Editorial Review

Is intracellular sodium increased in hypertension?

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INTRODUCTION

It is generally assumed that the intracellular Na⁺ content (Na⁺i) of vascular smooth muscle is increased in essential hypertension and several experimental models of hypertension [1, 2]. The assumption dates back to the original reports of increased Na⁺ content of arteries in human and experimental hypertension by Tobian [3]. Based on the finding of an elevated arterial wall Na⁺/Cl⁻ ratio, he concluded that some of the excess Na⁺ was intracellular. However, an increased Na⁺/Cl⁻ ratio is also compatible with the accumulation of excess cation-binding glycosaminoglycans in the extracellular matrix of arteries. Indeed, in dogs with hypertension secondary to coarctation of the aorta, Hollander et al. [4] found that the glycosaminoglycan content of the 'hypertensive' portion of the thoracic aorta was increased and might have been the factor directly responsible for the accumulation of excess Na⁺ [4]. Extracellular bound Na⁺ was found to be increased also in the tail artery of rats with deoxycorticosterone acetate (DOCA)-salt and two-kidney, one-clip (2K,1C) hypertension [5, 6].

ERYTHROCYTES AND LEUCOCYTES

The first direct evidence for increased Na⁺ in hypertension came not from the study of vascular smooth muscle but from the study of erythrocytes of hypertensive subjects [7]. Due to their ready availability and ease of handling, erythrocytes have been used extensively for study of the transmembrane distribution of cations in hypertension [8–11]. From the beginning, investigators had two methodological concerns; first, the separation of erythrocytes from leucocytes and platelets in whole blood, and secondly the trapping of plasma with its high Na⁺ concentration in the erythrocyte pellet after centrifugation. Consequently, lengthy centrifugation procedures were used, followed by washing of erythrocytes in ice-cold Na⁺-free solutions. In ice-cold solutions, erythrocytes shrink with a reduction in the water content of the cells (22.9 ± 1.6 litres/kg dry weight, mean ± SD, n = 8, at 4°C vs 27.1 ± 1.6 litres/kg dry weight, n = 8, at 37°C, P < 0.01, by paired Student's t-test; G. Simon, unpublished work). This may lead to overestimation of Na⁺ when results are expressed per litre of erythrocytes [12]. In addition, in the majority of studies there was considerable delay between the withdrawal of blood and the measurements. Not surprisingly, the reported range of erythrocyte Na⁺ has been large (2.4–31.3 mmol/litre of erythrocytes), and the differences between hypertensive and normotensive subjects have been inconsistent [8–11]. There are several comprehensive reviews of erythrocyte Na⁺ in human hypertension [1, 13, 14]. In 1983, Parker & Berkowitz [13] reviewed 21 published reports: erythrocyte Na⁺ of hypertensive subjects was increased in 10 communications and unchanged in 11. Hilton [14] analysed data pooled from 20 published reports: in approximately half erythrocyte Na⁺ of hypertensive subjects was increased in 10 communications and unchanged in 11. Of the 10 positive reports, six originated from the laboratories of Losse, Wessels and co-workers. Their findings were summarized in 1980 [9]: they found a 12% increase (P < 0.01) in erythrocyte Na⁺ of 295 subjects with essential hypertension compared with 319 normotensive control subjects. Hilton [14] analysed data pooled from 20 published reports: in approximately half erythrocyte Na⁺ of hypertensive subjects was increased in the remaining half unchanged. Hilton found that the discrepancies between studies were far too great to be attributed to chance alone and suggested that either variation in methodology masked differences between hypertensive and control subjects, or that the populations sampled by different centres were not comparable. Mismatch of groups within the same study is also possible. It may cancel out differences that exist between groups or show differences
where there are none. In this regard, in both normotensive and hypertensive black subjects, erythrocyte \(Na^+\) is higher than in Caucasians [15, 16]. In one study, rigorous matching of subjects for race, sex, age and body weight eliminated the differences in erythrocyte \(Na^+\) that existed between groups before matching [17]. The direct correlation of erythrocyte \(Na^+\) with age and body weight may account for this finding [17]. Recent evidence also shows that antihypertensive therapy may influence erythrocyte \(Na^+\) transport and transmembrane \(Na^+\) gradient for several weeks after discontinuation of therapy. In comparison with values obtained in normotensive controls, erythrocyte \(Na^+\) was increased and the number of \(Na^+\)–\(K^+\) pump sites per erythrocyte was reduced in hypertensive black subjects who were withdrawn from diuretic therapy 2–6 weeks before the study [18]. In contrast, in previously untreated hypertensive black subjects erythrocyte \(Na^+\) and the number of pump sites were unchanged [18]. Zidek et al. [19] have shown that erythrocyte \(Na^+\) rises during antihypertensive therapy with a loop diuretic [19]. The rise in erythrocyte \(Na^+\) is detactable by 2 weeks, and there is an additional increase at 6 weeks after the initiation of therapy. In order to explain these prolonged and cumulative effects of diuretics on erythrocyte \(Na^+\) and \(Na^+\) transport, one has to assume that the changes occur in the bone marrow during erythropoiesis while erythrocytes are nucleated and capable of protein synthesis de novo. Steroids also have a prolonged effect on erythrocyte \(Na^+\) and transport, requiring 6–7 weeks before the changes can be detected in circulating erythrocytes, but unlike diuretics which raise \(Na^+\), steroids reduce it [20]. The life span of human erythrocytes being about 120 days, several weeks are needed before newly formed erythrocytes replace circulating ones in sufficient number for the differences in \(Na^+\) and pump activity to be detectable. Finally, there are also reports of increased \(Na^+\) in separated and washed erythrocytes of rats and pigs with DOCA-salt hypertension [21, 22]. Erythrocyte \(Na^+\) is unchanged in rats with renal and spontaneous hypertension [21].

