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Sodium-coupled amino acid transport in renal tubule

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More than 99% of nearly all amino acids filtered by the kidneys of humans and other mammals are reabsorbed in the renal tubule and returned to plasma [1, 2]. In man, glycine, histidine and taurine are exceptional in that they have urinary fractional excretions of 3.5%, 6% and 6%, respectively [2]. Considerable evidence exists that amino acids are reabsorbed predominantly in the pars convoluta of the proximal tubule and to a small extent in pars recta [1–4]. Micropuncture studies [2, 3] have shown that no significant reabsorption of amino acids occurs under normal conditions in nephron segments beyond the proximal tubule. However, as documented by Silbernagl [5], the loop of Henle may participate in amino acid resorption under conditions of an elevated filtered load.

Micropuncture experiments [3, 5–7] and studies utilizing isolated perfused proximal tubules [8, 9] have clearly demonstrated that amino acids are primarily reabsorbed from tubular lumen by an active uphill transport across the apical membrane followed by a downhill efflux across the peritubular membrane. Fox et al [10] first showed that amino acid uptake by renal tissue was greatly diminished in the absence of Na^+ in the incubation medium. A stop-flow micropuncture experiment by Ullrich and coworkers [6] showed that the transepithelial concentration difference of actively transported amino acids was abolished when Na^+ was removed from the peritubular and luminal perfusates. Subsequently the Na^+ dependence of amino acid transport across the renal tubular epithelium has been clearly established in multiple animal studies using isolated perfused tubules [8, 9], electrophysiological techniques [11–13], cultured renal epithelial cell lines [14, 15] and isolated brush border membrane vesicles [16–25]. In the latter experiments the rate of accumulation of amino acids by vesicles and the magnitude of the "overshoot", which indicates active concentrative transport, were markedly augmented by a Na^+ gradient across the vesicle membrane. Hence, it is widely accepted that uptake of most amino acids at the brush border surface occurs by sodium-amino acid cotransport driven by both the concentration and the voltage components [26], of the electrochemical Na^+ gradient from tubular lumen to cell [4, 27–29]. The energy maintaining the Na^+ gradient is established by the Na^+-K^+-ATPase which is located at the basolateral membrane and pumps Na^+ out and K^+ into the cell. In concert with this notion is the finding that ouabain, a specific inhibitor of Na^+-K^+-ATPase which dissipates the electrochemical Na^+ gradient across the cell membrane, diminishes amino acid uptake by renal epithelial cell line ($LLC\text{-}PK_1$) [14]. Na^+-amino acid symport, like the cotransport of Na^+ with other solutes, has been termed "secondary active transport" because its energy is derived from the electrochemical gradient of Na^+ rather than from direct coupling to a metabolic process [27–29]. The dependence of tubular amino acid transport on Na^+ gradient has been further demonstrated by the observation of decreased Na^+-linked accumulation of various amino acids into gramcidin- [22, 23, 25] and papain- [30] treated vesicles. Both compounds stimulate the entry of Na^+ into vesicle, via pathways not coupled to uptake of amino acids, thereby rapidly collapsing the Na^+ gradient necessary for amino acid transport. In the case of cystine, however, the findings that its Na^+-dependent uptake by vesicles did not display an overshoot [31] and that its accumulation in papain exposed vesicles was enhanced, rather than diminished [32], has led Hsu, Corcoran and Segal [32] to speculate that tubular cystine transport, unlike transport of other amino acids, is more closely coupled to Na^+ flux across the luminal membrane. Further studies are needed to substantiate this conclusion.

Kinetics of transport

Na^+-amino acid symport across the luminal membrane is a carrier-mediated saturable process obeying Michaelis-Menten Kinetics [2, 27, 28]. Kinetics of transport have been determined for various amino acids in multiple micropuncture, microperfusion and vesicle studies [2, 27, 28, 33]. A wide range of values of the apparent affinity constants (Km) and transport capacities (V_{max}) have been reported [2, 33]. The effectiveness of Na^+-dependent reabsorption process for a specific amino acid depends on the ratio V_{max}/Km [2, 7]. A low such ratio for taurine (low V_{max}), as well as for glycine and histidine (high Km values) may partially account for the observed high fractional excretion of these amino acids in man.

In analyzing the data obtained from studies using brush border membrane vesicles, for various amino acids including glycine [18, 34], cystine [31], arginine [35], glutamic acid [20] and glutamine [20], two or more Na^+-linked transport systems with different kinetic characteristics have been described. The same phenomenon is observed in microperfusion and micropuncture experiments [2, 8, 36]. The evaluation of kinetic parameters and their physiological significance, particularly when determined in vesicle experiments, have been largely questioned because of the known limitations of vesicle transport studies [37, 38] including heterogeneity in vesicle size, origin and sideness, binding of the substrate to membranes, leak permeabilities, and finally the difficulty controlling transmem-

