phages was still five times greater than control numbers and this could have been related to lack of complete recovery in permeability, which we found in our subjects after stopping smoking. We have not found a relation between amount smoked and the change in number of alveolar macrophages, and it would be interesting to compare the result of such a study with our finding of a relation between carboxyhaemoglobin and half-time clearance.

The large change in the results of the permeability test and lack of effect on tests of ventilatory function are noteworthy. This implies that the alveolar permeability to the tracer ^{99m}Tc-DTPA is a very sensitive test of pulmonary dysfunction in cigarette smokers and the results are correlated with exposure to tobacco smoke. During the 21 days after stopping smoking recovery into the normal non-smoking range was the exception, and the greatest degree of recovery occurred in those with least exposure to tobacco smoke.

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Familial abnormality of erythrocyte cation transport in essential hypertension

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Abstract

Cation transport across the red-cell membrane was studied in subjects with essential hypertension and their relatives using rubidium-86 as an analogue of potassium. The activity of the ouabain-sensitive sodium-potassium pump was significantly greater in patients with untreated essential hypertension than in controls ($p < 0.001$). No clear separation was seen between normotensive and hypertensive subjects. Activity of the sodium-potassium pump was also increased in a proportion of normotensive relatives of subjects with essential hypertension. Rubidium uptake was significantly lower in normotensive black subjects than in normotensive whites, the difference being in a ouabain-resistant pathway of cation transport.

These results provide further evidence that a defect in membrane cation transport contributes to the pathogenesis of essential hypertension.

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Introduction

Garay and his colleagues have reported finding an abnormality of sodium and potassium flux across the red-cell membrane in untreated essential hypertension which was absent in normotensive controls. In addition, some normotensive subjects whose parents had essential hypertension were found to have a similar abnormality.[1 2] These results were obtained by measuring sodium and potassium transport in sodium-loaded, potassium-depleted red cells during exposure to *p*-chloromercuribenzenesulphonate. The data have been criticised because of the unphysiological conditions of the experiment.[3] In view of the potential importance of identifying a familial biochemical abnormality in essential hypertension, we have examined erythrocyte cation transport using an isotopic method which allows the activity of the ouabain-sensitive sodium-potassium adenosine triphosphatase pump (sodium pump) to be measured under less artificial conditions. Red-cell uptake of rubidium, which is handled by the sodium pump in the same way as potassium,[4 5] was monitored with the gamma-emitting isotope ^{86}Rb. The same technique is used extensively to measure inhibition of the sodium pump by cardiac glycosides.[6]

Because of reported racial differences both in sodium handling[7] and in the prevalence of essential hypertension[8 9] we have also compared red-cell cation transport by the same method in normotensive black subjects and normotensive whites.

Subjects and methods

We studied four groups of subjects. *Group 1* comprised white normotensive adults (mean age 34·5 years, range 19-64) with no known family history of hypertension. Normotension was defined as a blood pressure not exceeding 140/90 mm Hg (phase V diastolic) in the sitting position. *Group 2* comprised normotensive black (West Indian) adults (mean age 34·4 years, range 16-59) with no known family history of hypertension. *Group 3* comprised white subjects (mean age 47·5 years, range 28-72) with essential hypertension (blood pressure exceeding 140/90 mm Hg) before treatment. Secondary hypertension was excluded by conventional clinical, laboratory, and (where indicated) radiological criteria. *Group 4* comprised normotensive, white, first-degree relatives of subjects with essential hypertension (mean age 33·2 years, range 19-49).

The normotensive groups were drawn from hospital and laboratory staff, factory workers undergoing blood-pressure screening, and hospital patients awaiting minor elective operations. The hypertensive subjects were untreated outpatient referrals or factory workers whose condition was detected during screening. Normotensive relatives were contacted through patients attending the hypertension clinics. For all groups a careful drug history was recorded and subjects taking any form of prescribed or proprietary medication excluded. None of the subjects was grossly obese.