In 1983, we set out to confirm the finding of increased erythrocyte \(Na^+\) in human hypertension by studying the cells under experimental circumstances that resembled conditions \textit{in vivo} [23]. Samples for measurement were prepared within seconds after venepuncture, by centrifuging whole blood over oil. This was possible because the relatively small number of leucocytes and platelets in the cell pellet had no detectable effect on erythrocyte cation measurements. Because extracellularly trapped (plasma) \(Na^+\) may account for 20–30% of the total \(Na^+\) content of the cell pellet, plasma trapping was measured in every experiment by adding ^125I-albumin and ^51Co-labelled ethylenediaminetetra-acetate to whole blood. Two extracellular fluid markers were used, because each gives a slightly different estimate of extracellular fluid space. To our surprise, we found that erythrocyte \(Na^+\) in Caucasian male hypertensive subjects was reduced, not increased. There was in addition a small increase in the intracellular \(K^+\) content (\(K_s\)) of erythrocytes. We have since confirmed these findings [24]. Our finding of reduced erythrocyte \(Na^+\) and \(Na^+\)/\(K^+\) in hypertension is not unique. Trevisan et al. [11] reported reduced \(Na^+\) in previously untreated Caucasian males with borderline hypertension but did not comment on this interesting finding. Low erythrocyte \(Na^+\)/\(K^+\) was reported in children with mild blood pressure elevation and a positive family history of hypertension, a finding that is compatible with reduced total body \(Na^+\) content in early hypertension [25, 26]. Erythrocyte \(Na^+\) was also reduced in hypertensive patients with Cushing's syndrome (glucocorticoid excess) and in patients receiving pharmacological doses of prednisone, many of whom were hypertensive [20, 27]. Last year, Engelhardt & Scholze [28] confirmed our finding of reduced erythrocyte \(Na^+\) in mild human hypertension [140/90 mmHg (18.7/12 kPa) < blood pressure < 160/95 mmHg (21.3/12.7 kPa)]. Interestingly, in more severe hypertensive subjects, they found erythrocyte \(Na^+\) to be unchanged. Finally, erythrocyte \(Na^+\) is increased in the Milan strain of spontaneously hypertensive rats [29].

In contrast to conflicting reports regarding erythrocyte \(Na^+\), there is, with some exceptions, agreement that the \(Na^+\) of leucocytes in general and of lymphocytes in particular is increased in both human and experimental hypertension [14, 30–41]. The separation of leucocytes from whole blood is a more cumbersome and time-consuming process than that of erythrocytes, during which leucocytes incubate in the subjects' own plasma for 30–60 min at room temperature. After separation, the cells are either washed in ice-cold \(Na^+\)-free solution or resuspended in tissue culture medium and then pelleted in the presence of an extracellular fluid marker. Four major laboratories specializing in the study of membrane phenomena in hypertension have used whole leucocyte fractions or lymphocytes [14, 30–38]. In all but one of them [35–38], increased leucocyte \(Na^+\) has been a consistent finding. Increased leucocyte \(Na^+\) has been reported in human hypertension [14, 30, 31, 33] and in rats with renal, DOCA-salt and spontaneous hypertension [32, 34]. However, no differences in leucocyte \(Na^+\) were detected between hypertensive and normotensive subjects [35, 37, 38] or between rats with renal, DOCA-salt and spontaneous hypertension and their controls in one laboratory [36]. The \(K_s/Na^+\) ratio of isolated leucocytes in the latter reports were consistently higher (5.1 to 13:1) [35–38] than that in other laboratories (2:1 to 5:1) [30–34]. This finding suggests important differences in the processing of leucocytes. The main difference appears to be the incubation of isolated leucocytes in tissue culture medium at 37°C for 30–60 min before electrolyte measurements by the group that reported the higher \(K_s/Na^+\) ratios. There are also isolated reports of increased leucocyte \(Na^+\) in human hypertension [39–41].

