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Tissue uptake of thyroid hormone by amino acid transporters

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Thyroid hormones (THs) – thyroxine (T_4) and tri-iodothyronine (T_3) – are iodinated derivatives of the amino acid tyrosine, which regulates growth, development and critical metabolic functions. THs are taken up by target cells and act at the genomic level via nuclear thyroid receptors. Saturable transport mechanisms mediate the greater part of TH movement across the plasma membrane. System L1 permease is a transporter of THs and amino acids in mammalian adipose tissue, placenta and brain. T_3 is also a substrate of a putative System T transporter, which is selective for aromatic amino acids. The activity and functional mechanisms of these transporters can be crucial to cells in determining both their hormone sensitivity and their responses to change in circulating hormone concentrations or availability of competing substrates (e.g. amino acids). TH transporters are potentially important pharmacological targets in the design of novel or improved therapies for thyroid-related disorders.

Key words: amino acid; membrane transport; nuclear hormone action; thyroid disease; thyroid hormone; tryptophan.

INTRODUCTION

The formation of thyroid hormones (THs) within the thyroid follicle involves the coupling of iodinated L-tyrosine residues in thyroglobulin.[1] Indeed, THs retain an amino acid moiety within the iodothyronine molecular structure (Figure 1A). The plasma-free hormone concentration appears to be the key determinant of biological activity[2,3], so THs must

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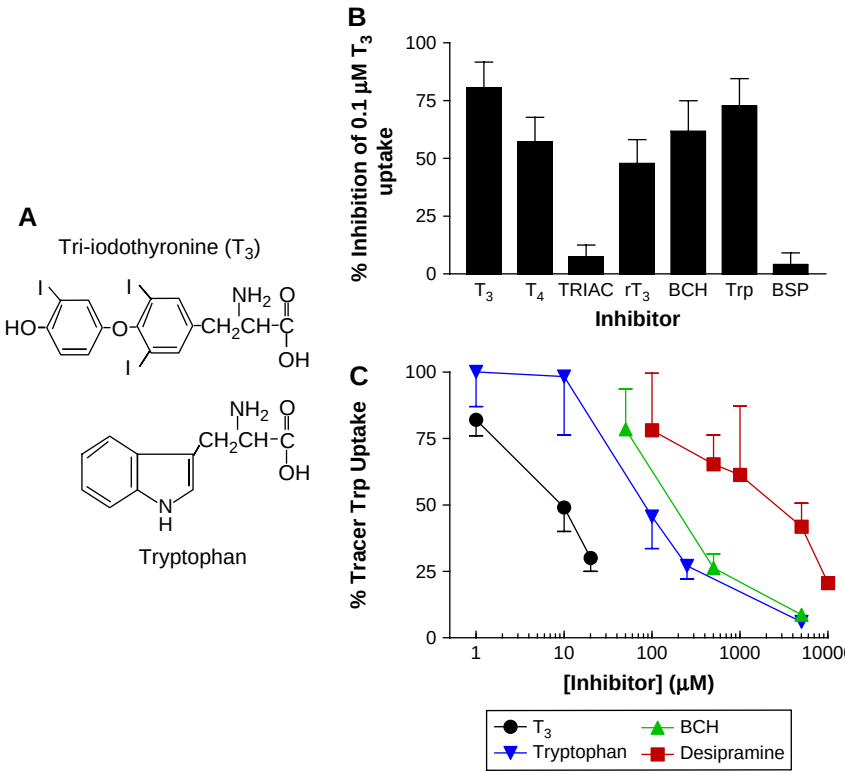

