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MODIFICATION BY ENDOGENOUS PHOSPHOLIPIDS OF PORCINE KIDNEY
Na, K-ATPase

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The specific nature of the transport of ions in individual segments of a nephron is interconnected with the heterogeneity of the properties of transport ATPases, which depends to a considerable degree on the phospholipid composition of the membrane-bound ATPase complex [1]. The replacement of certain phospholipids (PLs) strongly bound to the enzyme molecule is accompanied by a substantial change in its activity [2-4]. However, literature information on the role of various PLs in the reactivation of delipidated Na, K-ATPase is contradictory and does not take the functional features of the objects investigated into account.

We have studied the protein-lipid ratios and phospholipic compositions of membrane-bound Na, K-ATPases from the cortical, medullary, and capillary zones of the porcine kidney, and also the influence of endogenous PLs on the activity of the enzyme. A decrease in the protein:lipid ratio in preparations of membrane-bound Na, K-ATPase was found along the cortical-papillary gradient: in the cortical zone it was 1:0.5; in the medullary zone, 1:0.9; and in the papillary zone, 1:1.4. The amount of PLs in the total lipid of the extract estimated on the basis of inorganic phosphorus (P_i) also differed over the zones of the kidney: in the cortical zone 54; in the medullary zone 60; in the papillary zone 62 $\mu g \; P_i/mg \; lipid$, which corresponds, when calculated to protein, to a threefold increase in the amount of PLs along the cortical-capillary gradient. The main differences in the phospholipid composition of the membrane-bound enzyme complex from functionally different sections of the nephron were observed in the amounts of polar PLs — phosphatidylserine, phosphatidylinositol, and sphingomyelin (Table 1). The enrichment of the composition with polar phospholipids correlates with a change in the activity of the Na, K-ATPase (% on the total ATPase activity (Table 2).

The modification of delipidated Na, K-ATPase was carried out by reconstructing it in sonicated (22 kHz, 5 min) proteoliposomes formed from proteins and lipids from different

TABLE 1. Phospholipic Composition (% on the total lipid P_i , M \pm m) of Membrane-Bound Na, K-ATPase according to the Zones of the Porcine Kidney

| Phospholipid | Zone | | |
|--|--|---|--|
| | cortical | medullary | papillary |
| Phosphatidylethanolamine Phosphatidylcholine Phosphatidylinositol Sphingomyelin Phosphatidylserine % of the lipid P _i found | 48,2±1,31 40,5±2,43 8,1±0,57 — 96,8±1,64 | $\begin{array}{c} 41,0\pm1,89\\ 36,3\pm1,80\\ 12,2\pm0,43\\ 5,5\pm0,32\\ &&&\\ 95,0\pm1,21 \end{array}$ | $\begin{array}{c} 39,0\pm1,42\\ 32,0\pm1,61\\ 11,1\pm0,74\\ 9,2\pm0,43\\ 6,5\pm0,51\\ \end{array}$ $97,8\pm1,16$ |

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TABLE 2. Modification of the Specific Na, K-ATPase Activity (% of the total ATPase, M \pm m) by Lipids from Functionally Different Sections of Nephron

| | 1 | Lipids from the | | |
|--|-------------------------------------|-------------------------------------|--|--|
| Proteins | cortical zone | medullary zone | papillary zone | |
| Cortical zone Medullary zone Papillary zone Membrane-bound | 26,1±2,31 30,8±2,74 34,6±3,16 | 51,2±4,83 49,8±4,42 80,7±4,46 | 65,3±5,32 63,4± 6 ,83 63,4±6,89 | |
| enzyme | $22,0\pm 2,18$ | 44,1±3,82 | 57,4 <u>+</u> 3,48 | |

sections of the nephron. The proton:lipid ratio was kept in agreement with the ratio in the native preparations of membrane-bound complex. The amount of protein in a sample was $5-10~\mu g$.

In our experiments, the lipids of the medullary zone possessed a considerably more pronounced capacity for modifying specific Na, K-ATPase activity than the lipids of the cortical zone (Table 2).

The results obtained, together with information from the literature [5, 6] on the restoration of the activity of Na, K-ATPase by individual polar PLs, from evidence in favor of the hypothesis put forward [1-3] of the determining role of polar PLs in the modification of the biological activity of ion-transporting proteins of functionally different sections of the nephron.

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