INTERACTIONS BETWEEN PHOSPHATE TRANSPORT AND OXIDATIVE METABOLISM

IN THE RABBIT PROXIMAL TUBULE

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INTRODUCTION

In proximal renal tubules, inorganic phosphate is both a transported solute and a substrate for intracellular metabolism. While these two functions may involve separate pools of phosphate, it is more likely that there is some interaction between phosphate transport and the metabolic processes which use inorganic phosphate. Within the proximal tubular cell, several types of metabolic processes involve inorganic phosphate. Glycolysis and oxidative phosphorylation utilize inorganic phosphate in the production of ATP. On the other hand, gluconeogenesis and the sodium-potassium ATPase hydrolyze ATP to generate inorganic phosphate. Furthermore, the intracellular concentration of inorganic phosphate may contribute to the phosphorylation potential, i.e. [ATP]/[ADP]·[Pi], which regulates the rates of these metabolic processes.

The objective of the present study was to examine the relationship of one aspect of intracellular metabolism, oxidative phosphorylation, to phosphate transport in proximal renal tubules. Our approach was to inhibit or stimulate oxidative metabolism and to determine the effect of each maneuver on specific transport processes. The data indicate that phosphate transport in proximal convoluted tubules is closely coupled to the rate of oxidative phosphorylation.

METHODS

Two basic methods were used to study transport and
metabolism in renal tubules. 1. Transport studies. Proximal convoluted tubules from rabbit kidney were isolated and perfused by previously described techniques. These tubules were perfused with a physiological fluid that resembled an ultrafiltrate of rabbit serum. The bathing medium was either commercial rabbit serum or the perfusion fluid plus 6 g/dl defatted albumin or 3 g/dl dialyzed dextran (40,000 mol. wt.). Lumen-to-bath fluxes of phosphate and glucose were measured with radioisotopes. Fluid absorption rates (Jv) were measured with a water marker, radiolabelled iothalamate. 2. Metabolic studies. Oxygen consumption rates were measured in suspensions of cortical tubules from rabbit kidney. Separated tubules were prepared by a collagenase infusion of the kidney by previously described techniques. These tubules were then suspended in a physiological fluid containing dextran. The final concentration of tubules was 3-5 mg tubular protein per ml. Suspensions were placed in a closed chamber where the oxygen tension was monitored as a function of time with a Clark electrode. The slope of this plot reflected the oxygen consumption rate which was normalized per mg tubular protein in the suspension. Data are expressed as a mean ± SE for a number of tubules or suspensions examined. Each tubule or suspension served as its own control prior to the addition of the test substance.

RESULTS

Effects of Arsenate

Arsenate is an inorganic analogue of phosphate in many transport and enzyme systems. In renal brush-border membranes, arsenate is a competitive inhibitor of phosphate uptake. In the intact epithelium, however, the primary action of arsenate is as an uncoupler of oxidative phosphorylation. We examined the effects of arsenate on tubular transport function to determine the relationship of specific transport processes to mitochondrial function. It should be noted that arsenate is a relatively weak uncoupler of oxidative phosphorylation. The addition of 1 mM arsenate to suspensions of cortical tubules increased oxygen consumption rates by 20.5 ± 2.9 percent. Thus, this concentration of arsenate may be expected to cause only a partial reduction in mitochondrial production of ATP.

The effects of arsenate on tubular transport were examined in isolated, perfused proximal convoluted tubules. The addition of arsenate to only the bathing medium had no effect on tubular function. The addition of 1 mM arsenate to the luminal fluid significantly reduced transport processes. Therefore, arsenate must enter the tubular cell across the brush-border membrane. The pattern of arsenate-induced inhibitions of transport was unusual.