Figure 1. (A) Chemical structures of tri-iodothyronine (T_3), the most potent biologically active iodothyronine, and the aromatic amino acid tryptophan. (B) Inhibition of [^{125}I]-T_3 (0.1 μM) uptake by iodothyronines and amino acids in *Xenopus* oocytes injected with 4F2hc-xLAT1 cDNAs or water. Inhibitor concentrations were 10 μM iodothyronines [T_3, T_4, triodothyroacetic acid (TRIAC) and reverse T_3 (rT_3)], 5 mM synthetic bicyclic amino acid analogue 2-amino-bicycloheptane-2-carboxylic acid (BCH), 10 mM tryptophan (Trp) and 200 μM bromosulphophthalate (BSP; a classic organic anion transporter substrate). Each bar represents uptake in the presence of inhibitor as a percentage of control uptake measured in the absence of inhibitor (mean value ± SEM for at least seven oocytes). (C) Concentration-dependent inhibition of 4F2hc-xLAT1-induced [^{3}H]-tryptophan uptake by unlabelled T_3 or tryptophan. Data show the uptake of tryptophan (1 μM tracer) in the presence of increasing concentrations of unlabelled inhibitor, as a percentage of control uptake in the absence of inhibitor. Each point represents mean value ± SEM for at least 8–11 4F2hc-xLAT1-injected oocytes, after appropriate correction for uptake in water-injected oocytes.

first cross the plasma membrane of target cells to exert their major effects at the genomic level through binding of hormone (predominantly tri-iodo-L-thyronine; T_3) to nuclear thyroid receptors.[4] The biologically active iodothyronines, principally T_3 and L-thyroxine (T_4), are relatively small (molecular weight of about 700) hydrophobic molecules, which at first glance should be able to enter target cells at reasonable rates by means of passive diffusion through the lipid bilayer of the cell membrane. However, the polar amino acid side-chain retards their passage across the cell membrane so that, once partitioned into the membrane, THs tend to remain in the outer half of the lipid bilayer.[5]

It is now clear that saturable transport mechanisms mediate the greater part of TH import and export at the plasma membrane of cells (see refs 6–8 for review). Studies

using isolated cells indicate that a broad range of transporter types accepts THs as substrate, including amino acid transporters[9–11], monocarboxylate transporters[12] and classic multispecific organic anion/cation transporters[13,14], as well as multidrug-resistance (MDR) pumps[15,16] and fatty acid translocase.[17] It is also becoming clear that the transport of THs into their target tissues by saturable mechanisms is important for physiological control of both their action and metabolism.[3,18,19] This chapter focuses on the role of amino acid transporters in TH transport across cell membranes, and their functional significance. The numerous families of amino acid transporters have different substrate selectivities, mechanisms and physiology (see refs 20–24 for a review), and together are conventionally classified in terms of functional 'Systems' (see refs 25–27 for further information). The amino acid transport systems currently identified as TH transporters are classified as System L and System T.

AMINO ACID TRANSPORTERS ACCEPTING THYROID HORMONES AS SUBSTRATE

System L

System L (leucine preferring) is an ion-independent transporter for large, neutral amino acids (LNAA). It has a broad tissue distribution in mammals[22,28] and is generally characterized by the ability to transport branched-chain and aromatic amino acids (e.g. leucine and tryptophan) and to accept the synthetic bicyclic amino acid analogue 2-amino-bicycloheptane-2-carboxylic acid (BCH) as a substrate.

The earliest studies reporting an effect of TH on the transport of amino acids were performed on *Xenopus laevis* embryos.[29,30] System L was first clearly implicated in the uptake of TH into cultured rat astrocytes.[31] Two distinct L-type transporter systems (L1 and L2) were identified in the astrocytes, but only the high-affinity L1 system was demonstrated to be inhibited by T_3 in a competitive manner. The K_m for T_3 uptake into astrocytes was 2 μM, with a K_i of 2–3 μM for the T_3 block of tryptophan uptake by System L1. This indicated that T_3 and tryptophan were both substrates for the L1 transporter and that they therefore shared a common uptake mechanism. Studies using the JAR human placental choriocarcinoma cell line have clearly demonstrated that THs are high-affinity competitive inhibitors of System L.[32] The K_m value for T_3 transport in JAR cells was 0.8 μM, similar in magnitude to that obtained in astrocytes (2 μM)[31], although the authors expressed caution as to whether THs were substrates or merely inhibitors of the System L transport mechanism. Subsequent work, also using JAR cells[33], confirmed a mutual competitive inhibition between uptake systems for T3 and tryptophan but also indicated that T_3 was transported into JAR cells by at least two transport systems with differing substrate specificities, one of which resembled a System L-like amino acid transporter.[34,35] These studies also revealed that efflux of T_3 was progressively inhibited by increasing concentrations of both T_3 and tryptophan, i.e. it was saturable.

