

Transport of D-[1-¹⁴C]-amino acids into Chinese hamster ovary (CHO-K1) cells: implications for use of labeled D-amino acids as molecular imaging agents

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Abstract

Introduction: The fact that D-amino acids have been found in various tissues and are involved in various functions is a clue to how to develop new imaging agents. We examined D-amino acid transport mechanisms in Chinese hamster ovary (CHO-K1) cells because CHO-K1 cells are widely used in biomedical studies and are thought to be useful for expression of genes involved in metabolism of D-amino acids.

Methods: Uptake experiments were performed. CHO-K1 cells cultured in 60-mm plastic culture dishes under ordinary culture conditions were incubated with 18.5 kBq of radiolabeled amino acid in 2 ml of phosphate-buffered-saline-based uptake solution at 37°C. The following radiolabeled amino acid tracers were used: D-[1-¹⁴C]-alanine, L-[1-¹⁴C]-alanine, D-[1-¹⁴C]-serine, L-[1-¹⁴C]-serine, D-[1-¹⁴C]-methionine, L-[1-¹⁴C]-methionine, D-[1-¹⁴C]-phenylalanine, L-[1-¹⁴C]-phenylalanine, D-[1-¹⁴C]-leucine, L-[1-¹⁴C]-leucine, D-[1-¹⁴C]-valine, L-[1-¹⁴C]-valine, D-[1-¹⁴C]-tyrosine, L-[1-¹⁴C]-tyrosine, D-[1-¹⁴C]-glutamic acid, L-[1-¹⁴C]-glutamic acid, D-[1-¹⁴C]-lysine, L-[1-¹⁴C]-lysine, D-[1-¹⁴C]-arginine and L-[1-¹⁴C]-arginine. We tested the inhibitory effects of the following compounds (1.0 mM) on transport: 2-(methylamino) isobutyric acid (a specific inhibitor of system A, in Na⁺-containing uptake solution) and 2-amino-bicyclo[2,2,1]heptane-2-carboxylic acid (a specific inhibitor of system L, in Na⁺-free uptake solution).

Results: D-[1-¹⁴C]-methionine, D-[1-¹⁴C]-phenylalanine and D-[1-¹⁴C]-tyrosine accumulated mainly via system L. D-[1-¹⁴C]-alanine and D-[1-¹⁴C]-serine accumulated primarily via system ASC. High uptake of D-[1-¹⁴C]-alanine, D-[1-¹⁴C]-methionine, D-[1-¹⁴C]-phenylalanine and D-[1-¹⁴C]-leucine was observed. The uptake of radiolabeled serine, valine, tyrosine, glutamic acid and arginine into CHO-K1 was highly stereoselective for L-isomers.

Conclusions: We observed high uptake of D-[1-¹⁴C]-alanine via system ASC (most likely alanine–serine–cysteine-selective amino acid transporter-1) and high uptake of D-[1-¹⁴C]-methionine and D-[1-¹⁴C]-phenylalanine via system L (most likely L-type amino acid transporter-1).

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1. Introduction

With the exception of glycine, all natural amino acids have two optical isomers: L and D. After injection into

animals, ¹⁴C-labeled and ¹²⁵I-labeled D-amino acids localize preferentially in tumors and the pancreas [1,2]. Previous studies have used PET to image D-[1-¹¹C]-methionine [3], D-[1-¹¹C]-tyrosine [4] and D-[1-¹¹C]-phenylalanine [5] in tumors and the pancreas. It is not yet known why D-amino acids accumulate in tumors and the pancreas, but it is likely that membrane transport plays a very important role.

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With the exception of certain mechanisms in earthworms, silkworms [6,7] and a few bacteria [8], all known biological mechanisms involve only L-amino acids. However, due to recent advances in amino acid analysis, several D-amino acids have been detected in high concentrations in the bodies of mammals [9]. Free D-amino acids have been found in various tissues, including the brain; their presence suggests that they perform physiological functions [10–12]. The presence of D-amino acids in mammalian tissue is itself a major finding that overturns previously accepted biochemical theories. D-Amino acids have been found in various tissues, and studies indicate that they are involved in various functions, but numerous issues about D-amino acids remain unclear, including their membrane transport.

