plasma membrane (23), they represent a suitable model for uptake studies involving ionic mechanisms. As a first step we investigated the general characteristics of T_3 entry in thymocytes.

Received November 14, 1988.
Address all correspondence and requests for reprints to: Marco Centanni, M.D., 2° Cattedra di Endocrinologia % 2° Clinica Medica, Policlinico Umberto I°, 00161 Rome, Italy.
* This work was supported by CNR, Institute of Experimental Medicine and Ministero della Pubblica Istruzione. A preliminary report was presented at the 16th Annual Meeting of the European Thyroid Association, 1987.

Materials and Methods

All chemicals used in this study were purchased from Sigma (St. Louis, MO), except for ouabain which was obtained from Calbiochem-Boehring (La Jolla, CA). [^{125}I]L-T_3 (3300 μCi/μg) was purchased from New England Nuclear (Boston, MA). The purity of labeled and unlabeled hormones was assayed by TLC on silica gel using formic acid/methanol/chloroform (1:3:16) according to the method of Sato and Cahnmann (24). TLC was also performed on labeled T_3 released during wash-out experiments after extraction with butanol as described by Sato and Robbins (25). Negligible deiodination was found under all conditions tested.

Thymocyte preparation

BALB-c mice, 3–6 weeks old, were obtained from Charles River Ltd. (Firenze, Italy). The animals were obtained at least

3 days before the experiment, had free access to water and commercial food pellets, and were maintained in a light-dark cycle of 12 h. Mice were killed by cervical dislocation, the thymus was removed, and thymocytes were prepared as described for rats by Segal and Ingbar (19) with some modification. Briefly, the thymus glands were washed in Krebs-Ringer-25 mM Tris-HCl (KRT) buffer, pH 7.4 (containing 2.5 mM $CaCl_2$), and finely minced. The cells were then filtered through a nylon mesh, centrifuged at 1500 rpm for 10 min at 4 C, and finally resuspended in the same buffer up to a concentration of 20×10^6 cells/ml.

Uptake studies

Before the experiments thymocytes were equilibrated for 30 min at room temperature unless otherwise specified. Incubation was carried out in KRT buffer at constant pH (7.35–7.45) by adding 25 pM [^{125}I]T_3 to 500-μl aliquots of cell suspension for the times and conditions specified in the figure legends. At the end of incubation, 200-μl aliquots were layered over an equal volume of 30% sucrose in a Beckman microtube (Beckman, Palo Alto, CA) and centrifuged at 13,000 rpm for 30 sec. The tip of the tubes was cut, and the radioactivity in the pellet was counted in a γ-counter. Negligible radioactivity was found in the sucrose cushion when counted as control. When uptake studies were performed in the presence of inhibitors, these drugs were added only during preincubation; in control experiments they were also present during incubation, but no further effects occurred compared to those in the standard experiments.

The uptake of thyroid hormone was measured as the percentage of total radioactivity per 10^6 cells. Nonspecific uptake was determined as the uptake of labeled thyroid hormone in the presence of an excess (10 μM) of unlabeled T_3. Half-maximal inhibition of T_3 was obtained by plotting the concentrations of T_3 (x-coordinate) against the percentage of specific T_3 uptake (y-coordinate); the value was obtained by the intersection on the x-axis of the line starting from the 50% inhibition point on the displacement curve.

All experiments were carried out at least in triplicate.

Washout experiments

At the end of incubation with [^{125}I]T_3 for 30 min, samples were centrifuged at 4 C for 10 min at 1500 rpm, resuspended in buffer containing 0.005% BSA for 2 min, and recentrifuged. Cells were then resuspended at 4 C in the incubation medium containing 10 μM unlabeled T_3 for different periods of time. At the end of each period 200-μl aliquots were centrifuged as described for the uptake studies. The radioactivity in the supernatant was counted to determine the labeled T_3 released into the medium. Negligible radioactivity was found when sucrose was also counted.

Cell viability was checked by trypan blue exclusion before and after the experiments; viability was greater than 95%, and no differences were found under any of the experimental conditions employed.

Protein content was measured by the method of Lowry *et al.* (26), using BSA as standard, and was found to be 40 μg protein/10^6 cells.

Statistical analysis was performed using Student's *t* test for unpaired data, checking both upper and lower limits of the samples.