The laboratory procedure was based on the method of Aronson *et al.*[6] Red cells were separated from heparinised blood by centrifugation and washed three times in isotonic saline. Packed cells (1 ml) were mixed with 1·5 ml potassium-free Ringer's solution and 0·5 ml ^{86}RbCl solution (Radiochemical Centre, Amersham; final concentration 3 μmol/l; 26 μg/100 ml). The final composition of the medium was sodium 143·4 mmol(mEq)/l, calcium 1·34 mmol/l (5·4 mg/100 ml), magnesium 1·26 mmol/l (3·1 mg/100 ml), chloride 126 mmol (mEq)/l, and glucose 5·56 mmol/l (100·2 mg/100 ml).

Two 0·5 ml aliquots were removed immediately for counting total radioactivity. The remaining cell suspension was incubated for one hour in a shaking water-bath at 37°C. Three further 0·5 ml aliquots were then removed and the cells separated by centrifugation at 12 000 *g* for one minute. The cells were washed three times in cold isotonic saline, radioactivity of the final cell pellet counted, and rubidium uptake calculated (μmol/l cells/h; ^{86}Rb: 1 μmol = 86 μg). For each blood sample a replicate experiment was performed simultaneously in the presence of ouabain 1 g/l to derive uptake by the ouabain-sensitive sodium-potassium pump.

The coefficient of variation for multiple determinations on a single blood sample was 3·9% (n = 10). The mean coefficient of variation for ^{86}Rb uptake measured on four or five successive days in four subjects was 4·8%.

Statistical—Rubidium uptakes (total, ouabain-sensitive, and ouabain-resistant) were compared between groups by the Mann-Whitney test, since a normal distribution of the data could not be assumed.

Results

Age and sex—Rubidium uptake was not related to age in a group of 41 normotensive subjects aged 19-64 years. Mean uptakes did not differ significantly between men and women.

Race—Figure 1 shows the rubidium uptakes in normotensive black and white volunteers. Total uptake was significantly lower in blacks than in whites. The difference was entirely due to the ouabain-resistant component of rubidium influx ($p < 0·0001$). Sodium-potassium pump activity was the same in both groups. In view of this finding the following studies on subjects with essential hypertension and their relatives all refer to white people.

Essential hypertension—Total rubidium uptake was greater in subjects with essential hypertension than in normotensive controls ($p < 0·001$) (fig 2). This difference was due entirely to increased activity of the ouabain-sensitive sodium-potassium pump in the hypertensive subjects (fig 3). Ouabain-resistant rubidium uptake was not significantly different in the two groups. Despite the highly significant population differences in total rubidium uptake and ouabain-sensitive uptake, individual values showed some degree of overlap between the two groups of subjects.

Normotensive relatives of hypertensive subjects—Total rubidium uptakes among the normotensive relatives extended over the entire range of normotensive and hypertensive values (fig 2). Owing to the overlap between normotensive and hypertensive groups we could not divide normotensive relatives into those with normal rubidium uptake and those with unequivocally abnormal values. Considerably larger numbers of normotensive relatives will be required to establish whether their uptake is bimodally distributed. Analysis of the normotensive relatives as a single group showed that ouabain-resistant transport was similar to that of the normotensive controls and hypertensive subjects but that the ouabain-sensitive component of rubidium flux was intermediate between the two other groups (fig 3). Mean blood pressure (diastolic plus one-third of the pulse pressure) in the normotensive relatives was not significantly higher than in the normotensive controls (mean ± SD: relatives 91·0 ± 8·5 mm Hg; controls 93·8 ± 8·6 mm Hg).