**VASCULAR MUSCLE**

Implicit in the investigation of erythrocytes and leucocytes in hypertension is the assumption that these cells are representative of vascular smooth muscle cells. This remains to be proven. Measurement of \(Na^+\) in the intact...
artery, the target organ of hypertension, is more difficult
than in erythrocytes or leucocytes. There are at least four
compartments of Na⁺ in the blood vessel wall: Na⁺
dissolved in extracellular or intracellular water, and Na⁺
bound to extracellular or intracellular matrix [5, 6,
42-45]. Only about 18% of arterial wall Na⁺ is intra-
cellullary located. There are several approaches to the
measurement of Na⁺ in blood vessels, but none of them
may be precise enough to detect small differences [44,
46-48]. They include chemical dissection, flux analysis,
cold Li⁺ washout, electron probe analysis, ion-selective
microelectrodes, and, more recently, nuclear magnetic
resonance techniques. Chemical analysis is based on the
rationale that, if one can account for all non-cellular Na⁺,
the cellular component can be computed as the difference
between the total and the extracellular Na⁺ content [46].
Extracellular fluid markers, each yielding a different
result, are used to estimate the amount of Na⁺ dissolved
in extracellular water. The estimate of extracellularly
adsorbed Na⁺ is based on measurements of the glyco-
aminglycan content of the vessel wall. Flux analysis using
22Na or 24Na requires prolonged incubation of vascular
tissue in artificial salt solution for loading so that the
results of measurements may not represent conditions in
vivo. The radionuclide efflux curves can at best be
resolved into three components which makes the identifi-
cation of the four Na⁺ compartments difficult. Also,
efflux rate constants may be affected by diffusion delay
across the extracellular space [46]. Diffusion delays may
differ between the hypertrophied or hyperplastic arteries
of hypertensive animals and the arteries of normotensive
controls. The Li⁺ exchange method is based on the slow
exchange of Li⁺ for intracellular Na⁺ in vascular tissue at
2°C [44]. The rate of exchange of Li⁺ for intracellular
Na⁺ is sufficiently slow in rat arteries to distinguish
between the rapid washout of extracellular Na⁺ and the
much slower washout of intracellular Na⁺. In dog and
rabbit arteries and veins, this distinction is more difficult
due to the more rapid rate of cation exchange across the
cell membrane [49]. Extrapolation of the slow phase of the
washout curve to time zero provides an estimate of Na⁺.
For accurate results, multiple sampling of vascular tissue
for up to 90 min of incubation in cold Li⁺ solution is
required. In the case of a small artery, such as the rat tail
artery, the amount of tissue available is inadequate for
accurate measurement of Na⁺. Arteries from several rats
have to be studied to construct a washout curve, and the
estimated Na⁺ is, therefore, the group mean. Recently, in
phasisically contracting (spike-generating) vascular muscle,
a rapidly exchanging and temperature-insensitive
exchange of Li⁺ for intracellular Na⁺ was identified [48].
It remains to be seen whether such a pathway also exists
in non-spiking generaing vascular muscle such as the
peripheral arteries. If it does, it may result in the under-
estimation of Na⁺ by the cold Li⁺ exchange method and
by flux analysis. The technological requirements for
electron probe analysis of vascular tissues are beyond the
means of all but a few laboratories [48]. Tissue specimens
have to be frozen instantaneously to stop the diffusion
of ions and crystallization of water. Thin frozen sections are
placed in an electron beam. The ions in the field emit
X-rays of characteristic energy that are detected and
counted. Using this method, Somlyo and co-workers [48,
50, 51] found a uniform distribution of Na⁺ in the cyto-
plasm and intracellular organelles of vascular smooth
muscle cells but a wide cell-to-cell variation in Na⁺. They
also found that the Na⁺ of the phasically contracting
(spike-generating) rabbit portal vein was about twice as
high as the Na⁺ of the tonically active (non-spike-generat-
ing) pulmonary artery [51]. The latter is more in line with
Na⁺ values obtained in the peripheral arteries of rats [46].
Ion-sensitive microelectrodes have been used sparingly
for measurements of intracellular cation activities in
vascular muscle, not to mention group comparisons of
Na⁺. Considering the small diameter of the cells and the
abundance of connective tissue in the vessel wall, there is,
even in skilled hands, a large proportion of inadequate
impalments which must be distinguished from the valid
ones by firm criteria [47]. The wide cell-to-cell variation
in Na⁺ would necessitate the sampling of a large number of
cells for group comparisons. While the role of high-reso-
lution nuclear magnetic resonance in the measurement of
Na⁺ of vascular muscle has not been established yet, it will
not obviate the problem of distinguishing between extra-
cellular and intracellular Na⁺ [52].