Membrane transport is a first step in metabolic pathways of D-amino acids. Chinese hamster ovary (CHO-K1) cells are easy to culture and are widely used in genetic and biochemical studies [13]. CHO-K1 cells have been used as host cells for transient or stable expression of genes [14]. This suggests that these cells could be useful tools for the study of metabolism of D-amino acids and the possibility of using D-amino acids as molecular imaging agents, as well as studying the roles of D-amino acids in biological functions.

In the present study, to examine transport of D-amino acids, we performed uptake experiments using ^{14}C -labeled D-amino acids and CHO-K1, whose transport of L-amino acids has been studied in detail by Shotwell et al. and Bass et al. [15–17]. Those studies have revealed that CHO-K1 has at least three cellular uptake mechanisms for neutral amino acids: transport system L, transport system A and transport system ASC. Transport systems A and ASC are Na^+ -dependent systems and can be differentiated from each other based on pH dependence and specific inhibition by artificial amino acids. System A is specifically inhibited by 2-(methylamino)isobutyric acid (MeAIB), whereas system ASC is not inhibited by MeAIB. Transport system L is not Na^+ dependent and is specifically inhibited by 2-aminobicyclo[2,2,1]heptane-2-carboxylic acid (BCH) [16,17].

Recent molecular biology studies of amino acid transport have revealed much about solute carrier (SLC) genes, which are the genes encoding proteins that transport substances found in liquids [18–20]. These genes are classified into various SLC families according to the classification system of the Human Gene Nomenclature Committee. Genes encoding amino acid transporters belong to the SLC1, SLC3, SLC6, SLC7, SLC16, SLC25, SLC38 and SLC43 families. Neutral amino acid transporters belong to the SLC1, SLC6, SLC7, SLC16, SLC38 and SLC43 families. SLC1, SLC7 and SLC38 are particularly important transporter families in CHO-K1 cells.

In the present study, we examined the transporter isoforms involved in D-amino acid transport, based on the findings of recent molecular biology studies involving CHO-K1. The results for the D-amino acids were compared with those of the corresponding L-amino acids. To the best of our

knowledge, this is the first report of the characterization of transport of D-amino acids in CHO-K1, which is a useful cell line for biomedical studies of gene expression and metabolism. The fundamental data acquired in such studies should be useful for the development of molecular imaging agents based on radiolabeled D-amino acids.

2. Materials and methods

2.1. Chemicals and labeled compounds

Reagent-grade chemicals (Aldrich Chemical Co., Milwaukee, WI, USA) were used in this experiment. We purchased the following D-amino acids from American Radiolabeled Chemicals Co. (St. Louis, MO, USA), along with their L- ^{14}C -isomers: D-[^{14}C]-alanine (D- ^{14}C -ALA), D-[^{14}C]-serine (D- ^{14}C -SER), D-[^{14}C]-methionine (D- ^{14}C -MET), D-[^{14}C]-phenylalanine (D- ^{14}C -PHE), D-[^{14}C]-leucine (D- ^{14}C -LEU), D-[^{14}C]-valine (D- ^{14}C -VAL), D-[^{14}C]-tyrosine (D- ^{14}C -TYR), D-[^{14}C]-glutamic acid (D- ^{14}C -GLU), D-[^{14}C]-lysine (D- ^{14}C -LYS) and D-[^{14}C]-arginine (D- ^{14}C -ARG). Specific activity ranged from 1.85 to 2.22 GBq/mmol.

2.2. Cell culture

CHO-K1 cells were purchased from Riken Cell Bank (Cat. No. RCB0285, Tsukuba, Japan). CHO-K1 cells (10^5 cells/ml) were incubated in 6-cm dishes (Cat. No. 150288, Nalge Nunc International, Roskilde, Denmark) with 5 ml of Dulbecco's modified Eagle's medium (Cat. No. D5796, Sigma Chemical Co., St. Louis, MO, USA), which contained 10% fetal calf serum, at 37°C in a 10% CO_2 atmosphere (pH 7.4). A cell proliferation curve was prepared to determine the duration of the logarithmic growth phase.

2.3. Uptake experiments with CHO-K1 cells

To identify the transport systems involved in uptake of various neutral D-amino acids and to assess the contributions of each transport system, we investigated intracellular uptake of ^{14}C -labeled D-amino acids with or without Na^+ , using artificial amino acids and a modification of the method of Shotwell et al. [16].