branal pH and voltage gradients. However, the demonstration of multiple transport systems for the same amino acid becomes more meaningful if the reduced concentrations of filtered amino acids presented to the proximal straight tubule following their high reabsorption rate in the proximal convoluted tubule is considered. Thus, in the case of glycine, for example, two Na^+-dependent active transport systems have been demonstrated along the luminal membrane of the isolated perfused proximal tubule [8]; a low affinity (Km—11.8 mM), high capacity (J_{max}—28.5 pmol/min/mm) system in the convoluted segment, and a high affinity (Km—0.7 mM) low capacity (J_{max}—2.5 pmol/min/mm) system in the straight segment. The latter system, which is also operating in parallel to a lower apical membrane backflux permeability in the proximal straight tubule [27], absorbs lesser amounts of glycine against a greater concentration gradient and probably permits the reduction of the luminal glycine concentrations to lower levels than could be achieved in the proximal convoluted tubule [8, 27]. This axial heterogeneity of Na^+-linked amino acid uptake systems with respect to kinetic characteristics has been recently demonstrated for L-serine [39], L-phenylalanine [40] and L-proline [41] in brush border membrane vesicles derived from pars convoluta and pars recta of the proximal tubule. This pattern, however, may not hold for all amino acids. A progressive increase, rather than decrease, in Km of L-histidine transport along the proximal convoluted tubule has been reported [27, 42]. In addition, a microperfusion experiment [36] showed two transport systems for L-proline with different affinities and capacities, which were located in the proximal convolution thereby operating in parallel.

The effect of Na^+ concentration on kinetics of amino acid uptake has been extensively studied, and in most instances an increase in Na^+ concentration decreased Km of transport but did not alter V_{max} [16, 17, 23, 25].

Electrogenicity of transport

The electrogenic positive nature of Na^+-linked amino acid transport, which is favored by a negative intracellular potential, has been clearly established for most amino acids [2, 29, 43, 44]. Electrophysiological micropuncture experiments on rat proximal tubule showed that intraluminal application of various amino acids in the presence of Na^+ depolarized the luminal membrane, indicating a net transfer of a positive charge across the membrane [11, 13, 43]. In the absence of Na^+ no significant potential changes were observed. In studies using brush border membrane vesicles, markedly enhanced Na^+ gradient-dependent amino acid accumulation was demonstrated upon imposition of an inside negative potential using the K^+ ionophore valinomycin in the presence of an outwardly-directed K^+ gradient, or the proton ionophore, carbonyl cyanide P-trifluoromethoxyphenylhydrazone (FCCP), in the presence of an outwardly-directed H^+ gradient or by the use of highly permeant anions in the external medium [16, 19, 22, 23, 25, 39]. Moreover, the imposition of an inside negative voltage accelerated Na^+-linked amino acid uptake by vesicles even in presence of Na^+-equilibrated conditions [16, 26, 45, 46].

In the case of basic amino acids, which carry a net positive charge at physiological pH, disagreement exists whether their uptake into brush border membrane vesicles is driven by the total electrochemical Na^+ gradient or by membrane potential alone [12, 19, 44, 47]. While electrophysiological experiments showed that basic amino acids were reabsorbed and depolarized the luminal membrane only in the presence of luminal Na^+ [12], most vesicle studies demonstrated Na^+-independent L-arginine [35, 47, 48] and L-lysine [45, 49] uptake. In the latter experiments an interior negative potential induced accumulation of basic amino acids into vesicle, even in the complete absence of Na^+. A reasonable explanation for this data, as originally proposed by Fox et al [10] and later by Hammerman [47], is that the terminal cation moiety of dibasic amino acids could substitute for Na^+ at the binding site on the carrier protein. According to this hypothesis the Na^+-dependent and Na^+-independent transport of dibasic amino acids is accomplished by a single transport system which, depending on the availability of Na^+ is driven by the total Na^+ electrochemical gradient or by membrane voltage alone [44, 47].

An electroneutral transport system for acidic amino acids has been demonstrated by direct potential measurements in the microperfused newt proximal tubule [50] and, for Na^+- and K^+-dependent glutamate uptake, in rabbit renal brush border membrane vesicles [51, 52]. These findings, however, are in contradiction to electrophysiological experiments demonstrating a Na^+-dependent depolarization of rat tubular epithelial cells in the presence of L-glutamate and L-aspartate in the lumen [13], and to a positive electrogenic K^+-dependent Na^+-glutamate cotransport observed in rat renal brush border membrane vesicles [53]. Species differences may underly these discrepancies.

Stoichiometry and dependence on other ions

Investigations into the stoichiometry of Na^+-linked amino acid transport have shown that one or more sodium ions are transported per amino acid molecule translocated [29, 46, 54, 55]. In general, Na^+-solute stoichiometry determines the electrogenicity of the transport system and it determines the maximal concentration gradient against which solute transport can occur, namely the efficiency of the system [29, 44]. In accordance with the latter notion was the demonstration of two distinct proximal tubular transport systems for D-glucose, a low affinity system with 1:1 Na^+:hexose coupling ratio and a high affinity system with 2:1 Na^+:hexose stoichiometry, in brush border membrane vesicle derived from the convoluted segment and the straight segment, respectively [56, 57]. A Na^+/L-phenylalanine stoichiometry of 1:1, however, for both a low affinity system located in the pars convoluta and a high affinity system situated in the pars recta were recently found in a vesicle study [40]. Further studies are needed to elucidate whether higher Na^+-amino acid stoichiometries may account for the more efficient amino acid transport systems located in the pars recta of the proximal tubule.