By chemical dissection (inulin space) of vascular
muscle, Villamil et al. [53, 54] found increased Na⁺ of the
carotid artery in dogs made hypertensive with long-term
intravenous infusion of angiotensin II and of the aorta of
rats with DOCA-salt hypertension [53, 54]. The authors
themselves acknowledged that Na⁺ in these experiments
may have been overestimated due to the poor penetration
of inulin into the extravascular structures. In the dog
carotid artery, sucrose space exceeded inulin space by
about 28%, but was less reproducible than inulin space.
There is no agreement as to which extracellular fluid
marker provides the true extracellular space. The cold Li⁺
exchange method was used to measure the Na⁺ of arterial
smooth muscle in rats with DOCA-salt and spontaneous
hypertension [5, 44, 55-57]. Distinctly different results
were obtained depending on the preparation of tissue
samples before measurements. The Na⁺ of arteries from
hypertensive rats was found to be increased when the
removed arteries were immersed at once in cold Li⁺-
substituted salt solution for washout of extracellular Na⁺.
When, after removal, the arteries were allowed to reach a
new steady state in vitro by prolonged incubation in
physiological salt solutions, the opposite was found,
namely a reduction in the Na⁺ of arteries from
hypertensive rats. The K⁺/Na⁺ ratio of fresh arteries
ranged between 3:1 and 5:1, and that of incubated arteries
was about 8:1. This is similar to what was found in freshly
isolated and in preincubated leucocytes (see above)
[30-38]. The Na⁺ of arteries from rats with DOCA-salt
and spontaneous hypertension was also measured by flux
analysis [58]. Flux analysis requires prolonged incubation
of tissues in physiological salt solutions; however, using
this methodology, no differences were found between the
Na⁺ of arteries from hypertensive and normotensive
control rats. It is unclear why the study of fresh arteries
and incubated arteries should yield such discrepant results. It is possible that in vivo, arteries are in a state of partial depolarization due to the stretch associated with the high intraluminal pressure. There is also evidence that the high Na⁺ of freshly removed arteries is in part the result of the trauma of dissection [5]. The rougher the handling of arteries, the higher is their measured Na⁺, indicating that Na⁺ is taken up by the cells during dissection. As to the differences detected between the Na⁺ of freshly removed arteries of hypertensive and normotensive rats, hypertension per se is not a factor because the Na⁺ of arteries from vessels of hypertensive and normotensive rats had doubled. Na⁺ accumulates intracellularly also when cells are deprived of serum in tissue culture [61]. The changes are rapid and dramatic: Na⁺ more than doubles by 1 h, and quadruples by 2 h. The uptake of Na⁺ is accompanied by the loss of K⁺, indicating increased membrane permeability to these ions. Na⁺→K⁺ pump activity does not change significantly during the first 2 h of serum depletion. Na⁺ also accumulates in arteries removed from experimental animals and placed in Krebs–Ringer solution [62]. Na⁺ uptake is accompanied by depletion of intracellular K⁺. It takes 90 min of incubation in Krebs–Ringer solution at 37°C before Na⁺ and K⁺ contents are restored to levels found in freshly excised arteries [62]. In this regard, leucocytes seem to behave like vascular muscle. Changes of temperature and separation of cells from plasma (or serum) would be expected to have a more pronounced effect on leucocytes than on erythrocytes because the Na⁺ permeability of the former (840 mmol min⁻¹ litre⁻¹ of cell water) is about 20 times greater than the latter (35 mmol min⁻¹ litre⁻¹ of cell water) [63–66]. This may explain why in the majority of studies the K⁺/Na⁺ ratio of erythrocytes is about 10:1 and that of leucocytes about 4:1. The influx of Na⁺ during cell preparation and the difference between the P Na⁺ of erythrocytes and leucocytes may also explain why increased leucocyte Na⁺ in hypertension has been a more consistent finding than the increased Na⁺ of erythrocytes.