When cells had been cultured to a density of about 80% of confluence, the medium was replaced with 5 ml of phosphate-buffered saline (PBS) containing Na^+ (37°C , pH 7.4). The cells were then incubated at 37°C for 10 min. Next, the PBS was replaced with 2.0 ml of a solution containing a ^{14}C -labeled D-amino acid and 1.0 mM of an amino acid transport system inhibitor: MeAIB (a specific inhibitor of system A) or BCH (a specific inhibitor of system L). The cells were then incubated for 10 min. Next, the medium was removed, and the cells were washed twice using 5.0 ml of cold PBS and were then lysed using 2.0 ml of NaOH (0.1 N). A liquid scintillator cocktail (Clear-sol II; Nacalai Tesque, Kyoto, Japan) was mixed with 200 μl of the resulting lysate, and this mixture was placed in a scintillation vial. A liquid scintillation counter (LS-6500; Beckman Instruments, Fullerton, CA, USA) was used

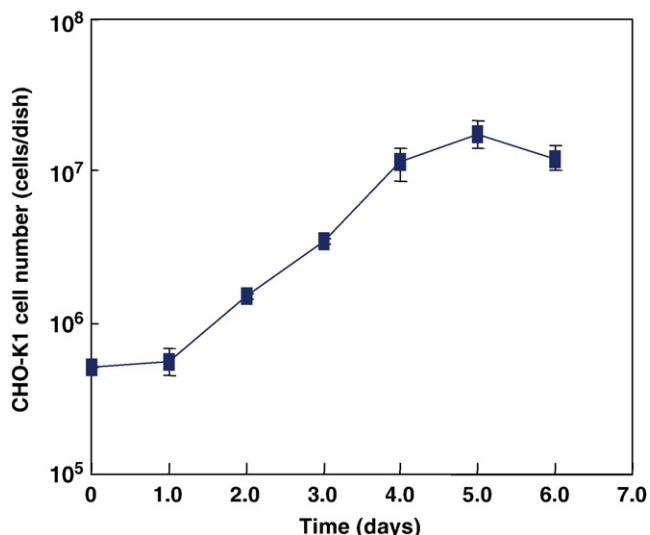


Fig. 1. Growth curve of CHO-K1. The logarithmic growth phase is for 1 to 4 days after inoculation. It took 5 days to reach confluence.

to measure radioactivity. The experiment was repeated using PBS without Na⁺. The result for each set of experimental conditions was the mean of five dishes.

3. Results

Fig. 1 shows the growth curve of CHO-K1. For the uptake experiments, we used CHO-K1 cells in the logarithmic growth phase, 4 days after inoculation.

The highest uptake was observed for D-¹⁴C-ALA (about 30% of the administered dose), followed by D-¹⁴C-PHE (25%). Approximately 15% of the administered dose of D-¹⁴C-MET and D-¹⁴C-LEU was taken up by CHO-K1 cells. The uptake of D-¹⁴C-TYR was about 5% of the administered

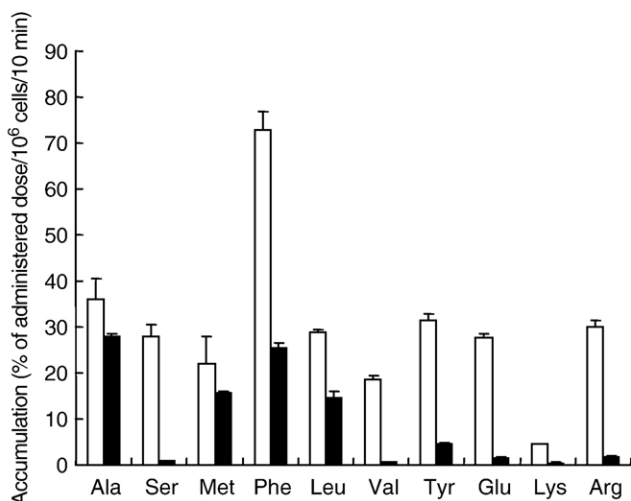


Fig. 2. Percent uptake of L-[¹⁴C]-amino acids (open column) and D-[¹⁴C]-amino acids (filled column) into CHO-K1 cells. Each column represents mean±S.D. of five CHO-K1 monolayers. All differences between L-isomer and D-isomer were significant ($P<.005$).