Results

In time-course experiments at 37 C T_3 uptake in mouse thymocytes was linear up to 2 min, then approached a plateau. At 4 C it was much slower, and at 30 min was only one third that at 37 C (Fig. 1). The same figure shows that incubation at 37 C in the presence of an excess (10 μM) of unlabeled T_3 resulted in a decrease in [^{125}I]T_3 uptake at all time intervals, that was 48 ± 6% at 30 min. The remaining fraction represents nonspecific uptake and was used as a reference throughout the study. The addition of 10 μM unlabeled T_3 during incubation at 4 C produced no further decrease in [^{125}I]T_3 uptake compared to the total uptake at that temperature (Fig. 1). At 37 C specific T_3 uptake (Fig. 1, – – –), obtained by subtracting the nonspecific from control uptake, reached a plateau between 5 and 15 min of incubation (Fig. 1).

Specific T_3 uptake was saturable in the presence of increasing concentrations of unlabeled T_3 (Table 1). By plotting specific [^{125}I]T_3 uptake *vs.* competitor T_3 concentrations we obtained a displacement curve (not shown); from this curve (see *Materials and Methods*) it appeared that the concentration of unlabeled T_3 necessary to obtain 50% inhibition of [^{125}I]T_3 uptake was 550 nM. A comparison of the analog effect on [^{125}I]T_3 uptake is also reported in Table 1. The effects of unlabeled L-T_4, D-T_3, and triiodothyroacetic acid (TRIAC; 1 or 10 μM) were compared at 30 min of incubation to that of equal amounts of L-T_3. The results show that L-T_3 is the most powerful in displacing labeled T_3 from these low

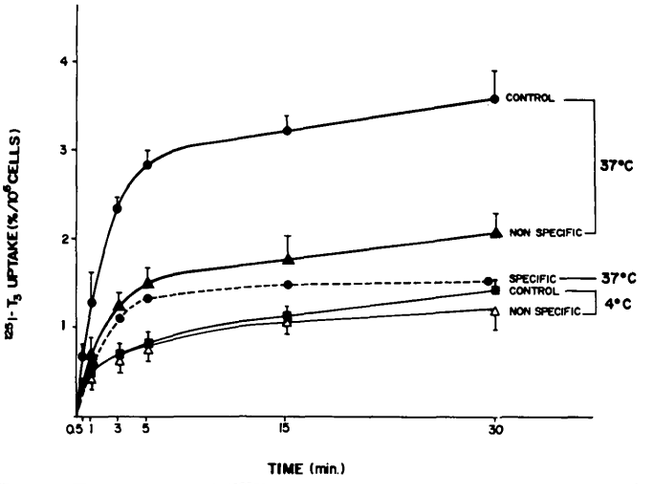

FIG. 1. Time course of [^{125}I]L-T_3 uptake. Thymocytes (10–20 × 10^6 cells/ml) were incubated with [^{125}I]T_3 (25 pM) for the indicated times at pH 7.4 (±0.05) in the absence (●, 37 C; ■, 4 C) or presence (▲, 37 C; △, 4 C) of 10 μM unlabeled T_3. The values are the mean ± SD of four experiments. The specific T_3 uptake at 37 C (●– –●) was calculated by subtracting nonspecific from total uptake.

TABLE 1. Effect of increasing concentrations of iodothyronine analogs on [^{125}I]T_3 uptake

Conc. (M)	% of specific uptake/10^6 cells	% of control
L-T_3		
10^{-9}	1.13 ± 0.21	68.5
10^{-8}	1.10 ± 0.20	67.0
10^{-7}	1.00 ± 0.18	60.1
10^{-6}	0.70 ± 0.15	42.0
10^{-5}	00 ± 0.13	0.0
D-T_3		
10^{-6}	1.27 ± 0.47	77.0
10^{-5}	1.20 ± 0.17	72.7
L-T_4		
10^{-6}	1.23 ± 0.15	74.5
10^{-5}	0.90 ± 0.13	54.5
TRIAC		
10^{-6}	1.27 ± 0.25	77.0
10^{-5}	1.25 ± 0.20	75.8

Thymocytes (10–20 × 10^6/ml) were incubated in KRT at 37 C, pH 7.40 (±0.05) with 25 pM labeled T_3 in the presence or absence of the indicated concentrations of analogs. Results are the mean ± SD of at least four different experiments.

affinity binding sites, and the analogs can be placed in rank order of decreasing inhibitory effect as follows: L-T_3 > L-T_4 > D-T_3 = TRIAC, showing that the binding sites are stereospecific.

