Altered activity of the sodium–potassium pump in arteries of rats with steroid hypertension

MOTILAL B. PAMNANI, DAVID L. CLOUGH AND FRANCIS J. HADDY

Department of Physiology, Uniformed Services University of the Health Sciences, Bethesda, Maryland U.S.A.

Summary

1. Ouabain-sensitive uptake of $^{86}$Rb, a measure of the Na$^+$$-$K$^+$ pump activity, was studied in tail arteries of rats made hypertensive with deoxycorticosterone and saline.

2. Decreased activity of the ouabain-sensitive Na$^+$$-$K$^+$ pump supports the hypothesis that the activity of Na$^+$$-$K$^+$ pump is suppressed in volume-expanded hypertension.

Key words: dexamethasone-hypertension, deoxycorticosterone-hypertension, hypertension, Na$^+$$-$K$^+$ pump.

Introduction

We have previously shown that ouabain-sensitive $^{86}$Rb uptake, a measure of the Na$^+$$-$K$^+$ pump activity (Bernstein & Israel, 1970; Ku, Akera, Pew & Brody, 1974), is reduced in mesenteric arteries and veins of dogs with hypertension produced by cellophane wrapping of a single kidney (Overbeck, Pamnani, Akera, Brody & Haddy, 1976). We have also shown that myocardial membrane Na$^+$$-$K$^+$-dependent ATPase, the pump enzyme, is depressed in microsomes prepared from either the left or the right ventricle of rats with one-kidney Goldblatt hypertension (Clough, Pamnani & Haddy, 1977). Because the Na$^+$$-$K$^+$ pump is electrogenic in vascular smooth muscle (Thomas, 1972; Hendrickx & Casteels, 1974), decreased activity of the Na$^+$$-$K$^+$ pump would partially depolarize the muscle cell membrane. Depolarization of the vascular smooth-muscle cells could help account for many manifestations of the hypertensive process, e.g. the elevated vascular resistance and decreased venous compliance (Overbeck et al., 1976; Simon, Pamnani, Dunkel & Overbeck, 1975).

The purpose of the present study was to determine whether or not a similar defect in pump activity occurs in other forms of experimental hypertension.

Methods

Male normotensive wistar rats weighing 220–260 g underwent unilateral nephrectomy and were randomly divided into three groups. Group 1 served as a control and was given tap water ab libitum. In Group 2 hypertension was induced by an initial injection of 12.5 mg of deoxycorticosterone acetate (DOCA) in oil (Percorten) followed by 6.5 mg weekly, coupled with 1% saline as drinking water. In Group 3, hypertension was produced by an initial injection of 12.5 mg of dexamethasone phosphate [DEXA (Decadron)] followed by 6-5 mg weekly. These animals, like the control rats, drank tap water. All three groups were maintained on normal rat chow. Systolic blood pressure (plethysmographic method) and body weight were monitored weekly in all rats. After 8 weeks of DOCA–saline or DEXA treatment, and at a similar time interval in the control group, rats were anaesthetized with sodium pentobarbital 75 mg/kg body weight intraperitoneally. Tail arteries were excised simultaneously from control (NT), DOCA–saline hypertensive (DOCA HT), and DEXA-treated hypertensive (DEXA HT) rats and immediately placed in Krebs–Henseleit solution aerated with O$_2$ + CO$_2$ (95 : 5).

The uptake of $^{86}$Rb was measured as in our previous study (Overbeck et al., 1976). The tissue
was first incubated at 0°C in K⁺-free Krebs-Henseleit solution to depress the pump and load the cells with Na⁺. Next, to stimulate the pump, the artery was incubated in 37°C K⁺-free Krebs-Henseleit solution containing 2 mM RbCl. Each tail artery was divided in half. One half was incubated in medium without ouabain and the other half in medium containing 0-8 mM-ouabain. 86Rb (New England Nuclear) was added to each medium to a standard concentration of 0-01 mM and the incubation continued for 18 min. The media were oxygenated with O₂ + CO₂ (95:5). At the end of incubation, tissues were rapidly washed with K⁺-free Krebs-Henseleit solution containing 2mM-RbCl, blotted to remove surface fluid, weighed and placed in a scintillation counter (1185 Searle) to determine 86Rb uptake (pmol/mg of tissue). The tissues were then placed in an oven at 100°C for 24 h and reweighed to determine dry weight.