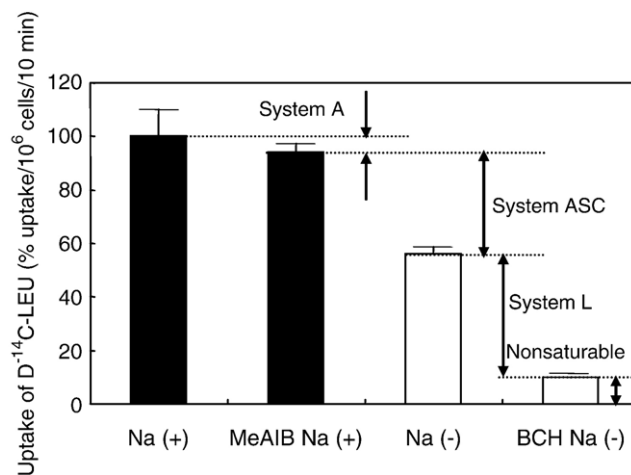


Fig. 3. Contributions of amino acid transport systems to total uptake of D-¹⁴C-LEU into CHO-K1 cells. The total uptake of D-¹⁴C-LEU was divided into the fractions mediated by different transport systems. System A contributed the portion of the total uptake that was inhibited by MeAIB. System ASC and other systems contributed the portion of the Na⁺-dependent uptake that was not inhibited by MeAIB. System L contributed the portion of the Na⁺-independent uptake that was inhibited by BCH. Nonsaturable systems contributed the portion of the Na⁺-independent uptake that was not inhibited by BCH. Each column represents mean±S.D. of five CHO-K1 monolayers.

dose. Very little of the administered dose of D-¹⁴C-SER, D-¹⁴C-VAL, D-¹⁴C-GLU, D-¹⁴C-LYS and D-¹⁴C-ARG was taken up by CHO-K1 cells (Fig. 2).

Fig. 3 shows the contribution of each transport system to the total uptake of each radiolabeled amino acid. The main contributor to uptake of D-¹⁴C-ALA was system ASC. The main contributor to uptake of D-¹⁴C-PHE and D-¹⁴C-MET was system L (Tables 1 and 2).

For all of the tested amino acids, the uptake into CHO-K1 was stereoselective in favor of the L-enantiomer. The following stereoselectivity ratios [(uptake of L-amino acid)/(uptake of D-amino acid)] were observed: alanine, 1.3; serine, 31.0; methionine, 1.4; phenylalanine, 2.9;

Table 1
Contributions of systems A, ASC and L to transport of D-[¹⁴C]-amino acids into CHO-K1

D-Isomer	System A	System ASC	System L	Nonsaturable
D- ¹⁴ C-ALA	15.39±7.27	71.88±4.73	10.78±0.51	1.9±0.20
D- ¹⁴ C-SER	7.80±7.47	72.64±6.05	6.01±4.63	14±1.75
D- ¹⁴ C-MET	4.48±5.98	0.00±11.98	95.36±10.75	9.3±0.50
D- ¹⁴ C-PHE	11.28±11.86	4.67±9.61	78.14±3.79	5.9±0.36
D- ¹⁴ C-LEU	5.69±7.40	38.09±3.49	46.33±2.47	9.9±0.14
D- ¹⁴ C-VAL	0.00±4.98	21.09±14.07	54.60±11.51	28±5.49
D- ¹⁴ C-TYR	10.30±11.17	0.00±16.97	79.68±14.30	17±1.20
D- ¹⁴ C-GLU	7.53±15.62	74.91±9.66 ^a	4.20±4.99	13±2.27
D- ¹⁴ C-LYS	14.36±4.39	21.04±7.78 ^b	12.41±9.97	52±6.26
D- ¹⁴ C-ARG	0.00±10.18	67.74±9.31 ^b	5.62±5.36	27±3.67

Percentage of administered dose/monolayer of noncarrier added D-[¹⁴C]-amino acid; mean±1 S.D. of five monolayers.

^a Acidic amino acid transport systems.

^b Basic amino acid transport systems.

