Posttranscriptional regulation of the proximal tubule NaPi-II transporter in response to PTH and dietary Pi

HEINI MURER, IAN FORSTER, NATI HERNANDO, GEORG LAMBERT, MARTIN TRAEBERT, AND JÜRG BIBER
Institute of Physiology, University of Zurich, CH-8057 Zurich, Switzerland

Murer, Heini, Ian Forster, Nati Hernando, Georg Lambert, Martin Traebert, and Jürg Biber. Posttranscriptional regulation of the proximal tubule NaPi-II transporter in response to PTH and dietary Pi. Am. J. Physiol. 277 (Renal Physiol. 46): F676–F684, 1999.—The rate of proximal tubular reabsorption of phosphate (Pi) is a major determinant of Pi homeostasis. Deviations of the extracellular concentration of Pi are corrected by many factors that control the activity of Na-Pi cotransport across the apical membrane. In this review, we describe the regulation of proximal tubule Pi reabsorption via one particular Na-Pi cotransporter (the type IIa cotransporter) by parathyroid hormone (PTH) and dietary phosphate intake. Available data indicate that both factors determine the net amount of type IIa protein residing in the apical membrane. The resulting change in transport capacity is a function of both the rate of cotransporter insertion and internalization. The latter process is most likely regulated by PTH and dietary Pi and is considered irreversible since internalized type IIa Na-Pi cotransporters are subsequently routed to the lysosomes for degradation.

phosphate; reabsorption; parathyroid hormone; sodium-phosphate co-transport

RENAL HANDLING of inorganic phosphate (Pi) is of primary importance to control the extracellular concentration of Pi. Under normal physiological conditions, ~80% of Pi contained in the primary urine is reabsorbed along the proximal tubules. However, under conditions of deviated concentrations of Pi in the plasma, the rate of proximal tubule Pi reabsorption is adjusted rapidly by various hormonal regulatory circuits to achieve a correct Pi homeostasis (3, 6). So far, our knowledge about a possible handling of Pi in the distal segments of the nephron is rather limited.

In proximal tubules, Pi reabsorption is initiated by a sodium-dependent transport step(s) located in the luminal (brush border) membrane. Functionally, this transport step has been thoroughly characterized in studies using isolated brush-border membrane vesicles and in vitro and in vivo microperfusions (3, 55). On the basis of the knowledge accumulated over the last few years about the identity of Na-Pi cotransport proteins, much has been learned about the molecular basis of proximal tubular apical Na-Pi cotransport and about the mechanisms involved in the regulation of this transport step, which is most crucial for Pi homeostasis. Most physiological and pathophysiological situations resulting in an impaired renal Pi handling are due to an altered rate of Na-Pi cotransport through the proximal tubular apical membrane. This is reflected by changes of the maximum transport velocity (Vmax). It became evident that changes in the Vmax value can be explained by changes in the total amount of Na-Pi cotransporters (specifically of the type IIa Na-Pi cotransporter) residing in the brush-border membrane. Although a full explanation is beyond the scope of this article, it should be mentioned that proximal tubular Pi reabsorption can be influenced additionally by the luminal pH (36) and the membrane potential (21).

In this review, we will briefly describe the chief players responsible for proximal tubular Pi reabsorption (the NaPi protein family). Then we will discuss the phenomenology and the molecular mechanisms (known and speculative) involved in alterations of proximal tubule Pi reabsorption, such as by parathyroid hormone (PTH) and acute changes of the dietary Pi content. In the present context, we will concentrate on posttranscriptional mechanisms, since it appears that transcriptional mechanisms are not involved in the rapid adjustment of proximal tubule Pi reabsorption.

Na-Pi Cotransporters Expressed in the Proximal Cell

Mammalian Na-Pi cotransporters cloned to date fall into three dissimilar families (type I, type II, and type III; see Table 1). Each type has been identified in a number of mammals (for review, see Ref. 72), and some have also been identified in fish (74) and Xenopus laevis...
Members of all types are expressed in proximal tubule cells. The current picture of the cellular location of type I, type II, and type III Na-Pi cotransporters is shown in Fig. 1. As illustrated, type I as well as type II cotransporters are located in the apical membrane, whereas type III cotransporters are most likely located at the basolateral membrane. As demonstrated in a recent study, in targeted type IIa knockout mice, Na-Pi cotransport in brush-border membrane vesicles was reduced by 70%, indicating that most of proximal tubular Pi reabsorption is via the type IIa cotransporter (2). However, it is not known via which pathway(s) the remaining 30% is transported; one candidate would be the type I cotransporter or an as yet unidentified Na-Pi cotransporter or both.

