Overview of Sodium-Phosphate Cotransporter Family

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Abstract

Inorganic phosphate (Pi) is transported across the plasma membrane by sodium-dependent phosphate transporters belonging to solute carrier families 34 (SLC34) and 20A (SLC20A). SLC34 has been well characterized and studied. Recently, the role of SLC20A in many pathological conditions is emerged. In this review, a brief overview of characteristics of this family will be provided.

Introduction

P_i is transported across the plasma membrane by two solute carrier families 34 (SLC34) and 20A (SLC20). Both of them use electrochemical gradient of Na^+ to transport P_i[1, 2]. SLC34 family has 3 members, including sodium-phosphate cotransporter type IIa (NaPi-IIa), type IIc (NaPi-IIc), which are mainly expressed in the brush border membrane of proximal tubules and type IIb (NaPi-IIb), whose expression primarily is in the GI tract [1-4]. SLC20A is comprised of two members, PiT-1 and PiT-2, which are ubiquitously expressed [2, 4]. The role of SLC34 families in P_i transport has been extensively studied and well-characterized [3, 5, 6]. SLC20A family has been considered as playing a housekeeping role in P_i homeostasis [4]. Recently, however, their roles in vascular calcification [7-9] and in P_i-induced apoptosis in endothelial cells [10], epithelial cells, and podocytes [11] have emerged.

SLC34 family

All three members of SLC34 transport HPO_4^{2-} preferentially and are affected by extracellular pH, which can influence the distribution of $H_2PO_4^-$ or HPO_4^{2-}[2, 3, 12]. A series of dedicated studies using patch clamp technique in *Xenopuslaevis* oocytes by Forster and his group showed that NaPi-IIa/NaPi-IIb are electrogenic and NaPi-IIc is electroneutral with Na:Pi

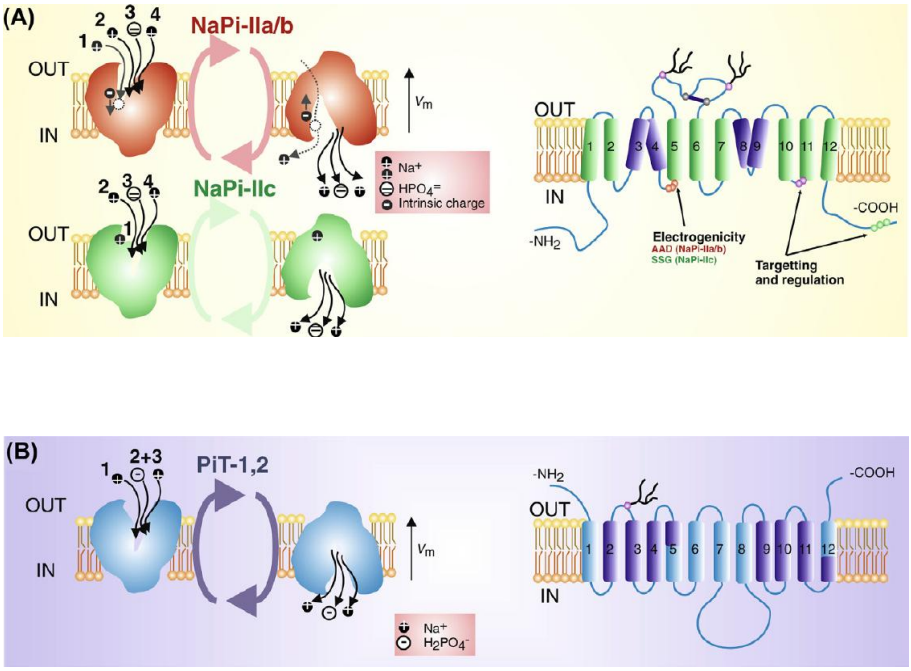
stoichiometry is 3:1 and 2:1, respectively [1, 3, 5, 13] (Table 1). Kinetic studies revealed that 2 first Na^+ bind to the transporters, subsequently, P_i and the 3rd Na^+ binding transition implicated a rate-limiting of carrier [2, 3] (Fig 1A). This family is presumed to be a functional monomer, however, it is also suspected to form dimers or tetramers (for review,[2, 3]). The sequences of these proteins are composed of 599 to 640 amino acids [2, 12] with 12 transmembrane domains. Both C- and N-terminus face the intracellular [2], and the linker region is located in the extracellular side (Fig. 1A).