**DISCUSSION**

How do we explain the inconsistencies in the measurements of Na⁺ in hypertension? We suggest that they are due to changes in Na⁺ during preparation of tissue specimens. In the course of measurement, cells undergo two drastic changes: one is cooling, and the other is the separation of cells from plasma. Both result in Na⁺ uptake. That the transmembrane Na⁺ gradient falls when the temperature is reduced has been known for a long time [46]. Cell shrinkage resulting in stimulation of inward-directed Na⁺→K⁺ co-transport may explain in part this temperature-dependent influx of Na⁺ [21]. Less well known is the fact that Na⁺ also accumulates intracellularly after separation of cells from plasma. This was documented in erythrocytes by high-resolution ²³Na-nuclear magnetic resonance techniques [52]. The authors were comparing the Na⁺ of erythrocytes derived from stroke-prone spontaneously hypertensive and Wistar–Kyoto normotensive rats. The initial Na⁺ of freshly isolated and washed erythrocytes, was low (approximately 5 mmol/litre of cell water) and increased progressively with prolonged incubation in isotonic salt solution at 37°C. While no statistically significant differences were detected between the two groups, the Na⁺ of erythrocytes from spontaneously hypertensive rats was somewhat lower during the first 2 h and somewhat higher during the last 4 h of incubation. By 6 h, the erythrocyte Na⁺ of rats had doubled. Na⁺ accumulates intracellularly as well as in normotensive and spontaneously hypertensive rats, which is not a factor because the Na⁺ of arteries from vessels of hypertensive and normotensive rats has doubled. Na⁺ accumulates intracellularly also when cells are deprived of serum in tissue culture [61]. The changes are rapid and dramatic: Na⁺ more than doubles by 1 h, and quadruples by 2 h. The uptake of Na⁺ is accompanied by the loss of K⁺, indicating increased membrane permeability to these ions. Na⁺→K⁺ pump activity does not change significantly during the first 2 h of serum depletion. Na⁺ also accumulates in arteries removed from experimental animals and placed in Krebs–Ringer solution [62]. Na⁺ uptake is accompanied by depletion of intracellular K⁺. It takes 90 min of incubation in Krebs–Ringer solution at 37°C before Na⁺ and K⁺ contents are restored to levels found in freshly excised arteries [62]. In this regard, leucocytes seem to behave like vascular muscle. Changes of temperature and separation of cells from plasma (or serum) would be expected to have a more pronounced effect on leucocytes than on erythrocytes because the Na⁺ permeability of the former (840 mmol min⁻¹ litre⁻¹ of cell water) is about 20 times greater than the latter (35 mmol min⁻¹ litre⁻¹ of cell water) [63–66]. This may explain why in the majority of studies the K⁺/Na⁺ ratio of erythrocytes is about 10:1 and that of leucocytes about 4:1. The influx of Na⁺ during cell preparation and the difference between the P Na⁺ of erythrocytes and leucocytes may also explain why increased leucocyte Na⁺ in hypertension has been a more consistent finding than the increased Na⁺ of erythrocytes. There is evidence for increased P Na⁺ of erythrocytes in hypertensive subjects and of erythrocytes, leucocytes and vascular smooth muscle cells in rats with spontaneous and DOCA-salt hypertension [34, 44, 56, 58, 64, 67–69]. Increased membrane P Na⁺ is demonstrated not only at 37°C but also at 16°C and 4°C [34, 70]. At 16°C, the degree of depolarization and the associated transmembrane movement of cations were greater in vascular muscle removed from spontaneously hypertensive rats than in vascular muscle from normotensive control rats [70]. Taken together, this means that during delays in processing and lengthy separation procedures Na⁺ may accumulate at a more rapid rate in cells derived from hypertensive subjects and experimental animals than in cells from normotensive controls. In other words, there is a good possibility that the finding of increased Na⁺ in hypertension is experimentally induced, and it is a reflection of increased membrane P Na⁺ rather than of increased Na⁺ in vivo. In vivo, the Na⁺→K⁺ pump appears to compensate for and, in some cases, may overcompensate for increased P Na⁺. For instance, we found a negative correlation between the reduction of erythrocyte Na⁺/K⁺ and the number of pump sites per erythrocyte in hypertensive subjects [24]. Reductions of erythrocyte Na⁺ in prednisone-treated subjects and in the Milan strain of hypertensive rats are accompanied by increased cation transport [20, 29]. Thus, under certain circumstances enhanced active Na⁺ extrusion may dominate the transmembrane Na⁺ balance. This suggestion was made by Friedman and
co-workers who found reduced steady-state Na, and increased \( P_{Na} \) of arteries removed from prehypertensive DOCA-salt-treated rats [57]. The same explanation was given by Engelhardt & Scholze [28] for their finding of reduced erythrocyte Na, in mild human hypertension.