Various routes have been suggested via which Pi exits the cell at the basolateral site (see Fig. 1). None of the indicated Na-independent transporters has been identified. Interestingly, Na-dependent Pi transport at the basolateral membrane seems also to occur, most likely via the type III cotransporter. The broad expression pattern of type III transcripts, however, suggests a housekeeping role of type III cotransporters rather than a role in the regulation of proximal tubule Pi reabsorption.

Table 1. Na-Pi cotransporter protein families

<table>
<thead>
<tr>
<th>Family Name</th>
<th>Type I</th>
<th>Type II</th>
<th>Type IIa</th>
<th>Type IIb</th>
<th>Type III</th>
</tr>
</thead>
<tbody>
<tr>
<td>Molecule name</td>
<td>NaPi-I/rabbit, rat, mouse or human</td>
<td>NaPi-I/IIa/mouse, rat, human, rabbit or opossum</td>
<td>NaPi-IIb/mouse, human, flounder or Xenopus</td>
<td>Glvr-1 (Pit-1) Ram-1 (Pit-2)</td>
<td>human, mouse, rat 679, 656</td>
</tr>
<tr>
<td>Amino acids</td>
<td>=465</td>
<td>=640</td>
<td>=690</td>
<td>Na-Pi cotransport, electronegic, pH dependent</td>
<td>Na-Pi cotransport, electronegic</td>
</tr>
<tr>
<td>Function (in Xenopus oocytes)</td>
<td>Na-Pi cotransport, Cl channel activity, interaction with organic anions</td>
<td>Na-Pi cotransport, electronegic, pH dependent</td>
<td></td>
<td>Na-Pi cotransport, electronegic</td>
<td></td>
</tr>
<tr>
<td>Tissue expression (mRNA and/or protein)</td>
<td>Kidney cortex/PT, liver (brain)</td>
<td>Kidney cortex/PT</td>
<td>Small intestine, lung and other tissues</td>
<td>Ubiquitous</td>
<td></td>
</tr>
<tr>
<td>Regulated by PTH/Pi diet?</td>
<td>no</td>
<td>yes</td>
<td>yes</td>
<td>yes</td>
<td></td>
</tr>
</tbody>
</table>

PTH, parathyroid hormone; PT, proximal tubule.
sible role in glucose metabolism of type I Na-P$_i$ cotransporters.

Type II Na-P$_i$ cotransporter. A type II Na-P$_i$ cotransporter was first identified in cDNA libraries of rat and human kidney cortex tissue (47). Subsequently this protein was identified in a variety of other species and notably also in cultured opossum kidney cells (OK cells) (66). A similar protein (type IIb) has been described that is not expressed in mammalian kidney but in small intestine and other tissues (31). The subclassification of type IIa and type IIb (see Table 1) is based on differences of COOH-terminal amino acid sequences, although at the functional level they display essentially the same characteristics.

In mammals, expression of type IIa cotransporter mRNA is restricted to the proximal tubules as shown by in situ hybridization (65) and RT-PCR (15), although expression of type II-related mRNA in other tissues such as bone (25) cannot be excluded. By immunohistochemistry, it has been established that the type IIa cotransporter is located exclusively in the apical membrane of proximal tubular cells and uniformly distributed along the microvilli (15, 69). Thereby it was realized that the relative amount between the segments S1, S2, and S3 and also between the proximal tubules of superficial and juxtamedullary nephrons was dependent on the physiological/pathophysiological state. Under normal conditions, the type IIa protein is predominantly expressed in S1 segments of juxtamedullary nephrons.

The central role of the type IIa cotransporter in renal handling of P$_i$ became evident from the following observations: 1) Na-P$_i$ cotransport of isolated renal brush-border membranes isolated from Npt2 knockouts was reduced by $\sim 70\%$, indicating that most of the proximal tubular P$_i$ reabsorption is via the type IIa cotransporter (2); 2) intravenous injection of antisense oligonucleotides led to a reduced rate of proximal tubular Na-P$_i$ cotransport that was paralleled by a reduced amount of type IIa cotransporters (57); and 3) several studies performed under different physiological and pathophysiological conditions demonstrated that altered rates of Na-P$_i$ cotransport correlated with an altered amount of type IIa cotransporters in the apical membrane (53).