The involvement of additional ions besides Na^+ in the carrier complex translocating amino acids across the luminal membrane has been extensively studied [29, 44, 58]. Studies using brush border membrane vesicles demonstrated that Na^+ gradient-driven L-glutamate or L-aspartate uptake was stimulated by an outwardly-directed K^+ gradient [24, 52, 53, 58] (Fig. 1). Furthermore, as shown by Murer and coworkers, in the rat kidney [53, 58], but not supported by Sacktor and his colleagues in the rabbit kidney [24, 52], L-glutamate accumulation into vesicles was potential sensitive only in the presence of intrave-

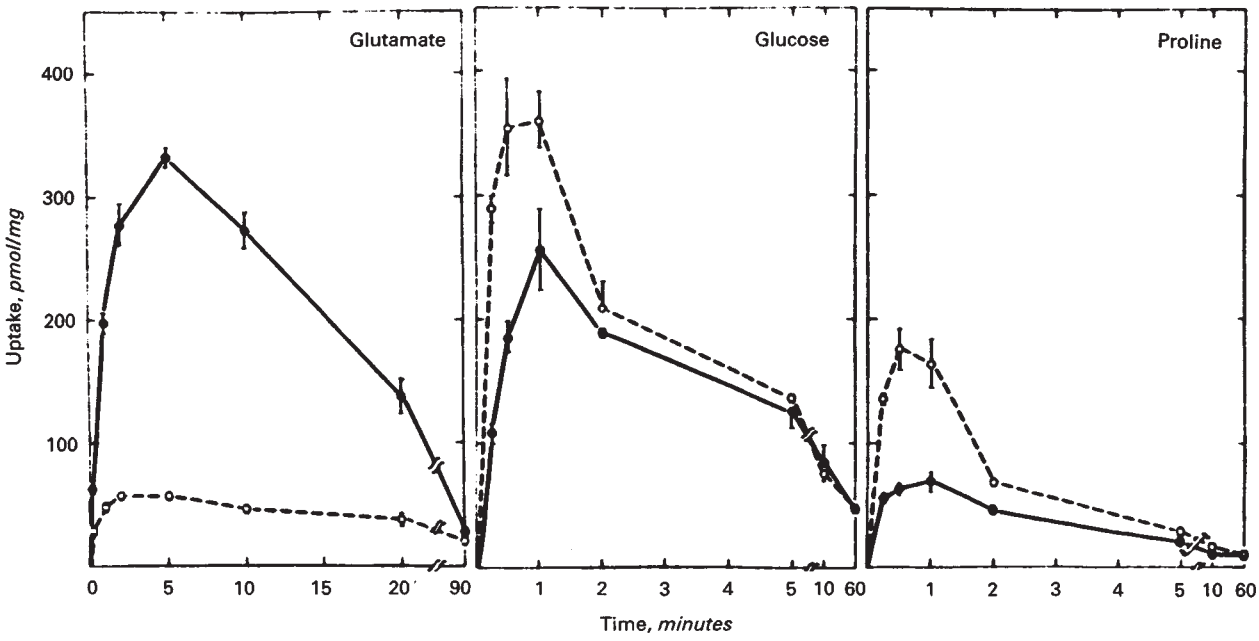

Fig. 1. *The effects of intravesicular K^+ on the Na^+ gradient-dependent uptakes of L-glutamate, D-glucose and L-proline.* Symbols are: (○) Na^+ out, mannitol in, (●) Na^+ out, K^+ in. [From (24) with permission].

sicular K^+. The observations that an outwardly-directed K^+ gradient could drive L-glutamate uptake against a concentration gradient even under Na^+-equilibrated conditions [24, 53] and that an inwardly-directed K^+ gradient stimulated Na^+-dependent L-glutamate efflux, [58, 59] supported the contention of K^+ countertransport simultaneous with Na^+-glutamate translocation. Recently, however, Nelson et al [60] and Fukuhara and Turner [55] provided evidence supporting a model in which rather than a carrier-mediated simultaneous exchange of Na^+ and glutamate for K^+, sodium and potassium act at different steps in the transport system; Na^+ facilitates the translocation of the glutamate carrier complex and K^+ acts to facilitate the recycling of the unloaded carrier (Fig. 2). The exact nature of this complex translocation process awaits further studies.

For the positive electrogenic process operating in the rat kidney a stoichiometry of $3Na^+$:1 glutamate for influx coupled to the efflux of $1K^+$ has been suggested [53, 58]. For the electroneutral transport system in the rabbit kidney, on the other hand, a symport of $2Na^+$:1 glutamate and countertransport of $1K^+$ has been postulated [24, 59].