Molecular cloning of System L[28,36] revealed a holotransporter consisting of two subunits, a hydrophobic permease light chain (LAT1; SLC7A5) and a regulatory glycoprotein heavy chain (4F2hc/CD98; SLC3A2) (Figure 2). Several additional permease subunits have subsequently been identified, although only two (LAT1, LAT2) have the transport characteristics of System L (see ref. [22] for a review). Following up on the observations that suggested a close relationship between TH transport and System L, we demonstrated that a heterodimer of 4F2hc and xLAT1 (the *Xenopus*

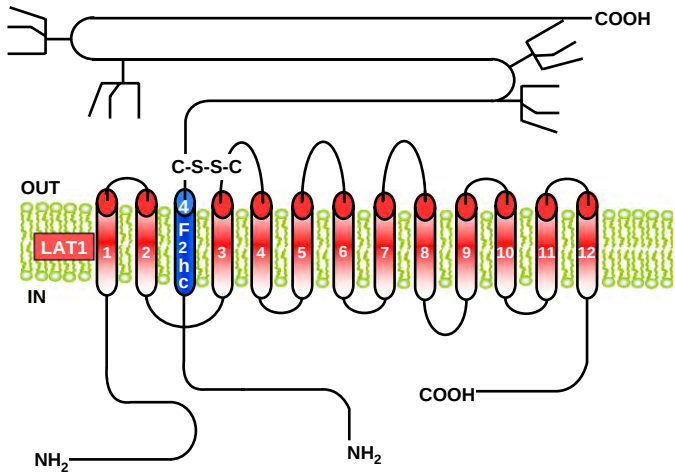

Figure 2. Predicted structures of 4F2hc, the light-chain permease LAT1 and their interactions. The putative transmembrane domains of the light chain are numbered 1–12. The potential glycosylation sites of 4F2hc are designated as 'forks', and the location of the cysteine residues forming the intermolecular disulphide bridge are indicated (C–S–S–C). Adapted from ref. 36.

homologue of mammalian LAT1, also known as IU12) transports TH when expressed in amphibian oocytes.[9] The transport of TH by 4F2hc-xLAT1 heterodimers was competitively inhibited by both BCH and tryptophan, two classic substrates of System L. In addition, reverse T_3 (rT_3) but not tri-iodothyroacetic acid (TRIAC; a TH analogue lacking an amino acid moiety) inhibited TH uptake, demonstrating structural specificity of the transport system[9] (see Figure 1B), which was clearly different from specificity of intracellular TH binding sites. Kinetic studies showed that this transporter has K_m values of 1.8 and 6.3 μM for T_3 and T_4, respectively; these values are appropriate for the System L1 subtype. The K_m values obtained for TH uptake by the 4F2hc-xLAT1 holotransporter are markedly lower than for Trp transport (85 μM; see Figure 1C), Human LAT1 and LAT2 were subsequently shown to also mediate uptake of TH[11], with similar K_m values to those found for xLAT1/IU12. The glycoprotein heavy chain 4F2hc/CD98 is a multifunctional protein with involvement in integrin signalling[37], as well as delivery of the catalytic permease light chain(s) to the plasma membrane from the cytosol.[38]