A possible model of the secondary structure of the type IIa Na-P$_i$ cotransporter is shown in Fig. 2. This model predicts that type IIa cotransporters exhibit eight transmembrane segments and that both the NH$_2$ and the COOH termini are located intracellularly. Two additional hydrophobic domains are positioned such that they may penetrate the membrane only partly.

---

**Fig. 2.** Putative model of the secondary structure of the rat type IIa protein. As described in the text, the location of some parts have been verified, but others remain speculative. The two N-glycosylation sites in the secondary extracellular loop are indicated by asterisks. Cysteine residues likely to form a disulfide bridge and possibly important for function are in black.
around 0.1 mM, the net rate of proximal tubular Pi reabsorption is also being transported together with one H$_2$PO$_4$ ion (23). The transport stoichiometry of three Na ions exhibited an inwardly directed current that was compatible with an electrogenic mode of Na-Pi cotransport suggests that the membrane potential (alteration of whole tissue (68), and evidence was obtained that in the type III cotransporters are found throughout the liver, which suggests a general "housekeeping" role for rather broad including small intestine, bone, heart, and liver, which suggests a general "housekeeping" role for PiT-2 (72).

Thus, on the basis of the observation that up to 70% of Pi is reabsorbed via the type IIa cotransporter, the electrogenic mode of Na-Pi cotransport suggests that the net rate of proximal tubular Pi reabsorption is also influenced by the membrane potential (alteration of driving force).

Type III Na-Pi cotransporter. Receptors for gibbon ape leukemia (Glvr-1) and mouse amphotropic (Ram-1) retroviruses have been shown to exhibit Na-dependent transport of Pi; the two proteins were named PiT-1 and PiT-2 (72).

The expression pattern of PiT-1 and PiT-2 mRNAs is rather broad including small intestine, bone, heart, and liver, which suggests a general "housekeeping" role for type III cotransporters. In mouse kidney, transcripts of type III cotransporters are found throughout the whole tissue (68), and evidence was obtained that in proximal tubules type III cotransporters are located at the basolateral membrane (C. Silve, personal communication). Thus in proximal tubules type III cotransporters most likely do not contribute significantly to Pi reabsorption nor to its acute regulation. It is, however, of interest that in the Hyp mouse model, as well as after a treatment of normal mice with growth hormone, increased abundance of renal Glvr-1 mRNA was reported (68). Furthermore, in cell culture experiments, a transcription-independent increase of PiT-related mRNA due to P$_i$ depletion was described (14).

Regulation of Proximal Tubular Pi Reabsorption by PTH and Dietary Pi Content Is Via the Amount of Type IIa Na-Pi Cotransporters

In addition to other actions, the phosphaturic action of PTH is well documented (3, 6); this action of PTH is assigned to the proximal tubule. Elevated plasma levels of PTH lead to a phosphaturic response that has been shown, on the basis of studies performed with intact tubules and isolated brush-border membranes, to be due to a reduced capacity ($V_{\text{max}}$) of brush border Na-Pi cotransport (3, 6, 54). Direct evidence that the amount of the type IIa cotransporter in the apical membrane is modulated by PTH has been obtained in vivo studies (34, 69) and in vitro studies using OK cells (61, 62) and isolated perfused mouse proximal tubules (H. Voeikl and M. Traebert, unpublished observations). In all these experimental systems, immunohistochemistry in combination with Western blots and transport studies established that the phosphaturic action of PTH can largely be explained by a reduction of the amount of type IIa cotransporters in the microvilli.

Dietary Pi content (ranging from 0.1% to 1% P$_i$) has a marked effect on the net rate of proximal tubule Pi reabsorption due to an altered rate of type IIa-associated apical Na-P$_i$ cotransport. Interestingly, feeding animals with an altered P$_i$ diet leads within a few hours to two- to threefold changes of proximal tubular transport paralleled by changes of the amount of brush-border membrane-associated type IIa cotransporters (upregulation by low-P$_i$ diet and downregulation high-P$_i$ diet). Since under these acute adaptation conditions no change in the abundance of the type IIa transcript has been observed, this kind of regulation is thought to occur at the posttranscriptional level as well (41).