Since the rate of uptake was linear in the first minute, we studied the initial, unidirectional uptake of unlabeled T_3 in the presence of increasing concentrations of unlabeled T_3 to obtain kinetic parameters at very low substrate concentrations. By increasing the T_3 concentration we obtained a nonlinear correlation that indicates saturability of the process. The nonspecific linear component, measured at 10 μM unlabeled T_3, was subtracted from the total, and the saturable process was analyzed according to Lineweaver-Burk plot. One high affinity binding site was apparent, with a K_m of 0.8 nM and a maximum velocity of 1 pmol/10^6 cells·min, as shown in Fig. 2.

To investigate whether T_3 uptake represents membrane-associated or intracellularly located hormone, washout experiments were carried out. Figure 3 shows that efflux of labeled T_3, after incubation at 37 C, has two components. The first one, which may represent the hormone associated with the membrane (27), was rapid and caused the efflux of 21% of the hormone from the cell in 1 min. The second one, which is believed to be the efflux from the intracellular compartment, was slower and led to a maximum release of 35% at 30 min. When incubation was performed at 4 C, 40% of the hormone exited with the first component; a maximum of 70% was reached with the slower one. Therefore, it seems that at 37 C a larger hormone fraction is located in a less accessible compartment inside the cell.

To study further the characteristics of T_3 uptake we used inhibitors that are known to interfere with different processes: oligomycin as a metabolic inhibitor, ouabain

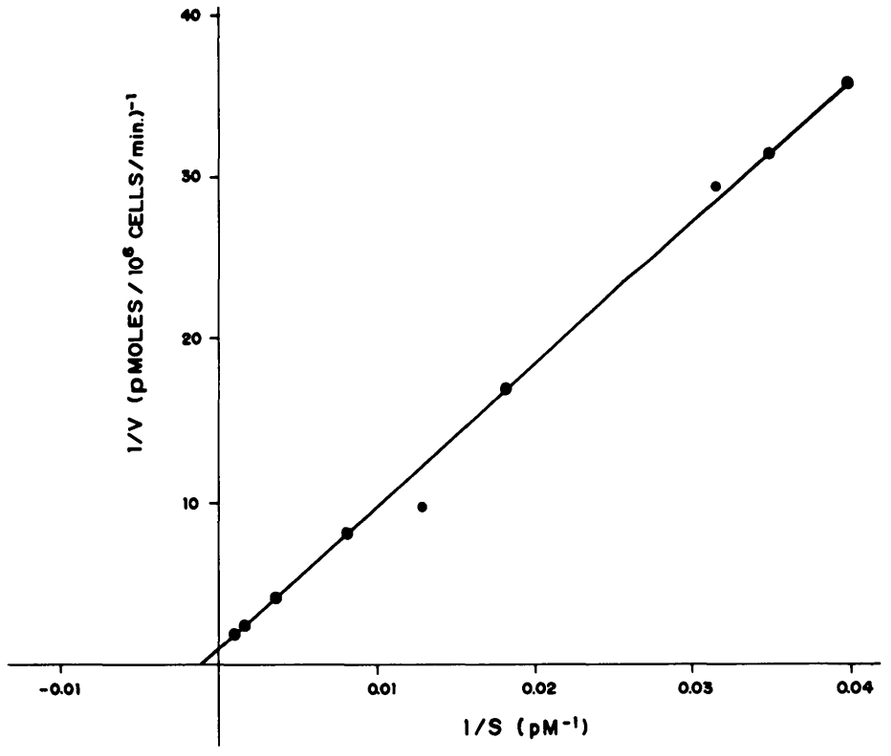

FIG. 2. Lineweaver-Burk plot of saturable T_3 uptake. Thymocytes (10–20 × 10^6 cells/ml) were incubated with 25 pM labeled T_3 for 1 min at 23 C in the presence of increasing concentrations of unlabeled T_3. Each *point* represents the reciprocal of the mean of three different experiments.