Specific 86Rb uptake (ouabain-sensitive) was calculated as the difference between the 86Rb uptake without and with ouabain. The paired t-test was used to compare group means of uptakes; P values < 0.05 were considered significant.

Results

Group 2 (DOCA—saline) rats showed a progressive rise in mean systolic pressure from a pretreatment value of 123.1 ± 1.3 to 135.9 ± 2.4 mmHg (P < 0.01) by the second week of treatment and 175.3 ± 2.4 mmHg (P < 0.001) by the eighth week. DEXA treatment also induced hypertension (Group 3). Pretreatment mean systolic pressure was 122.9 ± 2.2 mmHg whereas the pressures after 1 and 8 weeks of treatment were 133.0 ± 4.4 (P < 0.05), and 170.1 ± 1.2 (P < 0.001) respectively. No significant change in blood pressure occurred in the control rats (Group 1) during this time (120.0 ± 1.6 to 123.8 ± 1.1 mmHg, P > 0.5).

During 8 weeks of treatment the weight of the DOCA HT rats increased from 241.8 ± 4.5 to 413 ± 8.5 g and the weight of the DEXA HT rats increased from 252.0 ± 5.3 to 335.6 ± 15.5 g. However, during the same time period, the weight of the control rats increased from 238.5 ± 8.6 to 489.2 ± 13.2 g. Thus the DOCA HT and DEXA HT rats did not gain weight as fast as the control rats.

Compared with the arteries from normotensive control animals, the ouabain-sensitive 86Rb uptake in the arteries from DOCA—saline treated rats was decreased (Fig. 1, P < 0.02). In contrast, this uptake was significantly increased (P < 0.05) in arteries from DEXA treated rats. The ouabain-insensitive 86Rb uptake in tail arteries from DEXA HT rats was normal (P > 0.5). However, this uptake was significantly increased (P < 0.05) in the tail arteries from DOCA HT rats.

Discussion

The results of this study indicate that the ouabain-sensitive uptake of 86Rb is decreased in tail arteries of DOCA—saline treated rats. In contrast, this uptake is increased in arteries from DEXA-treated rats. This difference is probably attributable to the difference in the type of hypertension induced by
these two treatments. DEXA, unlike DOCA, lacks mineralocorticoid activity (Goodman & Gilman, 1970). It is therefore reasonable to assume that DEXA-treated rats are not as volume expanded as DOCA-saline-treated rats. On the other hand, DEXA, like many other steroids, may stimulate the synthesis of membrane Na\(^{+}\)–K\(^{+}\) ATPase (Hegyvary, 1977; Charney, Silva, Besarab & Epstein, 1974). This additional ATPase would be largely unopposed by the inhibitory effect of volume expansion (Overbeck et al., 1976; Gonick, Kramer, Paul & Lu, 1977). This might explain the increased activity of the Na\(^{+}\)–K\(^{+}\) pump in vessels from DEXA-treated rats.

Suppressed \(^{86}\text{Rb}\) uptake in DOCA-saline hypertensive animals contrasts with the finding of increased K\(^{+}\) uptake (Friedman & Friedman, 1976). Perhaps the difference is related to the time of study. We studied the arteries almost immediately after excision whereas some hours elapsed in the Friedman study.

Interpretation of the increased ouabain-insensitive \(^{86}\text{Rb}\) uptake in DOCA-saline hypertension must await further studies. This may reflect cell membrane permeability or other factors.

Decreased ouabain-sensitive \(^{86}\text{Rb}\) uptake is also found in one-kidney, chronic renal hypertension in dogs (Overbeck et al., 1976). Thus the findings in this study support our hypothesis that the activity of the Na\(^{+}\)–K\(^{+}\) pump in vascular smooth muscle is suppressed in volume-expanded hypertension.