We and others have examined the contribution of System L to TH transport in several mammalian tissues and cell types; a representative selection of our data is summarized in Figure 3. The pattern emerging from this type of study, which utilizes radiolabelled TH as a transportable tracer, has several key features: (1) a significant proportion of TH associating with cells appears to do so by non-saturable processes (this will include partitioning into the plasma membrane and passive diffusional uptake); (2) the extent of TH–amino-acid interaction is tissue specific; (3) aromatic amino acids (notably tryptophan) can interact with TH transport to a greater extent than can be accounted for by System L (which suggests an important role for another amino acid transporter, known as System T, as considered below). The data in Figure 3 also show the diversity of entry routes for TH in a cell-type-specific context, with organic anion transporters (e.g. in SH-SY5Y neuronal cells) and monocarboxylate transporters (e.g. in HepG2 liver cells) making major contributions in many cases. Nevertheless, we have

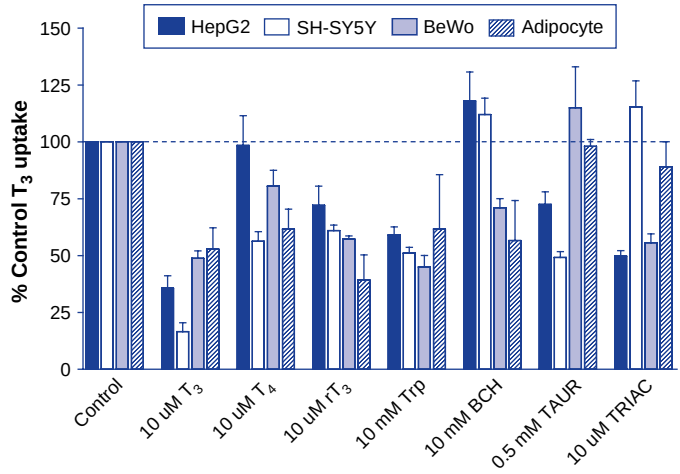

Figure 3. Inhibition of [^{125}I]-T_3 (0.05 μM) uptake by iodothyronines, amino acids and taurocholate at indicated concentration in four mammalian cell types: HepG2 human hepatoma cells, SH-SY5Y human neuroblastoma cells, BeWo human choriocarcinoma (placenta) cells and primary rat adipocytes. Each bar represents uptake in the presence of inhibitor as a percentage of control tracer uptake measured in the absence of inhibitor (mean value ± SEM for at least four separate experiments); inhibition largely reflects competitive inhibition of saturable T_3 uptake into cells under the experimental conditions employed. The iodothyronine concentrations used are close to the limit of free solubility and sufficient to saturate most known TH transporters, except for the iodothyronine analogue triodothyroacetic acid (TRIAC), which lacks the amino acid moiety but is a good substrate for the MCT8 monocarboxylate transporter. The amino acids tryptophan (Trp), the synthetic bicyclic amino acid analogue 2-amino-bicycloheptane-2-carboxylic acid (BCH) and the organic anion taurocholate (TAUR) are used in excess of saturating concentrations.

established that the mammalian System L permease LAT1 is a major effector of TH uptake into tissues, such as adipose tissue[39] and placenta[40], which express both LAT1 and 4F2hc.

We now know that more than one family of transporters contributes to system L transport activity. LAT3 and LAT4 exhibit Na^+-independent and BCH-sensitive leucine transport showing the properties of System L[41,42]; however, they do not require 4F2hc for functional expression. Based on substrate selectivity, affinity, and *N*-ethylmaleimide sensitivity, LAT3 at least is proposed to be a transporter subserving System L2. Human LAT4 exhibits 57% identity to human LAT3[42] and they have been proposed as a new family of organic solute transporters (SLC43), only weakly related to members of the heteromeric amino acid transport family (such as SLC7A5 and SLC7A8, which encode LAT1 and LAT2, respectively). Neither LAT3 nor LAT4 appears to recognize T_3 as a substrate and are therefore not TH transporters, which might account for the perplexing reports of the functional identification of System L activity in cell types that do not show evident System-L-type TH transport.