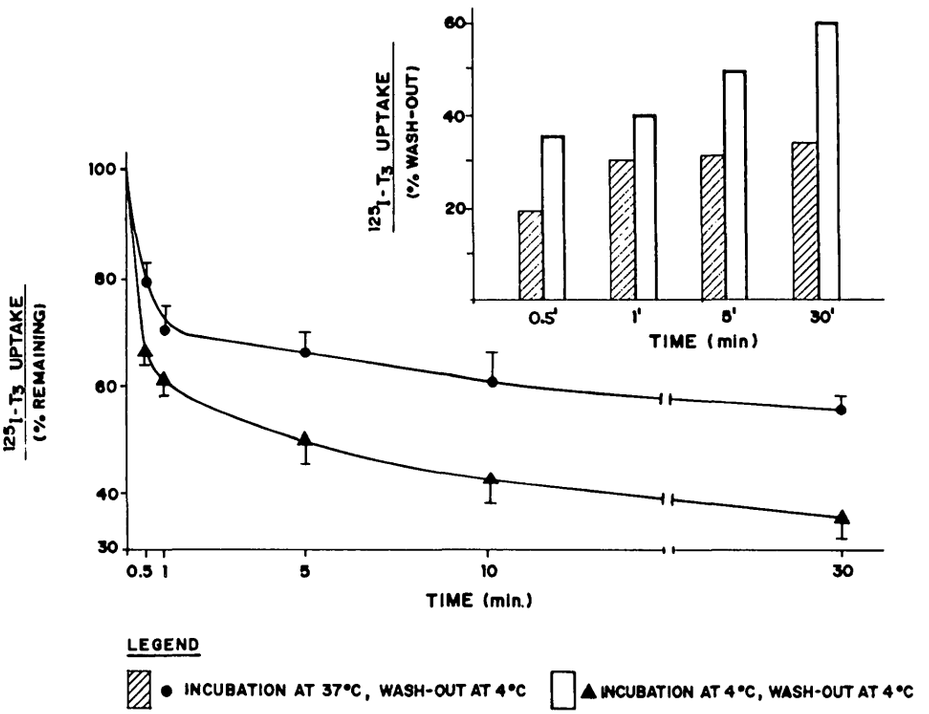

FIG. 3. Washout of the [^{125}I]L-T_3 incorporated into thymocytes. Cells (10–20 × 10^6/ml) were incubated for 30 min at 37 or 4 C, washed, and centrifuged in buffer containing 0.005% BSA, then incubated again at 4 C in the presence of 10 μM unlabeled T_3 for the indicated times. The values are the mean ± SD of five experiments.

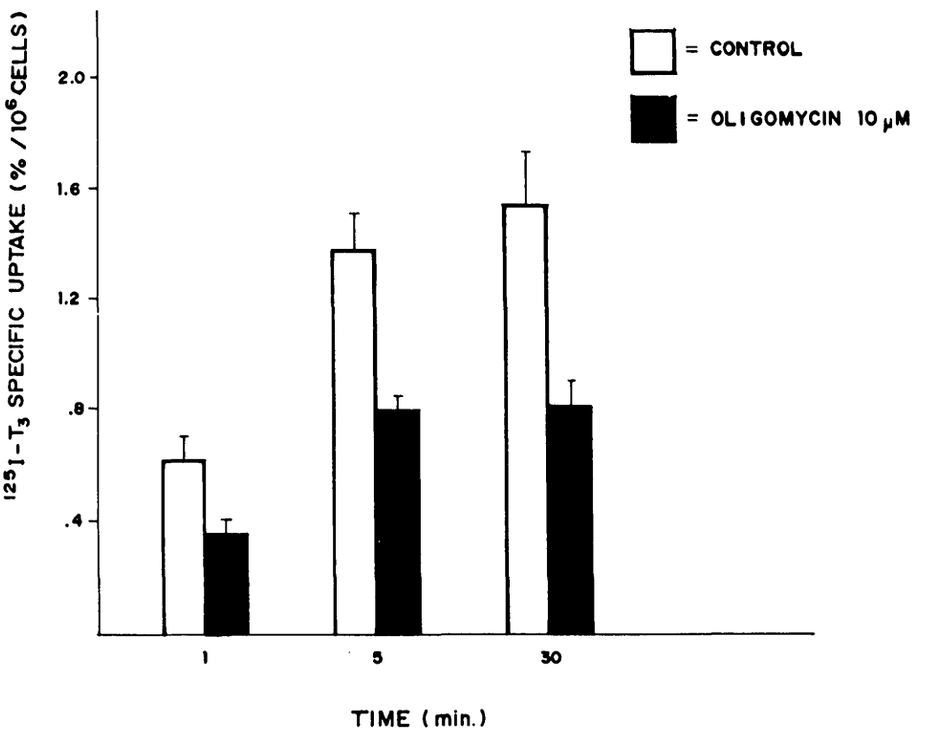

FIG. 4. Time course of the oligomycin effect on [^{125}I]L-T_3 uptake. Cells were preincubated for 30 min at 37 C and pH 7.45 (±0.05) in the presence or absence of 10 μM oligomycin, centrifuged, and then incubated in the presence of tracer amounts of labeled T_3 for 30 min under the same conditions. Results are the mean ± SD of three experiments.

