K-NPPase Activity along the Rabbit, Guinea Pig, Rat, and Mouse Nephron.

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Ouobain sensitive K dependent p-nitrophenyl phosphatase(K-NPPase) activity along the rabbit, guinea pig, rat, and mouse nephron was determined with the K-phosphatase cytochemical method by Mayahara et al. The pattern of enzyme activity was identical in the four species: with high activity in thick asending limbs and distal convoluted tubules(DCT), intermediate in proximal convoluted tubules(PCT), little or none in prximal straight tubules, collecting ducts, and thin limbs. In all species, K-NPPase activity was higher in medullary thick asending limbs(MAL) than in cortical thick asending limbs (CAL). This finding was especially apparent in the rabbit. The result that K-NPPase activity is higher in DCT than in PCT is in good agreement with biochemical report of Na-K-ATPase activity by Katz et al. However, higher activity in MAL than in CAL in rat and mouse differs from the result by Katz et al. who reported lower Na-K ATPase in MAL in the rat and similar activity in both limbs in the mouse.

Ultractyochemical Localization of Ca^{++}-ATPase Activity in the Mouse Pancreatic Exocrine Cells

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Ultrastructural localization of Ca^{++}-ATPase activity was investigated in the mouse pancreatic exocrine cells by a new one step method of Ando et al. (1981).

It was found that Ca^{++}-ATPase activity was localized in the whole plasma membrane, inner saccules of Golgi apparatus, membranes of zymogen granules, lysosomal membrane and some mitochondrial cristae. The activity observed at the apical membrane was, however, resistant to the EDTA treatment and heating (70°C, 5 min). When ATP was replaced by sod. β-glycerophosphate in the medium, no reaction products were observed, whereas the localization of ALPase activity demonstrated by the lead citrate method (Mayahara et al., 1967) was confined to the basal membrane adjacent to capillaries.

These results indicate that the pancreatic exocrine cells possess Ca^{++}-ATPase activity which is distinct from ALPase.

Na-K-ATPase Localization in the Rat Paw Eccrine Sweat Gland

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Localization of Na-K-ATPase in the rat paw eccrine sweat gland was investigated with the K-NPPase cytochemical method.

Rat paw was fixed in a mixture of 2% paraformaldehyde and 0.5% glutaraldehyde in 30mM PIPES buffer for 60 min. 14μm frozen sections or 50μm non-frozen Microslicer sections were made, and K-NPPase activity was detected at both light and electron microscopic levels with the lead citrate method (Mayahara et al., Histochemistry, 67, 125-138, 1980).

Intense K-NPPase activity was localized on the bosolateral, but not apical membranes of the secretory cells. The activity was faintly positive in duct cells, but not in myoepithelial cells. The activity observed showed K-dependency and ouabain-sensitivity. ALPase was positive only on the myoepithelial cell membrane. As the mechenism of sweat secretion, passive diffusion of Na through the paracellular pathways was not plausible in the rat sweat gland, since the precursor sweat is rich in KCl (Sato, 1979), and the zonula occuludens has at least several strands. Instead, KCl secretion across the apical plasma membrane (passive or active) was speculated.

Relation of Lysosomal Wrapping Mechanism (LWM) to Microfilaments and Ca^{++}-ATPase Activity

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We have proposed the lysosomal wrapping mechanism (LWM) as a reasonable process of the autophagic phenomenon. In this study we further investigated the relation of LWM to microfilaments and Ca^{++}-ATPase activity. Mouse subcutaneous histiocytes stimulated by 1% ovalbumin show strong autophagic respones of LWM and the lysosomal movement was followed by the demonstration of ACPase activity. To reveal the intracellular microfilaments of the histiocyte 1-3% saponin was added to the fixative. Ca^{++}-ATPase activity was detected by the method of Ando et al. (1981). Histiocytes fixed with saponin-containing fixative revealed the intracellular microfilaments, ca 6-8 nm, which are closely related to the wrapping lysosome. Ca^{++}-ATPase activity was localized of the plasma membrane, inner saccules of Golgi apparatus and lysosomal membrane in addition to the spotty activity seen scatteredly around the wrapping lysosome. These results suggest an intimate relation of LWM to microfilaments and Ca^{++}-ATPase.