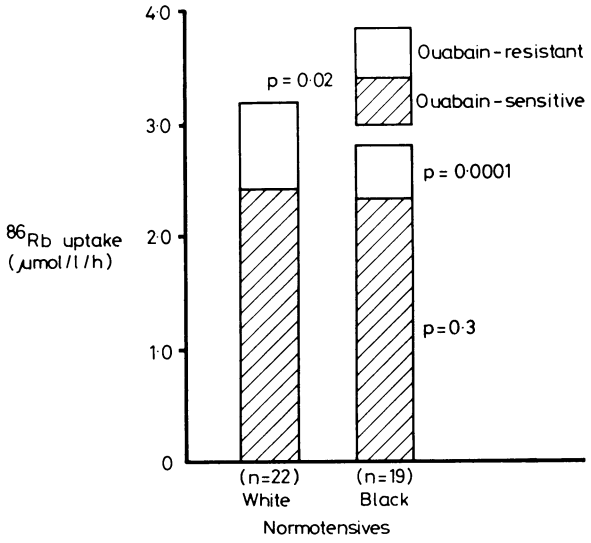

FIG 1—Red-cell rubidium uptake in normotensive white and black subjects. Statistical analysis by Mann-Whitney test. (^{86}Rb: 1 μmol = 86 μg.)

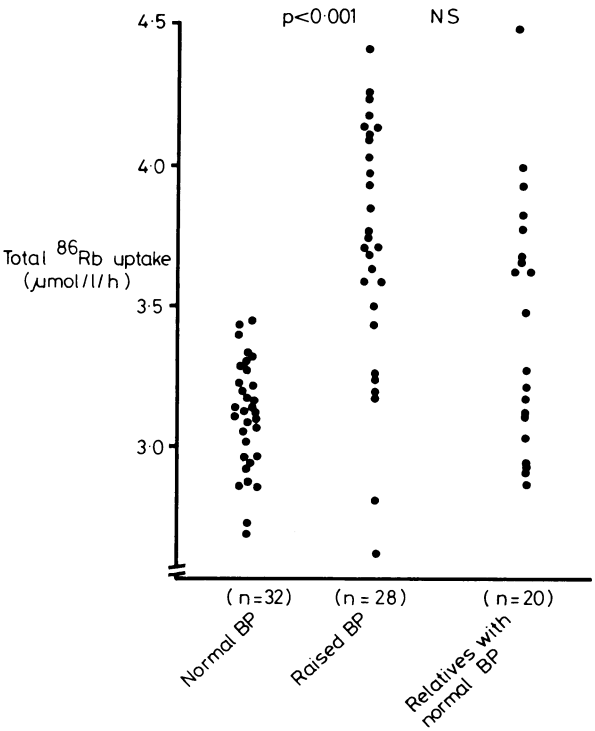

FIG 2—Total red-cell rubidium uptake in normotensive controls, subjects with essential hypertension, and normotensive first-degree relatives of subjects with essential hypertension. NS = Not significant. (^{86}Rb: 1 μmol = 86 μg.)