Table 2

Contributions of systems A, ASC and L to transport of L-[1-¹⁴C]-amino acids into CHO-K1

L-Isomer	System A	System ASC	System L	Nonsaturable
L- ¹⁴ C-ALA	5.81±13.17	88.25±1.28	4.00±0.53	1.95±0.36
L- ¹⁴ C-SER	0.00±8.79	93.43±1.86	4.70±0.65	2.02±0.34
L- ¹⁴ C-MET	9.50±6.40	0.00±4.02	84.98±3.22	11.68±0.60
L- ¹⁴ C-PHE	11.56±7.51	22.92±6.42	57.66±3.86	7.85±0.63
L- ¹⁴ C-LEU	0.00±3.73	39.27±4.41	50.87±2.82	15.86±0.19
L- ¹⁴ C-VAL	16.82±7.23	0.00±5.81	78.85±3.33	9.79±0.45
L- ¹⁴ C-TYR	15.08±4.71	13.68±5.17	61.85±4.32	9.39±0.48
L- ¹⁴ C-GLU	9.74±5.07	76.84±4.12 ^a	2.96±1.48	10.46±1.11
L- ¹⁴ C-LYS	2.02±4.46	33.15±5.12 ^b	0.00±4.79	66.03±1.69
L- ¹⁴ C-ARG	0.00±5.93	75.14±3.00 ^b	5.40±4.25	26.16±3.95

Percentage of administered dose/monolayer of noncarrier added L-[1-¹⁴C]-amino acid; mean±1 S.D. of five monolayers.^a Acidic amino acid transport systems.^b Basic amino acid transport systems.

leucine, 2.0; valine, 28.9; tyrosine, 6.9; glutamic acid, 17.9; lysine, 11.2; arginine, 17.2. The highest stereoselectivity was observed for uptake of serine and valine. The lowest stereoselectivity was observed for uptake of alanine and methionine.

4. Discussion

Understanding membrane transport of D-amino acids and their roles in biological functions is a gateway to exploring the possibility of using them as molecular imaging agents.

Na⁺-dependent transporters belong to the SLC1 family and consist of the alanine–serine–cysteine (asc)-selective transporter family that transports neutral amino acids and other groups of transporters for acidic amino acids. CHO-K1 cells contain cloned ASCT1/SLC1A4 complementary DNA (cDNA) [21]. System ASC is a transport system resembling the asc-selective sodium-independent transporter family (asc, asc-type amino acid transporter family). However, asc is Na⁺ independent and belongs to the SLC7 family. The SLC7 family is further divided into two classes: the L-type amino acid transporter (LAT) family and the cationic amino acid transporter family [20]. The LAT family consists of eight proteins that form a heterodimeric functional complex together with an additional protein, the heavy chain of 4F2 antigen (CD98; 4F2hc/SLC3A2). Of the eight known amino acid transporters, the most important transporter in CHO-K1 is thought to be L-type amino acid transporter-1 (LAT1; SLC7A), which is a major transporter of system L. Four LAT isoforms have been cloned: LAT1 and LAT2 (belonging to the SLC7 family) as well as LAT3 and LAT4 (belonging to the SLC43 family). System L activity in CHO-K1 has been examined at the cell biology level in experiments using the inhibitor BCH. Recent studies, using RT-PCR, have examined the expression of LAT1 and 4F2hc at the molecular level [22,23]. SLC38 is a Na⁺-dependent transporter family. SLC38 contains the alanine-selective

transporter family (system A) and other transporters (glutamate transporters).

In the present study, D-¹⁴C-ALA and D-¹⁴C-SER exhibited system ASC-selective transport. D-¹⁴C-ALA exhibited high uptake, but D-¹⁴C-SER exhibited low uptake. The uptake of serine via system ASC was highly stereoselective for L-¹⁴C-SER. It is unclear which isoform of system ASC transported D-¹⁴C-ALA. Two isoforms of system ASC have been identified: ASCT1 and ASCT2. D-Alanine appears to have a strong inhibitory effect on L-alanine transport via ASCT1 [24]. D-Alanine has a weak inhibitory effect on L-alanine transport via ASCT2 [25]. Thus, it seems likely that the present transport of D-¹⁴C-ALA occurred via ASCT1.