Downregulation of type IIa cotransporters. The decreased rates of proximal Pi reabsorption as observed after PTH infusion or after feeding a diet of high P$_i$ content (acute conditions) can be explained by a decrease in the amount of type IIa cotransporters. Immunohistochemical studies demonstrated that these treatments type IIa cotransporters are transiently accumulated in the so-called subapical compartment (Fig. 3) and are associated with small and large endocytic vacuoles. Internalization of type IIa proteins most likely occurs at the invaginated intermicrovillar regions (dext sites), possibly via clathrin-coated vesicles, since type IIa proteins were found in structures also containing clathrin and the adaptor protein AP2 (69). Furthermore, horseradish peroxidase injected prior to the treatment with PTH was associated with type IIa-containing small and large endocytic vacuoles. This suggests that type IIa cotransporters are internalized via the endocytotic pathway of soluble proteins.

Morphological data available to date suggest that unlike other regulated transporters such as the Glut4 transporter (58) or aquaporin-2 (19), internalized type IIa Na-P$_i$ cotransporters do not reside in an intracellular compartment from which they could be recruited, but are instead routed directly to the lysosomes. Under in vivo conditions, this pathway was described for both inhibitory factors PTH and high-P$_i$ diet (35) (Fig. 4). Similar observations were made also with OK cells (61, 62). In these studies, PTH led to a complete disappearance of type IIa cotransporters, which could be inhibited by the addition of inhibitors of lysosomal degradation such as leupeptin. Thus all data obtained so far suggest that inhibition of proximal Pi reabsorption is via an internalization of type II cotransporters and represents an irreversible process (48).
In summary, the endocytic retrieval of type IIa cotransporters from the apical membrane resembles the pathway as described for the internalization (via clathrin-coated vesicles at the deft sites) and processing (degradation in the lysosomes) of a variety of filtrated proteins/peptides in the proximal tubule cell (13). In contrast to the later constitutive process, internalization of type IIa cotransporters appears to be a regulated event for which we postulate the following mechanism. Type IIa cotransporters localized along the microvilli constantly move laterally along the microvilli and pass freely through the cleft sites. In contrast to soluble proteins, internalization of type IIa cotransporters would then not be an obligatory process but requires signaling, which may be based on a specific interaction(s) with yet unknown protein factors. Such interactions then would be either in an “on mode” (signaled by PTH or acute high-Pi diet) or in an “off mode” (signaled by low-Pi diet).

Upregulation of type IIa Na-Pi cotransporters. The most dramatic increase of proximal tubule Pi reabsorption is observed shortly after a low-Pi diet. Four hours after treatment, expression of type IIa cotransporters increased close to the maximum level that is observed after prolonged (days) adaptation to a low-Pi diet (41, 42, 65). Upregulation in midcortical and superficial nephrons occurs without a parallel increase of the type IIa transcript. This is in agreement with the observation that upregulation by an acute low-Pi diet could not be blocked by cycloheximide (41, 43). However, it has not been shown definitively whether protein synthesis in proximal cells is indeed blocked by intravenously injected cycloheximide.

Studies concerning depletion of PTH have so far been performed mostly in cell culture (OK cells) and indicated that upon removal of PTH the recovery (upregulation) of Na-Pi cotransport was entirely dependent on protein de novo synthesis (48, 61). In chronically parathyroidectomized rats, a specific upregulation of the type IIa protein was apparent in superficial nephrons without a parallel change of the type IIa transcript (34). These findings may point to similar (in vivo) mecha-
nisms independent of protein synthesis as discussed for the acute adaptation to low-Pi diet.

Mechanistic Views

Involvement of microtubules. Microtubules play a fundamental role in maintaining polarity and apical endocytosis of proximal cells. For example, disruption of the microtubular network by colchicine was reported to block the uptake of horseradish peroxidase almost completely and induced a redistribution of apical membrane proteins (7, 20).

In contrast to the described dependence of the endocytosis of soluble proteins on an intact microtubular network (20), internalization of type IIa cotransporters was not impaired by a colchicine-induced disruption of microtubules. In vivo, after disruption of the microtubules by colchicine, both PTH and an acute high-Pi diet was not impaired by a colchicine-induced disruption of microtubules. In vivo, after disruption of the microtubules by colchicine, both PTH and an acute high-Pi diet led to a similar transient accumulation of type IIa cotransporters in the subapical compartment as observed in control animals (41, 45, 46). Similarly, studies on OK cells also demonstrated that disruption of the microtubules did not impair inhibition of Na-Pi cotransport by PTH (26). On the other hand, it appeared that an intact microtubular network was necessary for the subsequent lysosomal sorting (46).