A requirement for H^+ ion has been reported for the uptake of various amino acids by renal brush border membrane vesicles [49, 60–62]. Glutamate accumulation into vesicles was stimulated by low external pH in both, the presence and the absence of Na^+-gradient or K^+-gradient driving forces, and was inhibited by low internal pH [60]. In addition, evidence was provided that H^+ may compete with K^+ for its binding site on the transporter [60]. Hence, as suggested by Nelson et al [60], H^+ may be cotransported with glutamate and, in the absence of internal K^+, H^+ can also fullfill the requirement for a countertransported ion (Fig. 2). The stoichiometry consistent with these data for an electroneutral process would be a symport of $1Na^+$:$1H^+$:1 glutamate and antiport of $1K^+$ [24, 60]. Recent studies demonstrated an enhancement of L-proline [61] and glycine [62] accumulation by vesicles in the presence of an inwardly-directed H^+ gradient. The H^+ gradient-stimulated uptake of both amino acids was electrogenic, occurred even in the absence of Na^+ and, in the case of L-proline, the enhancement of transport was mediated by an increase in V_{max}, but no change in Km [61, 62]. While the stimulation of L-proline accumulation by a pH gradient was additive with that produced by Na^+ [61], the glycine uptake data suggested a competitive interaction between Na^+ and H^+ for the transport of this amino acid [62]. Finally, Stieger et al [49] showed that an outwardly-directed H^+ gradient stimulated L-lysine uptake by vesicles, a finding which led the authors to postulate the existence of a proton-lysine antiport system. Clearly, further studies are needed to clarify the characteristics of tubular H^+-linked amino acid transport and to explore the interactions between Na^+ and H^+ during the translocation process.

Recent studies have focused on the anion dependence of tubular Na^+-amino acid symport. It has been shown that Na^+-dependent uptake of glycine [34], taurine [54, 63] and β alanine [46] into renal brush border membrane vesicles, is enhanced by an inwardly-directed Cl^- gradient. Br^-, unlike the other halides, fully [54] or partially [46] substituted for Cl^- in supporting the full overshoot of β amino acid accumulation. External Cl^- enhanced Na^+-linked uptake of glycine and β amino acids significantly more than the highly permeant anions SCN^- and NO_3^-, known to stimulate electrogenic positive transport processes [34, 63, 46]. Furthermore, β alanine uptake could be driven against a concentration gradient by a chloride gradient alone [46], providing strong evidence for a cotransport of Cl^- with the Na^+-β amino acid carrier complex across the luminal membrane. Taurine [54] and β alanine [46] uptake data suggested a stoichiometry of two or more Na^+:$1Cl^-$:1 β amino acid (Fig. 3), consistent with the electrogenic positive nature of this transport system.

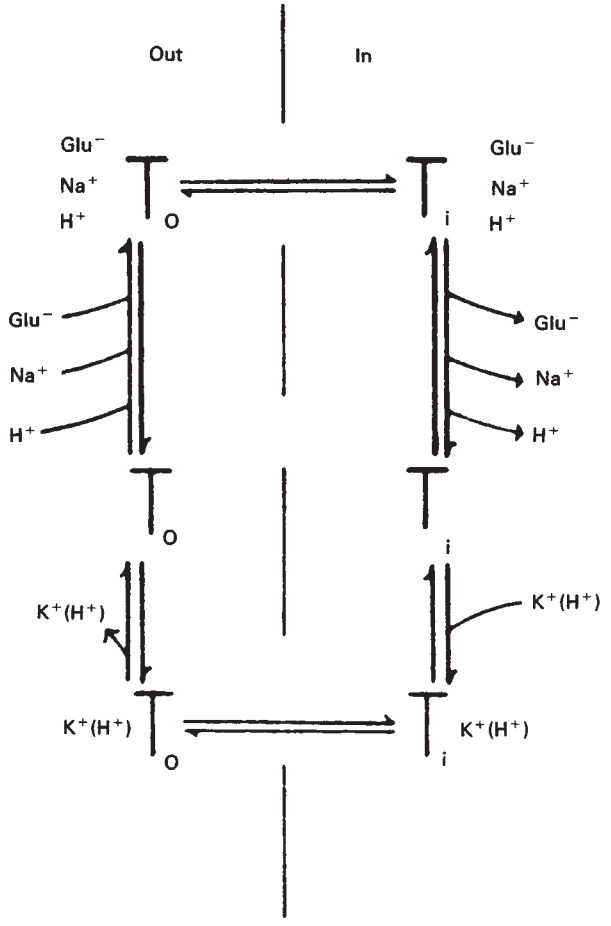

Fig. 2. *Possible mechanism for glutamate transport.* Ti and To represent two forms of the transporter with substrate binding sites exposed to the internal and external surface of the membrane, respectively. Interconversion of the two forms occurs either with Na^+, H^+, and glutamate bound (upper pathway) or with a cation (usually K^+ but possibly also H^+) bound (lower pathway). [From (60) with permission].