In the present study, D-¹⁴C-MET, D-¹⁴C-PHE and D-¹⁴C-TYR exhibited system L-selective transport. D-¹⁴C-MET, D-¹⁴C-PHE and D-¹⁴C-LEU exhibited high uptake, but D-¹⁴C-TYR exhibited low uptake. The uptake of tyrosine via system L was highly stereoselective for L-¹⁴C-TYR. Previous reports indicate that D-¹⁴C-MET, D-¹⁴C-PHE and D-¹⁴C-LEU are transported via LAT1, which is an isoform of system L [13]. Studies using expression cloning of LAT1 in *Xenopus* oocytes indicate that transport of serine, valine, tyrosine and arginine is highly stereoselective for the L-isomer and that transport of methionine, phenylalanine and leucine is not stereoselective [13]. Makrides et al. [26] recently reported preferred transport of O-(2-[¹⁸F]fluoroethyl)-D-tyrosine into the porcine brain in comparison to L-isomer, in addition to considerable differences in stereoselective amino acid transport at the blood–brain barrier among various species.

In the present study, system A exhibited lower transport activity than systems ASC and L. The greatest contribution of system A to transport of an amino acid was for D-¹⁴C-ALA, 15% of whose total uptake was due to system A; 14% of the uptake of D-¹⁴C-LYS was due to system A. Recent studies have examined functional expression of system A in *Xenopus* oocytes injected with mRNA from CHO-K1 cells [27]. To the best of our knowledge, homologues of cDNA of SLC38A1, A2, A4 and A6 have not yet been cloned in CHO-K1 cells.

In the present study, D-¹⁴C-ALA, D-¹⁴C-MET, D-¹⁴C-PHE and D-¹⁴C-LEU exhibited high uptake. Free L-alanine is one of the largest pools of natural amino acids in the human body; free L-methionine, L-phenylalanine and L-leucine are present in smaller pools [28]. It appears that there is no direct relationship between the rate of uptake of a free amino acid and the size of its pool. Moreover, it appears that the affinity of a transporter for a free amino acid is a more important factor in uptake of that free amino acid than the size of its pool. We anticipate that these issues will be clarified by future studies using D-amino acids.

In general, stereoselectivity of amino acid transport is well documented, especially at epithelial barriers such as those of the blood–brain barrier, renal proximal tubule and placenta epithelial cells. CHO-K1 is an epithelial-like cell

line derived from ovary tissue. In the present study, uptake of serine, valine, tyrosine, glutamic acid and arginine into CHO-K1 was highly stereoselective for the L-isomers. It is unclear whether endogenous D-¹⁴C-SER and isomerase are present in CHO-K1 cells. Blood-to-brain transfer of D-¹⁴C-SER and D-¹⁴C-TYR at the blood–brain barrier occurs at extremely low levels [29], and it is assumed that D-serine in the brain is generated by isomerization of L-serine [30,31]. In an investigation of the differential cerebral uptake of the D- and L-isomers of a PET tracer, Langen et al. [32] concluded that in rat and human brains, D-*cis*-4-[¹⁸F]fluoroproline and D-[³H]proline are preferably transported at the blood–brain barrier in comparison with their L-isomers and are isomerized to the L-form. Therefore, D-proline in plasma might be a source of intracerebral L-proline.

The present results suggest that in CHO-K1 cells, acidic amino acid transporters contribute to the uptake of D-¹⁴C-GLU and that basic amino acid transporters contribute to the uptake of D-¹⁴C-LYS and D-¹⁴C-ARG. These amino acids exhibited Na⁺-dependent uptake that was not inhibited by MeAIB.

The biodistribution of a functional molecular imaging agent should be such that it reflects tissue-specific metabolic or physiological function. Fig. 4 shows the general metabolic pathway of amino acids. The main amino acid metabolic pathway consists of membrane transport, the free amino acid pool and protein synthesis. Membrane transport and protein incorporation or other functions are important targets of functional imaging with labeled L-amino acids. The available evidence about D-amino acids suggests that future studies will reveal relationships between diseases and the biological functions and activities of D-amino acids, including their membrane transport, racemization and catabolism.