as a specific blocker of Na^+/K^+-ATPase activity, and dansylcadaverine as a substance interfering with receptor-mediated endocytosis (see *Discussion*). Table 2 shows that preincubation with oligomycin resulted in a dose-dependent reduction of specific T_3 uptake that was maximal at 100 μM (−85%), suggesting that the process is energy dependent. Its effect was present after 1 min of incubation, and the maximal effect was attained in 30 min (Fig. 4). Preincubation with ouabain also inhibited the uptake in a dose-dependent fashion (−79%), indicating the requirement for a sodium gradient across the membrane and the activity of the sodium pump for such a process (Table 2). This table also shows 80% reduction of T_3 specific uptake obtained by using dansylcadaverine.

TABLE 2. Effects of metabolic inhibitors on [^{125}I]T_3 specific uptake in thymocytes

Inhibitor	n	Conc. (μM)	[^{125}I]T_3 specific uptake (% of control ± SD)
Ethanol	3	0.1%	99 ± 4[a]
Oligomycin	3	10	55 ± 5
Oligomycin	4	100	15 ± 4
Dansylcadaverine	4	10	47 ± 6
Dansylcadaverine	4	200	20 ± 8
Ouabain	5	100	42 ± 15
Ouabain	5	1	21 ± 7

n, Number of experiments, each in triplicate. Cells were preincubated for 30 min with the inhibitors; incubation with tracer hormone was measured in both the presence and absence of the inhibitor. No differences were found between the two conditions. Inhibitors were shown not to interfere with nonspecific uptake. All inhibitory effects were statistically significant ($P < 0.001$) unless otherwise specified.

[a] P = NS.

Discussion

Whether thyroid hormone entry into cells is mediated by a passive, facilitated, or active process is still a matter of controversy. The dissimilar behavior of T_3 and T_4 in some tissues raises a further unresolved question.

The present results indicate that T_3 uptake in mouse thymocytes involves an energy-dependent, saturable process. At equilibrium, half-maximal inhibition of the saturable process was comparable to that of the low affinity system found in other tissues, whose K_m ranges between 50–400 nM (3, 6, 11, 12). Initial rate studies, however, performed at low substrate concentrations, revealed the presence of a further saturable process with an apparent K_m of 0.8 nM and a maximum velocity of 1 pmol/10^6 cells·min, that was not evident at equilibrium. These kinetic parameters are fairly similar to those reported for the high affinity system in hepatocytes (4), human red cell ghosts (7), and fibroblasts (8) and believed by the same researcher to represent an active transport system. Moreover, van Doorn *et al.* (15) showed a 2.26 tissue/plasma gradient in rat thymus; in the present study, however, free intracellular T_3 was not measured, preventing us from reaching a conclusion about active transport.

The low affinity saturable T_3 uptake was stereospecific, since it was able to discriminate between L and D isomers of T_3, and unlabeled L-T_3 was also more effective than L-T_4 and TRIAC in displacing [^{125}I]T_3 from the binding sites. Since L-T_4 partially reduced specific [^{125}I]T_3 uptake, the question arises whether T_4 can share the same transport system as T_3. Many of the previous studies agreed with the hypothesis that a transport system operates for T_3 (1–3, 5–7) but not for T_4 (6, 12, 14). In hepatocytes (4), fibroblasts (8), and neuroblastoma cells (28), however, T_4 was shown to cross the membrane by a carrier-mediated process. Our preliminary data (not shown) indicate that labeled T_4 uptake was not saturable even in the presence of 10 μM unlabeled T_4 and was equal to the nonsaturable fraction of T_3 uptake. This indicates that passive diffusion is the entry pathway for T_4 in these cells, in contrast with that of T_3, as reported by Galton *et al.* (6) in rat thymus cells.