System T

System T (tryptophan preferring) is an ion-independent transporter specific for aromatic amino acids. It does not accept BCH as a substrate. The first detailed report

of TH uptake by an amino acid transport system was in erythrocytes[43] and showed that all aromatic amino acids (tryptophan, phenylalanine and tyrosine) had an inhibitory effect on T_3 uptake and that T_3, T_4 and D-T_3 could inhibit L-tryptophan uptake, consistent with functional properties of System T. These interactions between T_3 and tryptophan were mutually competitive and the concentrative uptake of T_3 was by an exchange mechanism, whereby erythrocytes preloaded with tryptophan or phenylalanine produced a transacceleration of T_3 uptake in exchange for amino acid release. Similarly, T_3 shares a common transport mechanism with aromatic amino acids, via System T, in trout[44] and frog tadpole[45,46] erythrocytes. T_3 and T_4 uptake in the embryonic heart cell line H9c2 is also dose-dependently inhibited by tryptophan (maximum inhibition around 70%).[47] Furthermore, System T appears to contribute to T_3 uptake into liver.[48] Indeed, it is apparent from analysis of Figure 3 that tryptophan is the only non-iodothyronine interacting significantly with T_3 uptake in all cell types studied. Whereas System L probably accounts for this interaction in BeWo cells and adipocytes, it is clear that a distinct System-T-like transport mechanism for TH operates in liver cells, neuronal cells and erythrocytes at least.

The molecular identity of this transport activity is still under investigation. A System T amino acid transporter (TAT1, also called MCT10) has been characterized to transport aromatic amino acids but, surprisingly, initial reports indicate that it does not transport iodothyronines.[49,50] A very recent study (E.C.H. Friesema et al, manuscript under review) challenges these reports by showing that TAT1/MCT10 is indeed effective in mediating both influx and efflux of THs when transfected into COS (monkey kidney) cells. TAT1/MCT10 recognizes amino acid substrates as anions, consistent with its structural similarity to H^+/monocarboxylate transporters of SLC16 family. This transporter family includes the now well-established TH transporter MCT8. However, despite the fact that TAT1/MCT10 shares 50% amino acid identity with MCT8, the latter does not transport tryptophan or other amino acids[12], although it accepts TRIAC and organic anions such as BSP as substrates alongside T_3, T_4 and rT_3.[12] Furthermore, to our knowledge, none of the classic organic anion transporters accepts tryptophan as a substrate. It is conceivable that tryptophan and TH are as yet unrecognized competing substrates for other transporter types: recent pharmacological evidence links both the organic cation transporter (OCT) family and the P-glycoprotein/multidrug-resistance (MDR) export pumps in cellular TH transport.[15,33,51] The MDR transporters are members of the ATP-binding cassette (ABC) superfamily of transporter proteins and a related member of this family of transporters has a *Drosophila* homologue that is involved in cellular uptake of guanine and tryptophan.[52]

We have previously proposed that the uptake of T_3 via System T across rat liver plasma membrane involved a 'receptor–transporter' coupled mechanism[48], by which T_3 binds to a high-affinity receptor, which then facilitates the uptake of T_3 via a distinct System T transport protein. High-affinity binding sites for TH have in fact been identified in the plasma membranes of tissues, including placenta[53], liver[54] and brain.[55] Treatment of liver membranes with the detergent Triton-X100 results in removal both of T_3 binding sites and inhibition of tryptophan uptake by T_3, whereas basal tryptophan uptake itself is unaffected.[48] Further research is required to clarify whether these putative receptors need to be co-expressed alongside transporter proteins such as TAT1/MCT10 to produce the functional characteristics of System T transport for TH. An analogous receptor–transporter mechanism has been described for the uptake of folate into JAR cells[56], involving the interaction of a folate receptor, transporter and also the H^+-pump.