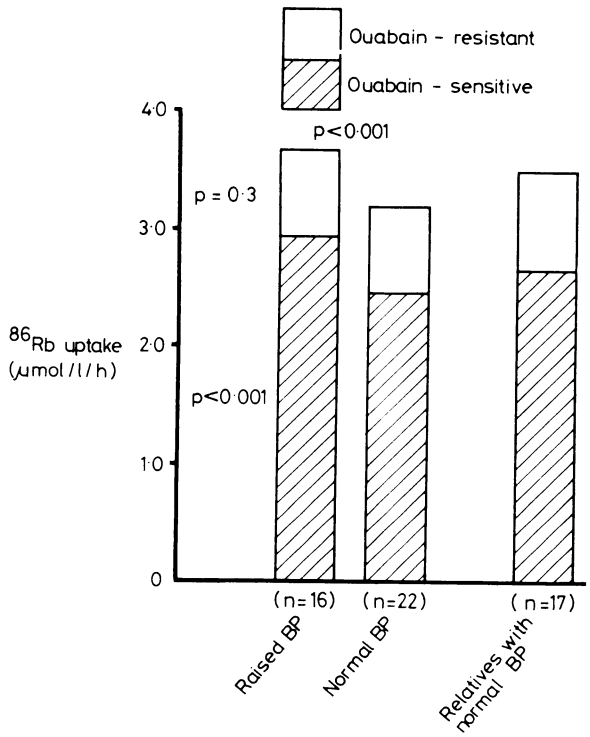

FIG 3—Comparison of ouabain-sensitive (sodium pump) and ouabain-resistant components of red-cell rubidium uptake in hypertensive and normotensive subjects and normotensive relatives. (^{86}Rb: 1 μmol = 86 μg.)

Discussion

Our results clearly show increased activity of the ouabain-sensitive sodium-potassium pump in erythrocytes of patients with essential hypertension. A similar finding was obtained with different techniques to measure sodium-potassium fluxes.[10] Such increased activity, however, may not be the primary abnormality of cation transport in these cells. It could result from increased passive influx of sodium ions or from a defect in an alternative, ouabain-resistant pathway of active sodium extrusion. Garay *et al*[10] found no evidence of increased red-cell sodium permeability to account for the observed increase in ouabain-sensitive sodium-potassium exchange but reported that a ouabain-resistant, frusemide-sensitive pathway of sodium extrusion is lacking in the red cells of hypertensives. This pathway, the sodium-potassium cotransport system, is present in the normal red-cell and uses the efflux of potassium down its electrochemical gradient to drive sodium "uphill," out of the cell. Increased activity of sodium-potassium adenosine triphosphatase in red-cell ghosts has recently been reported in essential hypertension by Wambach *et al*,[11] who used an enzymatic assay. It has also been reported that erythrocytes in essential hypertension show greater activity of a sodium-sodium (or sodium-lithium) countertransport pathway.[12] This does not bear an obvious relation to the abnormalities referred to above, since the countertransport pathway does not result in net sodium flux. In addition, studies on leucocytes in essential hypertension under steady-state conditions have shown an increased intracellular sodium concentration, normal net sodium efflux, and reduced ouabain-sensitive rate constant for sodium efflux.[13] The rise in intracellular sodium was suggested as secondary to impaired ouabain-sensitive sodium-potassium pump activity. Thus on present evidence we cannot provide a unifying hypothesis to reconcile all the available data on disturbed membrane transport of cations in essential hypertension.

Nevertheless, there is now impressive evidence that these abnormalities of cation transport are neither the consequence of raised blood pressure (since they are not seen in renal hypertension[2]) nor artefacts of treatment. Since the ouabain-sensitive sodium-potassium adenosine triphosphatase pump is a ubiquitous mechanism for maintaining transmembrane electrolyte gradients, there are several ways in which an abnormality of the pump might lead to altered handling of sodium by the kidney or to an increase in peripheral vascular resistance.[14] The role of the observed abnormalities in the pathogenesis of essential hypertension is speculative.

The evidence for a familial disturbance of cation transport reported here and elsewhere[1 10 12] does not prove a genetically based predisposition to hypertension. The unimodal distribution of blood pressure makes it likely that essential hypertension is multifactorial in origin rather than due to a simple interaction of a high dietary sodium intake with a single genetic defect in membrane transport. It is also noteworthy that we did not find a clear separation in ^{86}Rb uptake values between normotensive and hypertensive subjects. Further studies of normotensive relatives are clearly needed, particularly to see if indices of cation transport can be related to abnormalities of cardiovascular regulation.

The racial difference in ouabain-resistant rubidium uptake reported here is of particular interest, since normotensive black volunteers have been shown to excrete a sodium load more slowly than normal whites, with a greater and more persistent rise in blood pressure during saline infusion.[7] It has recently been found that in Birmingham there is a higher prevalence of hypertension in the black community than in the white.[15] The relation between cation transport and blood pressure in black subjects is being investigated.

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