T_3 uptake in thymocytes was reduced at low incubation temperature. Evidence supporting the hypothesis that T_3 enters the cells by a temperature-sensitive process comes from the fact that during wash-out experiments, more labeled hormone was released in the medium when previous incubation was carried out at 4 C instead of 37 C; this fact indicates that at 37 C a major fraction of the hormone is in a less exchangeable compartment that, as suggested by Henneman *et al.* (27), probably represents intracellularly located hormone.

The energy dependence of this process is indicated by the finding that oligomycin, a drug that blocks ATP production by inhibiting ADP phosphorylation at the mitochondrial level, also affects specific T_3 uptake. Oligomycin reduced, in a dose-dependent fashion, both the initial and the equilibrium T_3 uptake, showing a maximal reduction of 85% of the specific uptake. The possibility that this effect could be due to some nonspecific action at the membrane level is counteracted by the fact that the effect was also found when the inhibitor was present only during preincubation, and cell viability was unchanged at the end of the experiment.

The dose-dependent sensitivity of specific T_3 uptake to ouabain, the specific inhibitor of Na^+/K^+-ATPase activity, revealed the involvement of the membrane sodium pump; as in the case of oligomycin, the ouabain effect was present as early as 1 min after the addition of labeled T_3, reaching a maximum after 30 min. This finding also points to a role of the Na^+ gradient across the plasma membrane, as described for active or facilitated T_3 transport by skeletal muscle (13), hepatocytes (4), red blood cells (1, 7), and fibroblasts (8), but in contrast to the report from Blondeau *et al.* (29) that indicated the presence of a Na^+-independent transport of T_3 in isolated hepatocytes.

Cheng *et al.* (9) showed that T_3 enters the cultured fibroblast via receptor-mediated endocytosis. This finding was confirmed by others (5, 10, 12, 29, 30) based on dansylcadaverine inhibition of T_3 entry. The present results confirm the sensitivity of specific T_3 uptake to such an inhibitor; however, a nonspecific effect of this amine, by disturbing the pH gradient, cannot be excluded, since it was suggested that pH plays a role in T_3 entry in some tissues (13, 29).

In conclusion, the results indicate that T_3 enters isolated mouse thymocytes by a saturable stereospecific

energy-dependent process that involves the activity of membrane pumps and the sodium gradient. This fact suggests the existence of a carrier-mediated process and allows the study of thyroid hormone entry related to cation trafficking across the membrane in this tissue.

Acknowledgments

The authors thank Dr. Jacob Robbins for useful discussion, suggestions, and revision of the manuscript and Drs. A. Vacca and A. Taglienti for expert technical assistance. We also thank Ms. Leda Pallante for typing the manuscript.