PHYSIOLOGICAL RELEVANCE

Cellular thyroid hormone bioavailability and actions

The vast majority of THs in the serum are bound to proteins, including serum albumin, transthyretin and a number of lipoproteins; the resulting free TH concentrations are in the picomolar range. The overall values obtained for the K_m (0.2–7 μM) of saturable TH uptake by Systems L and T (and other TH transport mechanisms; see refs 6,7 for a review) are between 25 and 100 times higher than their total serum levels (2 nM and 110 nM in human serum for T_3 and T_4, respectively[1]), enabling a linear delivery of T_3 and T_4 to the cell via their saturable transport mechanisms under physiological circumstances. The K_m values for cellular TH uptakes by amino acid transporters are also 50–100 times lower than the corresponding values for tryptophan uptake (95–1800 μM) and T_3 at picomolar concentrations enters BeWo cells at measurable rates through System L even when amino acids are present externally at physiological plasma concentrations.[18]

Using human cell lines, we have shown that blockade of System L with tryptophan or BCH significantly reduces total T_3 uptake and T_3 binding to cell nuclei, apparently without disturbing nuclear binding kinetics (the nuclear:cytosol T_3 ratio is about 0.12 in all cases). Furthermore, we confirmed that these effects on nuclear binding of T_3 mirror activation of gene transcription by the hormone.[18] Cell membrane transport of T_3 therefore appears to be an important determinant of nuclear T_3 entry and TH action under such conditions. Both LAT1 and 4F2hc subunits of System L1 are highly regulated genes responsive to endocrine and stress stimuli[8,22] and, indeed, xLAT1 (IU12) was originally identified as an early T_3 response gene associated with cell activation, organ development and establishment of tissue TH competence.[57] Such up-regulation of transport activity by T_3 should help ensure both that sufficient amino acids enter the cell to sustain levels of growth and that sufficient THs reach the cell nucleus to maintain the high expression levels of metabolic and anabolic proteins required over these periods.

The different cloned TH transporters exhibit a range of functional properties. LAT, organic anion transporter (OATP), OCT and Na^+-taurocholate carrier protein (NTCP) carriers have recognized uptake or exchange properties, whereas MDR transporters are reported to direct movement of major substrates out of cells. Although their respective physiological significance for effecting or regulating TH bioavailability and action is yet to be fully elucidated, it is reasonable to suggest that exchangers such as LAT1 might either take up or release TH, depending on circumstances, such as the prevailing concentrations of competing substrates or potential exchange partners (helping to explain, for example, why external amino acids influence the steady-state distribution ratio of T_3 across the plasma membrane of BeWo cells[18]). It is now evident that transport proteins such as MDR, which mediate active cellular TH export, can have a significant influence on cellular TH levels.[15,33,58]

Both T_4 and T_3 are substrates for System L and System T amino acid transporters, with a slight preference for T_3. The naturally occurring iodothyronine rT_3 has an elevated plasma concentration in catabolic states, such as prolonged fasting, but is not transcriptionally active. Nevertheless, reports indicate that rT_3 might act as a modulator of TH action at the level of the cell membrane.[59–61] We now know that rT_3 is a potent competitive inhibitor of TH uptake by several different transporter types, and such

effects might provide a mechanism through which rT_3 acts as a previously overlooked negative regulator of both genomic and non-genomic TH actions.

