AN INVESTIGATION OF THE EFFECT OF GENTAMICIN ON Na^+-K^+ ATPase AS A POSSIBLE MECHANISM OF TOXICITY

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It has been suggested that inhibition of plasma membrane Na^+-K^+ ATPase by aminoglycoside antibiotics, such as gentamicin, may be a mechanism common to both their nephrotoxicity and ototoxicity. Iinuma et al (1967) and Kaku et al (1973) showed that aminoglycosides depressed Na^+-K^+ ATPase activity in inner ear tissues. Lipsky & Lietman (1980) reported that neomycin produced almost maximal inhibition of canine renal Na^+-K^+ ATPase at 0.55×10^{-3}M.

In the present work, a study was made of the effect of gentamicin on Na^+-K^+ ATPase (ouabain-sensitive Na^+-K^+ ATPase) of human erythrocyte membrane, cortical microsomal fraction of rat kidney and rat isolated renal tubules. In the first two of these systems Na^+-K^+ ATPase activity was determined by the release of inorganic phosphate using a phosphomolybdate method (Ueda & Wada 1970). It was necessary to develop a modification of this method to avoid interference from gentamicin. In rat isolated renal tubules Na^+-K^+ ATPase activity was assayed by following the uptake of ^{86}Rb (a congener of K^+) by the tubules.

Table 1. Percentage inhibition of Na^+-K^+ ATPase activity by gentamicin (NE = No effect)

Gentamicin concentration	Erythrocyte membrane	Microsomal fraction	Renal tubules
10^{-4} M	7 ± 2	NE	NE
10^{-3} M	25 ± 4	18 ± 2	NE
10^{-2} M	110 ± 11	59 ± 4	47 ± 3

There was no appreciable effect of gentamicin on Na^+-K^+ ATPase activity in any of the systems at a concentration of 10^{-4} M. Although there was some inhibition at 10^{-3} M in both the erythrocyte membrane and microsomal fraction preparations, the effect of gentamicin was marked only at a concentration of 10^{-2} M. This high concentration totally abolished the activity of Na^+-K^+ ATPase of the erythrocyte membrane. The lesser degree of inhibition at 10^{-2} M in the other preparations might be explained by the presence of other binding sites for the drug, thus effectively reducing the concentration of drug available to the Na^+-K^+ ATPase enzyme complex.

The isolated renal tubule preparation was included because of the problems of extrapolating results from fragmented membranes to intact cells. Although this system was less sensitive to the effect of gentamicin, the similarity of the pattern of inhibition in the different types of preparation suggested a valid relationship between the three systems. However, the very high concentration of gentamicin required to produce significant inhibition suggests that interaction with Na^+-K^+ ATPase is unlikely to be the primary mechanism of toxicity.

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0022-3573/81/120050P-01$02.50/0