What is the possible significance of an increased or reduced Na, of arteries in hypertension? As far as the resting membrane potential is concerned, the significance of Na, is probably minor. Wahlström [71] used the Goldman equation for diffusional potential:

\[
E_{\text{cell}} = \frac{RT}{zF} \ln \left( \frac{P_K[K^+]_o + P_{Na}[Na^+]_o + P_{Cl}[Cl^-]_o}{P_K[K^+]_i + P_{Na}[Na^+]_i + P_{Cl}[Cl^-]_i} \right)
\]

to calculate membrane potential from flux data obtained in the rat portal vein, a phasically contracting smooth muscle. The value of \(-42 \text{ mV}\) was in good agreement with the measured value [71]. The relative \( P_K, P_{Na}, \) and \( P_{Cl} \) were 1.0:0.034:0.816. From simple inspection of the Goldman equation, it is apparent that a 2-4 mmol increase or decrease in [Na+] has a negligible effect on the steady-state membrane potential. Similar observations were made in the phasic portal vein and the tonic main pulmonary artery of rabbits [46]. The steady-state (resting) membrane potential of vascular muscle appears to be determined by the permeabilities and distribution of \( K^+ \) and \( Cl^- \) and not Na+ [47]. In contrast, a potentially important role of Na, may be the regulation of Na+-Ca2+ countertransport, whereby small changes in Na, may have a large effect on intracellular Ca2+ concentration and tension development [2, 72]. The physiological significance of Na+-Ca2+ countertransport, however, has been questioned [73, 74]. While it may play a role under extreme conditions, under more physiological conditions vasoconstriction is mediated through a potential-dependent Ca2+ influx rather than through Na+-Ca2+ countertransport-dependent decrease in Ca2+ efflux [74].

While the ionic currents responsible for action potentials are not definitely known, Na+ appears to be the principal ion carrying the depolarizing current in spike-generating vascular muscle [47]. The role of Na+ in graded depolarization of the non-spike-generating muscle is uncertain [47]. More important than the possibility of a steady-state increase in Na, is the possibility that a small increase in \( P_{Na} \) may produce a major increase in the amplitude of the neurally or agonist-mediated action potential of spike-generating vascular muscle and, presumably, in the magnitude of the contractile response. In non-spike-generating vascular muscle, increased \( P_{Na} \) may facilitate the graded depolarization of plasma membrane and result in increased myogenic tone. In our attempts to understand the pathophysiology of hypertension, we should concentrate on measurements of Na+ fluxes during membrane depolarization. This, however, will be a more formidable task than the measurement of steady-state Na, in hypertension has proved to be.

In summary, the assumption that steady-state Na, in hypertension is increased is not justified at the present time. Findings of increased Na, of erythrocytes, leucocytes and vascular smooth muscle in hypertension may be due to increased membrane \( P_{Na} \) and the more rapid uptake of Na+ during tissue preparation than in normotensive controls. Recent evidence indicates that when measurements are taken within seconds of the withdrawal of blood, erythrocyte Na, is reduced, not increased, in human hypertension [23, 24]. For future studies, every attempt should be made to measure cell Na, in hypertension \textit{in vivo} or under experimental circumstances which closely resemble conditions \textit{in vivo}. Measurements should be made in the subject's or experimental animal's own plasma, as separation of cells from plasma results in the influx of Na+. Electron probe analysis of quick-frozen tissue makes measurement of Na, \textit{in vivo} under equilibrium conditions and during membrane depolarization possible and should be used to study the Na, of vascular muscle in hypertension.

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content in erythrocytes in essential hypertension. Hypertension, 8, 618-624.