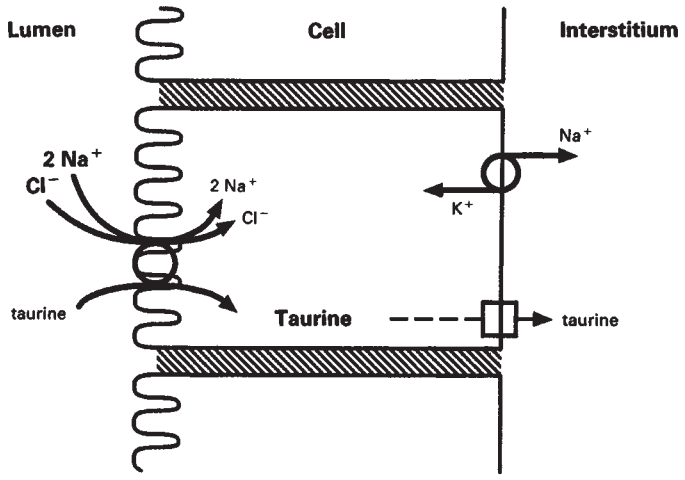

Fig. 3. *A schematic model for taurine transport across rat renal proximal tubular cell.* Modified from [54], with permission.

Specificity of transport

At least seven distinct group-specific Na^+-dependent transport systems for amino acids have been identified in the tubular luminal membrane [2, 26, 28]. Evidence for these systems is derived from a variety of microperfusion experiments, vesicle studies and in humans, from the existence of inborn errors of renal tubular transport which can only be explained by defects in specific transport pathways [28, 64]. These include systems for: 1) dibasic amino acids, 2) acidic amino acids, 3) "imino" acids, 4) glycine, 5) β- and γ-amino acids, 6) all other neutral amino acids, and 7) cystine/cysteine. Another characteristic of Na^+-linked tubular amino acid transport is its stereospecificity, because in general L-amino acids are transported to a much higher extent than D-isomers [2, 28].

There is good evidence that L-cystine and the dibasic amino acids L-lysine, L-arginine and L-ornithine share a common transport system [31, 65–67], as also suggested by the urinary hyperexcretion of all four amino acids observed in classic systinuria [68]. However, from the two Na^+-linked transport systems which have been identified for cystine, namely a high affinity-low Km system and a low affinity-high Km system, only the first appears to interact with dibasic amino acids [31, 65, 66]. Vesicle studies [69] and in vivo microperfusion experiments [70] showed that transport of L-cystine, but not L-cysteine, can also proceed via Na^+-dependent transport pathways for neutral amino acids. L-cysteine is probably reabsorbed by a separate and specific transport system [67, 70]. The investigation of the cystine/cysteine system and its interaction with other transporters has been complicated by the extremely low solubility of cystine, the rapid reduction of cystine to cysteine during or after the translocation process [66, 67], and the significant binding of cystine to vesicle membrane [31, 32]. Thus, the exact nature of this transporter remains unclear.

A Na^+-dependent transport system for glutamine, separate from the acidic amino acid transport mechanism, exists in the tubular luminal membrane [7, 20, 71]. However, the two pathways may be linked via the activity of the brush border enzyme γ glutamyltransferase which may hydrolyze glutamine to glutamate intraluminally, particularly during acidosis [71].

There is controversy in the literature whether glycine and L-proline share a transport system or have independent pathways [18, 44, 72, 73]. McNamara et al [18, 72] using brush borders membrane vesicles demonstrated dual transport systems for both glycine and proline, of which the low Km system for proline and the high Km system for glycine appeared to be shared, as shown by uptake competition studies. Vesicle studies by Hammerman and Sacktor [23, 73], however, demonstrated that both amino acids had separate, single high-affinity transport systems, and using the method of accelerated exchange diffusion (enhanced transmembrane flux of one amino acid induced by a second amino acid in "trans" position), the authors found the two systems to be independent. The discrepancy between the reports may be explained by the different methods used to explore the interaction between the transport pathways. As pointed out by Schafer et al [27, 44], great caution is needed in interpreting data from uptake competition studies because cotransport of one amino acid, or even other solute [74], with Na^+ across the membrane may collapse the electrochemical Na^+ gradient necessary for Na^+-dependent flux of the other amino acid.