The TH binding sites on plasma membranes[54,62–64] might facilitate cellular TH uptake of both free and serum protein-bound forms[3,48] by functional co-operation with TH transporters. A proteomic analysis of liver membrane proteins removed by Triton-X100 revealed a known TH binding protein – protein disulphide isomerase[65] – which is expressed at the cell surface in certain tissues[66] and might thus conceivably act as a TH receptor (F. Rafiqi and P. M. Taylor, unpublished observations). Serum albumin has been suggested to have a key role in carrier-mediated TH transport through the rat blood–brain barrier[67], a tissue that expresses LAT1 at high levels (see below). This process is believed to involve enhanced dissociation of TH from albumin as a result of transient conformational changes about the ligand binding site due to interaction of the serum protein with the surface of the microcirculation, possibly via surface receptors.[68] Similarly, albumin is reported markedly to stimulate iodothyronine uptake by MCT8 overexpressed in *Xenopus* oocytes.[12] It is intriguing to note that tryptophan is the only amino acid that binds substantially to serum proteins (80–90% bound to albumin), offering another possible site of tryptophan/TH competition for cellular uptake.

Tissue specificity of the TH transporter function

One key factor determining the likely importance of a transporter for TH signalling and action is the range of tissues in which it is expressed. The mammalian 4F2hc and LAT1/LAT2 proteins are widely expressed[28] and amino acid transporters might be the most important saturable transport mechanisms for TH transport across plasma membranes of TH target tissues, such as adipose tissue, as well as across barrier tissues, such as the placenta and the brain capillary endothelium (which forms the blood–brain barrier).

Adipose tissue

Fat is an important target tissue for TH action, the effects of which include regulation of expression of lipogenic enzymes[69] and modulation of hormone sensitivity via regulation of expression of receptor number.[70] System L appears to be the major TH transporter in white adipocytes[39] and it is noteworthy that the putative TH receptor protein disulphide isomerase (see above), which interacts with disulphide bonds in membrane proteins (such as found in the System L holotransporter; see Figure 2), is expressed at high levels in adipose tissue.[71] Hypothyroidism reduces TH uptake through System L in rat adipocytes (i.e. the process is regulated).[39]

Placenta

The transplacental supply of TH from maternal blood is essential for normal fetal development.[72] Maternal–fetal transfer supplies the human embryo with low levels of TH in the early gestation prior to onset of fetal thyroid function.[73,74] The levels of 4F2hc and LAT1 proteins in human placenta increase at full term compared with those at midtrimester[75], coinciding with peak TH concentrations in embryonic serum over the perinatal period.

Blood–brain barrier

T_4, T_3 and rT_3 are all capable of bidirectional transfer across the blood–brain barrier[76] and System L is thought to have a key role in these transfers, given that LAT1 is the principal neutral amino acid transporter expressed at the blood–brain barrier.[77] Nevertheless, the high-affinity T_4 transporter OATP1c1 is also expressed in capillaries throughout the brain and might be crucial for T_4 uptake over the blood–brain barrier (see ref.7 for a review). The K_m of blood–brain barrier transport of LNAA is in the 0.1–0.6 mM range, which approximates the physiological plasma concentrations and forms the basis of the unusual sensitivity of the brain to competition effects on LNAA transport. The activity of LNAA transport at the blood–brain barrier is influenced by changes in thyroid status[78] and brain plasma membrane vesicles derived from T_3-treated rats accumulate three times more LNAA than controls[79,80]; a finding that is consistent with the consensus that THs promote the establishment of neurotransmission in the developing nervous system.

Other tissues

Amino acid transporters are thought to be important for the availability of TH during the differentiation of heart muscle cells[47] and a tryptophan-inhibitable transport pathway accounts for approximately 40% of saturable T_3 uptake into differentiating skeletal muscle (C2C12) cells (C.J. Stockdale and P.M. Taylor, unpublished observations). System T appears to be the major TH transporter of erythrocytes, although the functional importance is unclear.

