Sodium Shows No Mercy on the Nanomechanics of Endothelial Cells

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einberger et al1 defined sodium sensitivity and sodium resistance of blood pressure in 576 women and men by 2 separate techniques. Sodium-sensitive individuals were older, had lower renin values, and were more commonly black. Follow-up of these subjects for ≤20 years was revealing. Initial baseline measurements were associated with subjects who had died compared with subjects known to be alive: age at study, blood pressure, hypertension, sodium sensitivity, baseline renin levels, and body mass index. When survival curves were examined, normotensive salt-sensitive subjects aged ≥25 years when initially studied were found to have a cumulative mortality similar to that of hypertensive subjects, whereas salt-resistant normotensive subjects had increased survival.2 Diagnosing sodium sensitivity is tedious, involving at least several days of volume expansion and contraction or observing blood pressure responses to a low-salt diet. Furthermore, the mechanisms of the phenomenon are imperfectly explained.

The Oberleithner laboratory offers some refreshing new views on the subject. Their focus is on endothelial cells, which harbor 4 mechanically distinct compartments, namely the glyco
calyx, the cell cortex, the cytoplasm, and the nucleus.3 In addition, endothelial cells have a slightly modified version of the epithelial sodium channel (EnNaC) that has considerable regu
latory importance in terms of endothelial cell behavior (Figure). The group has focused on nanomechanics of endothelial cells because stiffness of single cellular compartments has a crucial affect on endothelial cell function, subsequent vascular stiffness, and has direct implications for sodium-dependent effects.

The endothelial glyco
calyx is a thick proteoglycan-rich layer on the luminal side of the cell and directly faces the flowing blood plasma.4 The intensely negatively charged heparin sulfate residues offer binding sites for sodium and presumably the glyco
calyx is the site of the endothelial cell sodium sensor. The glyco
calyx is subject to injury, shedding, collapse, and subsequent increased endothelial cell permeability to sodium and decreased barrier function. The cell cortex, primarily a cort
cical actin web, is closely bound to the cytoplasm via actin stress fibers and the microtubular system. Cortical stiffness is mainly determined by the status of the underlying cortical actin web. The EnNaC, similar to its counterpart in the kidney, regulates mineralocorticoid receptor function and is important to shear stress and mechanosensitivity.5 Fortunately, the EnNaC is susceptible to amiloride so that pharmacologi
cal probes are available. Finally, the cell nucleus enables the
cell to synthesize more components, notably glyco
calyx and EnNaC subunits. Endothelial cells can respond accordingly, but erythrocytes cannot, a fact that the Oberleithner group has also used to its advantage (described below).

The Oberleithner laboratory has at its disposal atomic force microscopy. Atomic force microscopy is a high-resolution type of scanning probe microscope, with demonstrated reso
lution on the order of fractions of a nanometer, >1000-fold better than the optical diffraction limit. The atomic force microscopy is one of the foremost tools for imaging, measur
ing, and manipulating matter at the nanoscale level. The information is gathered by feeling the surface with a mechanical probe. Piezoelectric elements that facilitate tiny but accurate and precise movements on electronic command enable precise scanning. In some variations, electric potentials can also be scanned using conducting cantilevers. In more advanced versions, currents can be passed through the tip to probe the electric conductivity or transport of the underlying surface.

Oberleithner et al6 used the atomic force microscopy as a nanosensor to measure stiffness of live endothelial cells incubated in culture medium at variable sodium concentrations from 135 to 150 mmol/L. They found that the cells got stiffer within minutes as the sodium concentration increased. If aldosterone was eliminated from the medium or if EnNaC was blocked with amiloride, the response was attenuated or eliminated altogether. Furthermore, the endothelial cells subjected to high sodium concentra
tions and aldosterone produced decidedly less nitric oxide, compared with control endothelial cells. The findings gave in
teresting insights into the salt-intake hypertension debate.

In the report by Paar et al7 in the current issue of Hypertension, the group focused on the effects of age. Aortic endothelial cells of young and old mice were studied under controlled conditions. Endothelial cells from old mice had more EnNaC than those from young mice, and the cortical stiffness was significantly increased (Figure). When sodium concentrations were increased from 135 to 150 mmol/L, the endothelial cells from old mice became stiffer than those from young mice. Spirinolactone, amiloride, and decreased EnNaC abundance prevented endothelial cell stiffening.

Does hypernatremia cause hypertension in vivo? Indeed, patients with primary aldosteronism tend to have serum
sodium concentrations on the high side. Friedman et al\(^8\) raised serum sodium concentrations in rats with peritoneal dialysis and observed an increase in blood pressure. Serum sodium concentrations are also influenced by hemodialysis. Suckling et al\(^9\) observed a significant positive relationship between change in plasma sodium concentration and change in systolic blood pressure. However, dialysis patients are tricky and numerous confounders could have contributed. Some authorities claim that dietary sodium intake influences the plasma concentration\(^10\), however, whether or not the current recommendations would do so is uncertain.

The effects on the endothelial cell cortex and the endothelial glycocalyx presumably occur within the entire vascular tree, including the kidney, to account for a generalized increase in peripheral vascular resistance and increased blood pressure. The effects should extend to lymph capillaries and wherever endothelial cells are to be found. It would be interesting to inspect the endothelial cells within the kidney of desert rodents or camels, which can concentrate their urine to the level of sea water and must surely be exposed to higher sodium concentrations than those tested by the authors. Furthermore, imaging of the endothelial glycocalyx (Figure) by perhaps scanning microscopy or other methods would be of interest to allow inspection of the disheveled glycocalyx.

The Oberleithner laboratory has also been busy at developing clinical tests to identify at-risk sodium-sensitive individuals.\(^4\) Their salt blood test relies on the status of the glycocalyx of erythrocytes. The test is based on the sodium-dependent erythrocyte \(\xi\) potential, a scientific term for electrokinetic potential in colloidal systems. Their second sodium sensitivity (salt-provocation) test is based on testing endothelial permeability in the presence or absence of amiloride when subjects are given 2 oral 5-g salt loads. The 2 sodium-sensitivity tests do exhibit a correlation. Developing tests is clinically tedious, a gold standard is required; sensitivity, specificity, and predictor values must be calculated. Nevertheless, these approaches exhibit courage in translation. Finally, the Oberleithner hypothesis is reminiscent of a second recent idea, also based on sodium binding to proteoglycans that could result in an altered microenvironment.\(^11\) This idea involves sodium storage in interstitium, particularly skin, and includes implications for sodium sensitivity. Lymphatic vessels also have endothelial cells, and ENaC is important in keratinocytes. These ideas are by no means mutually exclusive. In any event, sodium still generates excitement, even for this aging sodium-sensitive, hypertensive individual.

**Disclosures**

None.

**References**


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