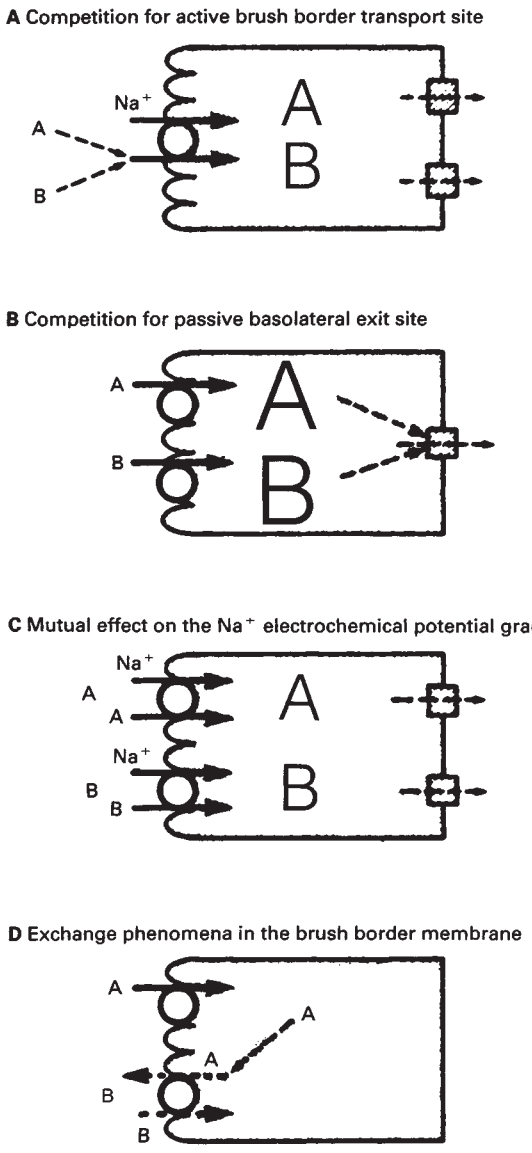

Fig. 4. *Potential mechanisms whereby absorption of one amino acid* (**A**) *may be reduced by the presence of a second amino acid* (**B**) *in lumen.* A. Competition for a common transport site. B. Competition for a common basolateral exit site. **C.** Competition on the Na^+ electrochemical gradient. **D.** Facilitated exchange diffusion of B for A. The resulting reduction in intracellular concentration of A reduces net transcellular absorption of this amino acid. [From (27) with permission].

Whatever the mechanism of the interaction between the various amino acids at the luminal membrane surface may be, whether competition for an active transport site (Fig. 4A) mutual effect on the Na^+ electrochemical gradient (Fig. 4C) or facilitated, carrier mediated, exchange diffusion (Fig. 4D), all these processes may be of great relevance in determining the patterns of various hereditary aminoacidurias in man, as reviewed elsewhere [2, 27].

The investigation of amino acid transport pathways using various tissue culture cells including kidney epithelial cell lines has delineated three major transport systems for neutral amino acids, namely Na^+-dependent, concentrative A and ASC systems and Na^+-independent, nonconcentrative L system [14, 15, 75, 76]. However, the exact nature of these pathways, their localization, (whether in the apical or the basolateral membrane) and the degree of correlation with the better defined tubular amino acid transport mechanisms found in vesicle and microperfusion studies is still uncertain [75, 76]. A recently characterized Na^+-dependent, PGE_1-responsive "G system" with broad amino acid specificity, in the apical membrane of kidney epithelial cell line, MDCK, may represent an energy-dependent transport route for amino acid reabsorption [15].

Basolateral membrane transport

The basolateral membrane plays a major role in tubular amino acid transport not only because it controls amino acid downhill efflux from the epithelial cell but also because it harbors Na^+-linked transport systems mediating amino acid uptake in the cell [27, 29, 44].

Studies using basolateral membrane vesicles isolated from renal cortex showed that the exit of neutral amino acids such as L-phenylalanine [16], L-serine [39] and L-proline [77] across the peritubular membrane is a saturable, Na^+-independent process probably occurring via facilitated diffusion. Thus, a polarity of the cortical proximal tubular cell with respect to Na^+-neutral amino acid cotransport is evident, a conclusion also supported by electrophysiological experiments [11].

Na^+-coupled active basolateral transport mechanisms have been identified for various amino acids. Studies using isolated perfused proximal tubules demonstrated active peritubular uptake of glycine [9] glutamine [78] and L-cystine [79] in the straight segment, which in the case of glycine and glutamine were Na^+-dependent. This mechanism may be of great physiological importance because it may provide the epithelial cell of the straight proximal tubule with essential nutrients in a nephron segment where the apical membrane transport of amino acids is markedly limited by their very low intraluminal concentrations [44]. An inwardly-directed Na^+ gradient and an outwardly-directed K^+ gradient stimulated an overshoot of glutamate accumulation by isolated renal basolateral membrane vesicles [80]. In addition, electrophysiological experiments [13] demonstrated Na^+-dependent depolarization of the basolateral membrane induced by peritubular application of acidic amino acids. As suggested by Sacktor et al [80], active accumulation of glutamate at both the luminal and the antiluminal sides of the tubular epithelial cell may provide the mechanism by which very high concentrations of this metabolically important amino acid are achieved in the renal tubule (Fig. 5). This notion applies also for glutamine, the major precursor of renal ammonia, for which Na^+-dependent transport pathways have been demonstrated in the luminal [7, 20], as well as in the basolateral [81, 82] membrane.

Amino acid transport across the peritubular membrane is potentially of major pathophysiological importance in humans. Competition between amino acids for a basolateral facilitating exit mechanism (Fig. 4B) or active Na^+-coupled uptake of amino acids across the basolateral membrane, combined with increased backflux across the luminal membrane, may result in net amino acid secretion and may underly some of the congenital aminoacidurias, as previously reviewed [2, 27, 64].