In contrast to the internalization process, an intact microtubular network is necessary for upregulation of type IIa cotransporters when induced, for example, by an acute low-Pi diet (41, 45). This observation is in agreement with the important role of an intact microtubular network for the apical routing of post-Golgi vesicles and for the maintenance of the dense tubular subapical network (20).

Possible signaling pathways. SIGNALING VIA THE PTH RECEPTOR. PTH and PTH-related peptide receptors respectively are localized in both the basolateral and apical membrane of proximal tubule cells (1). The binding of PTH-(1–34) to the PTH receptor leads to an activation of both adenylate cyclase and phospholipase C, with subsequent activation of protein kinases A and C (49). In OK cells, pharmacological activation of either protein kinase A by forskolin or protein kinase C by phorbol esters led to an inhibition of Na-Pi cotransport (54). A cAMP-independent mode of PTH action was suggested recently by the use of PTH-(3–34), which in OK cells inhibited Na-Pi cotransport and provoked internalization of type IIa cotransporters in the absence of an increase of cAMP (60). In oocytes, type IIa-associated Na-Pi cotransport was also reported to be inhibited by an activation of protein kinase C. This was paralleled by an internalization of type IIa proteins, which suggested that the type IIa protein represents a specific target for the protein kinase C pathway (23). However, type IIa-associated Na-Pi cotransport expressed in oocytes was not altered by a pharmacological activation of the cAMP pathway (28). Until now, the target(s) for protein kinases A and/or C activated by PTH remains unknown. One candidate hypothesis proposes that the degree of phosphorylation of type IIa cotransporter is itself important in determining the rate of internalization. Although a number of consen-
sus motifs for protein phosphorylation are contained in the amino acid sequence, mutation of these motifs did not affect inhibition of Na-Pi cotransport in oocytes by phorbol esters (28). In conclusion, the question of whether an alteration of the phosphorylation state of type IIa proteins plays any role in the regulation of proximal tubular Pi reabsorption remains unanswered.

SIGNS INVOLVED IN PHOSPHATE ADAPTATION. Until now, the signal involved in the acute changes of the type II cotransporters upon changes of the dietary Pi content is not known. Since in parathyroidectomized animals the adaptive response was of similar size, a major contribution by PTH was excluded (6, 55).

An asymmetry of the adaptive response has been suggested by experiments on OK cells (64). In cells grown on permeant filters, depletion of Pi at the apical site was sufficient to provoke an adaptive increase of apical Na-Pi cotransport, whereas removal of Pi at the basolateral site was without an effect. This may indicate that the concentration of Pi in the primary urine (reflecting the plasma concentration) as such elicits the signal for the appropriate adjustment of the amount of type IIa cotransporters. In this regard, it is of interest that intravenous injection of inorganic phosphate was sufficient to provoke an inhibition of proximal tubular Na-Pi cotransport (11). It remains to be shown whether in the apical membrane a specific phosphate sensor may exist or whether another sensor, such as the calcium sensor CaR (8), may indirectly signal changes of the tubular Pi concentrations in the primary urine.

UBIQUITINYLATION. In the last years ubiquitinylation has progressively emerged as a signal for downregulation of a variety of membrane proteins, including receptors, and possibly also of membrane transporters such as the Na channel ENaC. Ubiquitinated membrane proteins then are degraded by the proteasomes or the lysosomes or both (30). Although in OK cells proteasomal degradation of internalized type IIa cotransporters has been ruled out by the use of proteasomal inhibitors (61), ubiquitinylation of the type IIa protein can still not be fully excluded. Therefore, it is intriguing to speculate whether ubiquitinylation of type IIa cotransporters may serve as a signal to anchor a yet unknown adaptor-like protein that allows specific internalization of type IIa cotransporters.