D-Amino acids are much less common than L-amino acids but are widely distributed in most organisms [33]. Recent studies suggest that in vivo distribution of D-amino acids is related to their catabolism and possible neurochemical functions. For example, D-serine and serine racemase are present in the vertebrate retina and contribute to the physiological activation of N-methyl-D-aspartate receptors [34]. D-Alanine is catabolized by D-amino acid oxidase in

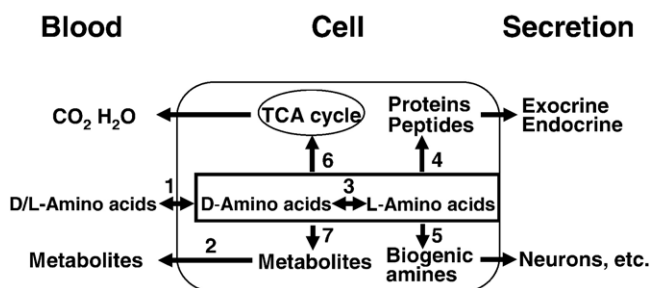


Fig. 4. General metabolic relationship of free amino acids. (1) Membrane transport, (2) racemization, (3) free D-/L-amino acid pool, (4) protein synthesis, (5) biogenic amine synthesis, (6) energy-producing metabolism and (7) catabolism. TCA cycle, tricarboic acid cycle.

mouse tissues including the kidneys, liver, brain, leucocytes and heart [35,36]. Nagata et al. [37] detected substantial amounts of D-serine, D-alanine and D-proline in plasma from patients with renal disease and in tissues from mutant mice lacking D-amino acid oxidase [35, 38]. Studies indicate that in humans, free D-amino acids exist in substantial amounts in various tissues, including the brain, during the growth and development stages, suggesting that they perform physiological functions. However, much remains unknown about the biological functions of D-amino acids, including their roles in brain function and diseases.

In addition, there have been interesting findings in studies of D-amino acids in proteins. D-Amino acids are present in proteins in tooth enamel [39], dentin [40], cerebral white matter [41] and the ocular lens of old animals and humans [42]. It has been reported that in the brains of deceased Alzheimer's patients, the ratio of D-amino acids in β -amyloid protein or tau protein was higher than that of healthy individuals [43], suggesting that D-amino acids are related to age-related diseases such as cataracts and dementia [39,44]. It appears that D-amino acids are not incorporated directly into protein during translation. Jilek et al. [45] recently reported cloning the cDNA of an isomerase that catalyzes posttranslational L–D-isomerization.

For more than 25 years, ¹¹C-labeled amino acids have been used for PET imaging, especially for tumor imaging using L-isomers (e.g., [*S*-methyl-¹¹C]-L-methionine) [3]. There has been much less use of labeled D-amino acids for imaging because D-amino acids are much less common in biological systems than are L-amino acids. Given how little is known about them, ¹¹C-labeled natural D-amino acids and their derivatives are interesting compounds for study. Studies of parent D-amino acids are generally essential, comparing transport characteristics between the natural parent amino acids and their corresponding derived amino acids, to clarify whether amino acids labeled with the relatively long-lived ¹²³I or ¹⁸F exhibit different transport characteristics than their natural parent amino acids.

We believe that clarification of the roles of D-amino acids may lead to the revision of common biological theories and the development of new diagnostic molecular imaging agents for use in studying biosystems involving D-amino acids. We plan to examine the relationship between the potential of D-amino acids as molecular imaging agents and their biological functions in disease, by performing experiments in which genes are introduced into CHO-K1.

5. Conclusions

We studied the transport of D-[¹⁴C]-amino acids, using CHO-K1 cells. CHO-K1 cells accumulated neutral D-[¹⁴C]-amino acids mainly via transport system L (D-¹⁴C-MET, D-¹⁴C-PHE and D-¹⁴C-TYR) and system ASC (D-¹⁴C-ALA and D-¹⁴C-SER). Although it was previously thought that D-amino acids have low affinity for biological systems,

we found that uptake of D-[1-¹⁴C]-amino acids (especially D-¹⁴C-ALA, D-¹⁴C-MET, D-¹⁴C-PHE and D-¹⁴C-LEU) by CHO-K1 cells was comparable to uptake of the corresponding L-amino acids. Uptake of serine, valine, tyrosine, glutamic acid and arginine into CHO-K1 was highly stereoselective for the L-isomers. To develop D-amino acids as diagnostic molecular imaging agents, there is a need for studies of specific D-amino acids in which genes are introduced into CHO-K1.

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