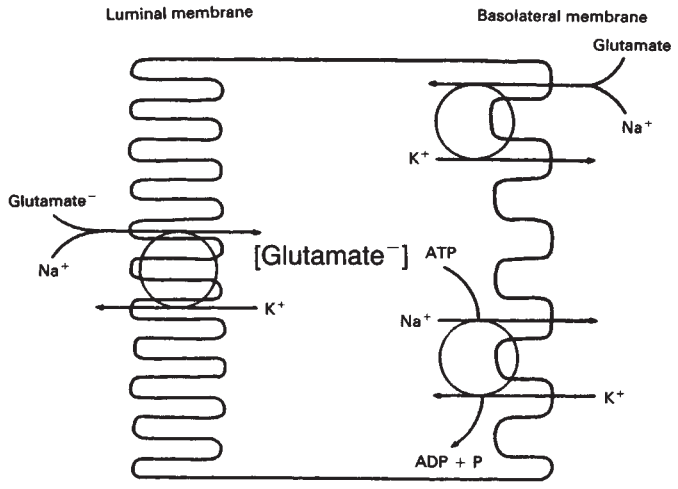

Fig. 5. *A schematic model for L-glutamate uptake by the renal tubular cell.* [From (80) with permission].

Adaptive responses

Reabsorption of amino acids in the proximal tubule increases during periods of reduced amino acid intake and decrease with dietary excess [22, 83, 84]. This renal adaptive response to diet is expressed at the tubular luminal membrane surface as evidenced by a greater Na^+-dependent accumulation of taurine into isolated renal brush border membrane vesicle prepared from rats fed low taurine diet [22]. Diminished uptake is found in vesicles from high taurine diet fed animals. This up-regulation and down-regulation of symport activity in response to diet is associated with a change in V_{max} of transport rather than in Km [22, 83]. The addition of β-alanine, a competitive inhibitor of Na^+-dependent taurine transport, to the drinking water of rats fed low, normal, and high taurine diets resulted in increased taurine uptake by brush border membrane vesicles in each group [85]. Since β-alanine reduced taurine content of renal cortex without changing plasma taurine values, it has been concluded that the renal adaptive response expressed at the brush border surface relates to changes in renal cortex taurine concentrations [85]. The cellular mechanism mediating this response remains to be established.

Noteworthy is the carrier-mediated, Na^+- and Cl^--dependent taurine transport in the renal peritubular membrane of various marine fish [86, 87] known to secrete taurine in their urine during exposure to fresh water in conjunction with cell volume regulation.

The effect of acidosis on Na^+-linked tubular glutamine transport has been thoroughly investigated [7, 88–90]. Controversy exists concerning the existence of such an adaptation at the luminal membrane surface. While brush border membrane vesicles isolated from chronically acidotic rats exhibited an enhanced Na^+-dependent glutamine accumulation [71, 88], and lowering the pH of the medium further increased glutamine uptake [88], no change in luminal membrane glutamine transport in response to acute acidosis was observed in micropuncture studies [7], in the isolated perfused proximal tubule [78] and in vesicle studies [89]. Indeed, an adaptation at the brush border surface seems unreasonable considering the very efficient reabsorption of filtered glutamine already present in the proximal convoluted tubule [7, 90]. The apparent adaptation observed in vesicle studies [71, 85] probably reflects an extravesicular γ-glutamyltranferase-mediated conversion to glutamate and subsequent accumulation [71, 90]. On the other hand, acute [78, 89] and chronic [91] acidosis stimulated Na^+-dependent glutamine uptake by basolateral membrane vesicles, and acute acidification of the peritubular medium enhanced antiluminal Na^+-linked glutamine uptake in the isolated perfused proximal tubule [78]. Thus, augmented Na^+-dependent glutamine transport across the basolateral membrane during acidosis may represent an adaptive mechanism which provides the tubular cell with increased amounts of substrate for ammoniagenesis.

Ontogeny of transport

Neonatal "physiological aminoaciduria" is characteristic of all mammalian species including man, and it reflects the immaturity of the renal tubular transport system [92]. The ontogeny of sodium-dependent tubular amino acid transport process has been the subject of recent studies [93–96]. A gradual age-related increase in Na^+-coupled uptake of taurine [93] and proline [96] by rat renal brush border membrane vesicles and of cystine [95] by isolated dog renal cortical tubules has been documented. While in the case of taurine [93] and cystine [95] this maturational process involved a change in V_{max} of uptake, the maturation of proline transport [96] was associated with a decrease in Km. Furthermore, using nursing rats suckling milk with low, normal and high taurine content from dams fed each of the respective diets, it was found that the renal brush border membrane isolated from the suckling 7-day-old rats did not exhibit the adaptive response to diet which existed in older rats [93, 94]. Other studies, however, using kidney cortical slices and isolated tubules from rats and dogs demonstrated increased intracellular levels of various amino acids in conjunction with slower efflux from the cell in immature animals [97–99]. Thus neonatal aminoaciduria may reflect a combination of reduced uptake at the luminal surface and an efflux block at the peritubular surface.