CLINICAL PERSPECTIVES

The discovery that TH transporters are also capable of transporting other molecules offers a potential pharmacological route for modulation of TH function. Given the tissue specificity of the expression of these transporters, one can envisage the use of different inhibitors to target TH function in specific organs/tissues. TH transport mediated by LAT1 is blocked by the tricyclic antidepressant desipramine (see Figure 1C), offering a possible explanation for the hypothyroid-like side-effects of desipramine treatment[6,9] and also providing a possible starting point for design of drugs targeted to TH transporters. The identification of novel high-affinity System L inhibitors (such as compounds based on the natural product brasilicardin A[81]) offers additional opportunities in this area.

Any change in the plasma TH/LNAA ratio can influence whole-body thyroid status by altering cellular TH delivery. Such effects might be too small or too transient to be of significance under normal circumstances, but can become important in situations where the TH/NAA balance is disturbed by marked changes in plasma NAA concentrations, for example in phenylketonuria or during tryptophan depletion.

Phenylketonuria

In maternal phenylketonuria, raised levels of phenylalanine can restrict delivery of amino acids such as tryptophan to the fetus.[82,83] It has been suggested that the resulting amino acid imbalance contributes to the congenital heart disease and mental retardation of maternal phenylketonuria[84], but effects of the concomitant reduction in fetal

TH delivery should now also be considered. There is some evidence that managed phenylketonuria subjects have subclinical TH imbalances (particularly with respect to T_4 and rT_3), although this issue is complicated by the fact that individuals consuming protein-restricted diets, such as patients with phenylketonuria, are at risk of selenium deficiency, which will also affect thyroid status.[85,86]

Acute tryptophan depletion

Tryptophan is the precursor of the neurotransmitter serotonin (5-HT) and acute tryptophan depletion (ATD), a means of reducing brain 5-HT synthesis, has emerged as an important tool for investigating 5-HT function (see ref.[87] for a review). ATD can achieve a marked lowering of tryptophan in both plasma and cerebrospinal fluid (by $>$ 85%) over periods of several hours.[88,89] Decreased serotoninergic neurotransmission has been proposed to play a key role in the aetiology of depression and ATD results in a depression of mood in normal subjects.[87,89] Plasma T_3 is elevated in tryptophan-deficient chicks, accompanied by lower rT_3[90], indicating that the possible effects of ATD on thyroid status (a variable known to influence 5-HT neurotransmission[91]) require further evaluation. This might have broader relevance in that chronic depletion of plasma tryptophan (and resultant impairment of brain 5-HT neurotransmission) is identified as a mechanism whereby persistent dieting can trigger the development of eating disorders, such as bulimia nervosa, in vulnerable individuals.[88,92] In this regard, it is noteworthy that rats placed on a tryptophan-deficient diet show a progressive decrease in thyroid function over 2 months.[93]

SUMMARY

Saturable transport mechanisms are responsible for the greater part of TH movement across the plasma membrane. The System L1 permease is a transporter of TH and amino acids in tissues, including adipose tissue, placenta and brain. T_3 is also a substrate of the System T transporter, which is selective for aromatic amino acids. The activity of these transporters is thought to be crucial for determining a cell's TH competence and response to changes in circulating levels of TH and nutrients. The function of TH-binding proteins and surface TH receptors in cellular TH delivery requires further investigation. TH transporters represent novel pharmacological targets for the design of improved therapies for thyroid disorders.

Practice points

- TH actions in target tissues may be influenced by changes in plasma concentration of substances competing with TH for cellular uptake by TH transport proteins (the latter include at least two types of amino acid transporter).
- The likely significance of such effects will be increased in situations where plasma amino acid concentrations are markedly disturbed, such as phenylketonuria or acute tryptophan depletion (ATD).

Research agenda

- Knockdown of System L expression (by short-hairpin RNA in cells or gene knockout in mice) should help elucidate the role in TH action.
- The function of TH-binding proteins (e.g. albumin) and surface receptors (e.g. protein disulphide isomerase) in cellular TH delivery requires further investigation.
- The influence of altered thyroid status on expression of TH transporters in different tissues needs to be examined.
- The effects of ATD on TH status should be fully evaluated.

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