NaPi-IIa and NaPi-IIc with main expression in the apical membrane of proximal tubules play an important role in P_i reabsorption. *NaPi-IIa*$^{-/-}$ mice or NaPi-IIa/-IIc double KO mice showed significantly low plasma P_i level with high P_i excretion, while *NaPi-IIc*$^{-/-}$ mice did not lower plasma P_i level meaningfully[4]. This data gives an insight that NaPi-IIa maybe chiefly regulates P_i reabsorption in the proximal tubules.

NaPi-IIb expressing in the small intestine determines P_i absorption from daily diet [14]. *NaPi-IIb*$^{-/-}$ mice died at the embryonic stage, whereas heterozygous mice showed lower plasma P_i level and hyperphosphaturia at 4 weeks old [15]. At 20 weeks, these heterozygous mice demonstrated a normophosphatemia with increased abundance of NaPi-IIa and NaPi-IIc[15]. This leads to the assumption that P_i absorbed via NaPi-IIb is critical for survival during embryogenesis.

SLC20A family

This family is comprised of two members, PiT-1 and PiT-2. These two isoforms of SLC20A family were originally discovered as retroviral receptor Glvr-1 (gibbon ape leukemia virus receptor) and Ram-1 (rat amphotropic leukemia virus receptors)[16], then were identified as sodium dependent phosphate transporters[17]. A number of studies showed that P_i transport via SLC20A in *Xenopus* oocytes are electrogenic with stoichiometry 2:1 of Na:Pi and rarely sensitive to pH (Fig 1B) [18-20] and phosphonoformic acid (PFA) [3]. However, measuring surface pH of oocytes that were injected with human PiT-1 plasmids might be affected by solution containing buffer molecules. $H_2PO_4^-$ has been suggested the preferential transport form of PiT-1/2 [2]. The current topology of PiT-1/2 provided 12 transmembrane domains structure with both C- and N-terminus facing extracellular space (Fig. 1B) [2].



(Adopted from Forster IC et al., 2013)

Fig. 1. Mechanism and molecular features of SLC34 and SLC20A

(A) For all SLC34 proteins transmembrane transport for the case of an inwardly directed Na^+ gradient, two Na^+ ions in the extracellular medium bind sequentially, followed by divalent P_i and a 3rd Na^+ ion. Substrate release to the cytosoloccurs after reorientation of the fully loaded carrier. (B) For SLC20 proteins, the transport sequence comprises an initial binding of one Na^+ ion, followed by a random interaction of monovalent P_i and a 2nd Na^+ ion. Reorientation of the loaded carrier then leads to substrate release in the cytosol.

Table 1. Kinetic properties of SLC34 and SLC20 proteins expressed in *Xenopus* oocytes

Family	**SLC34**			**SLC20**
Isoform	NaPi-IIa (SLC34A1)	NaPi-IIb (SLC34A2)	NaPi-IIc (SLC34A3)	PiT-1 (PiT-2) (SLC20A1/A2)
P_i species preferred	HPO_4^{2-}	HPO_4^{2-}	HPO_4^{2-}	$H_2PO_4^-$
Driving cations	Na^+ (Li^+)	Na^+ (Li^+)	Na^+	Na^+, Li^+
Stoichiometry (Na:Pi)	3:1	3:1	2:1	2:1
pH dependence	Strong	Strong	Strong	Weak
$K_{0.5}^{Pi}$ (μM)	50-54	7-250	70-80	24-120
PFA (K_i^{PFA} (mM)	1.05	0.16	0.9	2.71 (4.63)

(Adopted from Forster IC et al., 2012)

Recently, the emerging role of PiT-1 in high P_i conditions has been investigated. P_i uptake through PiT-1 is essential for bone formation and embryogenesis, however, excess P_i uptake via this transporter induces vascular calcification in chronic kidney disease, apoptosis, aging, etc.

Conclusion

The role of SLC20A in many pathological conditions has been investigated, particularly in the elevated Pi in the normal range. Of note, line of evidence showed that in the vascular calcification, PiT-1 and PiT-2 are critical. However, still needs it a lot of studies to figure out the mechanism and the role of this sodium-dependent phosphate cotransporter in this process.

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