References

1. Holm AC, Jacquemin C 1979 Membrane transport of L-triiodothyronine by human red cell ghosts. Biochem Biophys Res Commun 89:1006
2. Eckel J, Rao GS, Rao ML, Breuer H 1979 Uptake of L-triiodothyronine by isolated rat liver cells. Biochem J 182:473
3. Holm AC, Wong KJ, Pliam NB, Jorgensen EC, Goldfine ID 1980 Uptake of L-triiodothyronine into human cultured lymphocytes. Acta Endocrinol (Copenh) 95:350
4. Krenning E, Docter R, Bernard B, Visser TJ, Hennemann G 1981 Characteristics of active transport of thyroid hormone into rat hepatocytes. Biochim Biophys Acta 676:314
5. Halpern J, Hinkle PM 1982 Evidence for an active step in thyroid hormone transport to nuclei: drug inhibition of L-[^{125}I]triiodothyronine binding to nuclear receptors in rat pituitary tumor cells. Endocrinology 110:1070
6. Galton VA, St Germain DL, Whittemore S 1986 Cellular uptake of 3,5,3′-triiodothyronine and thyroxine by red blood and thymus cells. Endocrinology 118:1918
7. Holm C 1987 Active transport of L-triiodothyronine through the red cell plasmamembrane: true or false? Scand J Clin Lab Invest 47:185
8. Docter R, Krenning E, Bernard H, Hennemann G 1987 Active transport of iodothyronines into human cultured fibroblasts. J Clin Endocrinol Metab 65:624
9. Cheng S-Y, Maxfield FR, Robbins J, Willingham MC, Pastan I 1980 Receptor-mediated uptake of 3,5,3′-triiodothyronine by cultured fibroblasts. Proc Natl Acad Sci USA 77:3425
10. Horiuchi R, Cheng SY, Willingham M, Pastan I 1982 Inhibition of the nuclear entry of 3,3′,5-triiodo-L-thyronine by monodansylcadaverine in GH3 cells. J Biol Chem 257:3139
11. Docter R, Krenning EP, Bos G, Fekkes D, Hennemann G 1982 Evidence that the uptake of triiodo-*l*-thyronine by human erythrocytes is carrier mediated but not energy-dependent. Biochem J 208:27
12. Pontecorvi A, Robbins J 1986 Energy-dependent uptake of 3,5,3′-triiodo-L-thyronine in rat skeletal muscle. Endocrinology 119:2755
13. Centanni M, Robbins J 1987 Role of sodium in thyroid hormone uptake by rat skeletal muscle. J Clin Invest 80:1068
14. Rao GS, Rao ML 1983 L-thyroxine enters the rat liver cell by simple diffusion. J Endocrinol 97:277
15. van Doorn J, Roelfsema F, van der Heide D 1985 Concentrations of thyroxine and 3,5,3′-triiodothyronine at 34 different sites in euthyroid rats as determined by an isotopic equilibrium technique. Endocrinology 117:1201
16. van Doorn J, Van Der Heide D, Roelfsema F 1983 Sources and quantity of 3,5,3′-triiodothyronine in several tissues of the rat. J Clin Invest 72:1778
17. Larsen PR, Silva JE, Kaplan MM 1981 Relationships between circulating and intracellular thyroid hormones: physiological and clinical implications. Endocr Rev 2:87
18. Segal J, Ingbar SH 1982 Specific binding sites for triiodothyronine in the plasmamembrane of rat thymocytes. J Clin Invest 70:919
19. Segal J, Ingbar SH 1979 Stimulation by triiodothyronine of the *in vitro* uptake of sugars by rat thymocytes. J Clin Invest 63:507
20. Segal J, Ingbar SH 1980 Stimulation of 2-deoxy-D-glucose uptake in rat thymocytes *in vitro* by physiological concentrations of triiodothyronine, insulin, or epinephrine. Endocrinology 107:1354
21. Segal J, Ingbar SH 1981 Studies on the mechanism by which 3,5,3′-triiodothyronine stimulates 2-deoxyglucose uptake in rat thymocytes *in vitro*. Role of calcium and adenosine 3′,5′-monophosphate. J Clin Invest 68:103.
22. Goldfine ID, Simons CG, Ingbar SH 1975 Stimulation of the uptake of γ-aminoisobutyric acid in rat thymocytes by L-triiodothyronine: a comparison with insulin and dibutyryl cyclic AMP. Endocrinology 96:802
23. Grinstein S, Cohen S, Rothstein A 1984 Cytoplasmic pH regulation in thymic lymphocytes by an amiloride-sensitive Na^+/H^+ antiport. J Gen Physiol 83:341
24. Sato K, Cahnmann HJ 1980 Synthesis of 3,5-^{125}I-triiodothyronine of high specific activity. Anal Biochem 102:237
25. Sato K, Robbins J 1980 Thyroid hormone metabolism in cultured monkey hepatocarcinoma cells: monodeiodination activity in relation to cell growth. J Biol Chem 255:7347
26. Lowry OH, Rosebrough NJ, Farr AL, Randall J 1951 Protein measurement with the Folin phenol reagent. J Biol Chem 193:265
27. Henneman G, Krenning EP, Bernard B, Huvers F, Mol J, Docter R, Visser TJ 1984 Regulation of influx and efflux of thyroid hormones in rat hepatocytes: possible physiologic significance of the plasma membrane in the regulation of thyroid hormone activity. Horm Metab Res [Suppl 14] 1:000
28. Goncalves E, Lakshmanan M, Foti D, Robbins J, Thyroid hormone transport into different brain cell lines. 62nd Annual Meeting of the American Thyroid Association, Washington DC, 1987 (Abstract 80)
29. Blondeau JP, Osty J, Francon J 1988 Characterization of the thyroid hormone transport system of isolated hepatocytes. J Biol Chem 263:2685
30. Willingham MC, Pastan I 1984 Endocytosis and membrane traffic in cultured cells. Recent Prog Horm Res 40:569