Recently, alterations in phospholipid composition have been documented during the process of rat tubular brush border membrane maturation [100], suggesting that changes in fluidity of the membrane may account for the observed maturational changes in Na^+-linked tubular amino acid transport. Of great importance are recent studies showing an increased permeability to Na^+ [96] and an enhanced amiloride sensitive, Na^+/H^+ exchange activity [101] in neonatal rat renal brush border membrane vesicles. This increased luminal membrane Na^+/H^+ antiport, coupled with a diminished Na^+-K^+-ATPase activity known to exist in the basolateral membrane of the neonatal proximal tubular epithelium [102], may result in a rapid dissipation of the electrochemical Na^+ gradient necessary for Na^+-amino acid cotransport, thereby contributing to the aminoaciduria of early life [96, 101] (Fig. 6).

Summary

Amino acids are reabsorbed from the tubular lumen by a saturable, carrier-mediated, concentrative transport mechanism driven by a Na^+ electrochemical gradient across the luminal membrane. This process is followed by efflux mainly via carrier-mediated, Na^+-independent facilitated diffusion across the basolateral membrane. Individual amino acids may

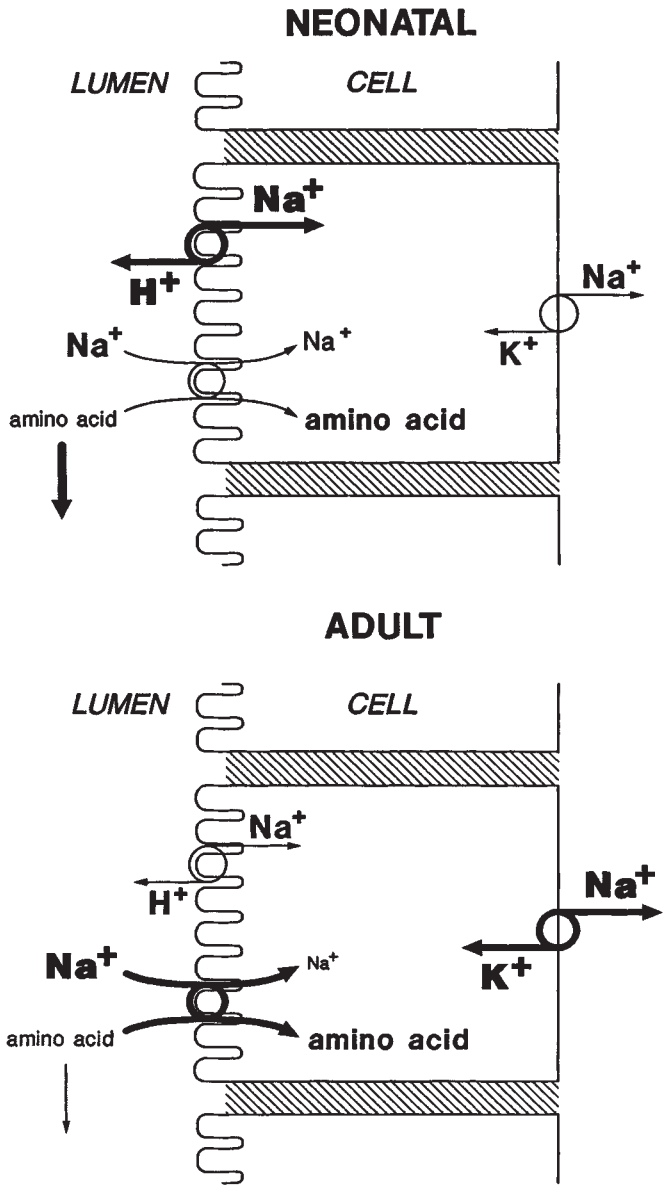

Fig. 6. *A hypothetical model for the pathophysiology of neonatal aminoaciduria.*

have two or more Na^+-dependent transport systems with different kinetic characteristics along the luminal membrane of the proximal tubule, thereby enabling very efficient amino acid reabsorption. Dual Na^+-coupled transport pathways for some amino acids located in both the luminal and the peritubular membranes may operate in concert to provide the tubular epithelial cell with essential nutrients. One or more Na^+ ions, H^+, Cl^- and in the case of acidic amino acids, K^+ ion, may be involved in the translocation of the carrier complex. For most amino acids this process is electrogenic positive, favored by a negative cell interior. At least seven distinct, but largely interacting, Na^+-dependent amino acid transport systems have been identified in the brush border membrane. A diet-induced adaptation in Na^+-coupled taurine transport and acidosis-induced adaptive response in Na^+-dependent glutamine transport are expressed at the luminal and the basolateral membrane surfaces, respectively. The aminoaciduria of early life may be related to a rapid dissipation of the Na^+ electrochemical gradient necessary for amino acid reabsorption.

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