PI-3 KINASE. An involvement of phosphatidylinositol-3 kinase (PI-3 kinase) has been described in a variety of membrane trafficking processes (18). A possible role of PI-3 kinase in the PTH-induced internalization of type IIa cotransporters was investigated in OK cells. By two approaches (use of an inhibitor and use of a negative dominant mutant) no evidence for a role of PI-3 kinase in the internalization of type IIa cotransporters was obtained (59). Interestingly, however, in OK cells, wortmannin has been reported to inhibit endocytosis of albumin (9). Thus these observations may provide evidence that the mechanisms involved in the (constitutive) endocytosis of soluble proteins and (regulated) endocytosis of type IIa cotransporters are different.
Motifs

Tyrosine-based motifs such as YXXO (where O is a bulky residue) and dileucine signals have been reported as being important in endocytic processes, for the biosynthetic targeting of lysosomal proteins, and for the basolateral sorting in polarized cells (32, 50). Such a tyrosine-based motif is contained in the sequence of the type IIa cotransporter (Y509RFW, fourth intracellular loop, see Fig. 2). Its possible role in PTH-mediated downregulation has been investigated in oocytes and OK cells using Y-mutated type IIa proteins coupled to the green fluorescent protein (Ref. 29 and unpublished data). These results indicated that the mentioned tyrosine-containing sequence does not represent a major determinant for the internalization of type IIa proteins. It remains to be shown whether other motifs such as LL motifs alone or in combination with tyrosine-based motifs are important.

Lipids

Chronic adaptation (for several days) to different Pi diets was reported to alter the cholesterol and glycosphingolipid content and consequently the membrane fluidity of brush-border membranes (39, 40, 52). Such alterations of the lipid content were not observed after acute adaptations (hours). Thus it seems unlikely that changes in the lipid compositions are involved for the down- or upregulation of type IIa cotransporters provoked by acute changes of the Pi diet. Nevertheless, changes of the lipid composition of microdomains such as in glycolipid-cholesterol rafts may facilitate or inhibit internalization of type IIa cotransporters.

Extracellular Proteolytic Cleavage

Western blot analysis using isolated proximal tubular brush-border membranes revealed that a large part of the type IIa protein exists as a cleaved entity yet structurally stabilized by a disulfide bridge (5, 75). Since proteolytic cleavage likely occurs between the two N-glycosylation sites, the two cysteine residues located in the second extracellular loop may form a disulfide bridge (see Fig. 2). However, it remains to be shown whether cleavage of type IIa cotransporter occurs in vivo or whether this observation reflects a preparation artifact.

With regard to a possible role of the proteolytic cleavage of the type IIa protein in the regulation of Na-Pi cotransport, it is of interest to note that Na-Pi transport in isolated brush-border membrane vesicles could be inhibited by disulfide bridge reducing agents and that the degree of inhibition correlated with the appearance of the cleaved moieties on Western blots (75). Although speculative, this would suggest that an oxidation-reduction mechanism may contribute additionally to the regulation of proximal tubular Na-Pi cotransport.

Associated Proteins

Type IIa cotransporters are localized along the entire microvillus and, upon a stimulus such as by PTH or high-Pi diet, are thought to be internalized at the clefts with an increased rate (see above). Certainly, specific interaction of type IIa cotransporters with other proteins would be required for 1) microvillar positioning and 2) a specific signaling either in the microvilli or in the clefts or both. Currently, the identity of such proposed proteins is not known. A PDZ domain-containing protein has been cloned and demonstrated to influence expression of type IIa Na-Pi cotransport in oocytes (17). Despite a strict proximal tubular and hence apical localization of this protein (unpublished observations), its exact role in either positioning and/or regulating the type IIa cotransporter is not yet established. Remarkably, similar PDZ domain-containing proteins have been described to interact with the sodium/hydrogen exchanger NHE-3 (71, 76).

Summary

Type IIa Na-Pi cotransporters located in the apical membrane contribute up to 70% of proximal tubular Pi reabsorption and represent the major target for physiological (e.g., PTH, Pi diet) and pathophysiological (e.g., X-linked Hyp mice, Ref. 67) regulation. On the basis of current data, we suggest that a change of the number of type IIa cotransporters is achieved by an altered rate of internalization via coated vesicles at the intermicrovillar invaginations. It is of interest that internalized type IIa Na-Pi cotransporters do not recycle but are routed to and degraded in the lysosomes. At the cleft sites an on-and-off switch is postulated that specifically targets type IIa cotransporters to clathrin-coated vesicles. Neither the single components nor the specific signaling reactions of these proposed on-and-off mechanisms are known.

REFERENCES


