Salt-sensing mechanisms in blood pressure regulation and hypertension

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Orlov SN, Mongin AA. Salt-sensing mechanisms in blood pressure regulation and hypertension. Am J Physiol Heart Circ Physiol 293: H2039–H2053, 2007. First published August 10, 2007; doi:10.1152/ajpheart.00325.2007.—High salt consumption contributes to the development of hypertension and is considered an independent risk factor for vascular remodeling, cardiac hypertrophy, and stroke incidence. In this review, we discuss the molecular origins of primary sensors involved in the phenomenon of salt sensitivity. Based on the analysis of literature data, we conclude that the kidneys and central nervous system (CNS) are two major sites for salt sensing via several distinct mechanisms: 1) [Cl\(^{-}\)] sensing in renal tubular fluids, primarily by Na\(^{+}\)-K\(^{+}\)-Cl\(^{-}\) cotransporter (NKCC) isoforms NKCC2B and NKCC2A, whose expression is mainly limited to macula densa cells; 2) [Na\(^{+}\)] sensing in cerebrospinal fluid (CSF) by a novel isoform of Na\(^{+}\) channels, Na\(_{\text{v}}\), expressed in subfornical organs; 3) sensing of CSF osmolality by mechanosensitive, nonselective cation channels (transient receptor potential vanilloid type 1 channels), expressed in neuronal cells of supraoptic and paraventricular nuclei; and 4) osmolality sensing by volume-regulated anion channels in glial cells of supraoptic and paraventricular nuclei. Such multiplicity of salt-sensing mechanisms likely explains the differential effects of Na\(^{+}\) and Cl\(^{-}\) loading on the long-term maintenance of elevated blood pressure that is documented in experimental models of salt-sensitive hypertension.

Numerous epidemiological and intervention studies have demonstrated a positive correlation between salt intake and elevated blood pressure in ~30% and 50% of hypertensive Whites and Blacks, respectively (38, 62, 168). It has also been shown that high salt intake provokes vascular remodeling and increases cardiac left ventricular mass as well as stroke incidence independently of blood pressure elevation (115, 126, 152). A priori, high salt intake may lead to the development of hypertension and cardiovascular complications by signals triggered by augmented extracellular Na\(^{+}\) (Na\(_{\text{e}}\)) concentration ([Na\(^{+}\)]\(_{\text{e}}\))

Relative changes of [K\(^{+}\)] in blood plasma are the largest compared with other major blood plasma ions in both physiological states and various pathologies. It is well known that Na\(^{+}\)-K\(^{+}\)-ATPase in skeletal muscle cells serves as a primary sensor for [K\(^{+}\)] in plasma and extracellular fluids. The Na\(^{+}\)-K\(^{+}\) pump in this tissue is particularly important because skeletal muscles constitute a main body store of intracellular K\(^{+}\). Half-maximal activation of α\(_{1}\), α\(_{2}\), and α\(_{3}\)-isoforms of Na\(^{+}\)-K\(^{+}\)-ATPase expressed in skeletal muscles occurs at extracellular K\(^{+}\) (K\(_{\text{e}}\)) concentrations ([K\(^{+}\)]\(_{\text{e}}\)) from 1 to 10 mM (15, 37, 59, 125), which is within the range detected in plasma (from 3.5 to 4.5 mM in healthy subjects to 2 and 7 mM in severe hypo- and hyperkalemia, respectively). In most cases, hypokalemia is a consequence of the augmented expression of skeletal muscle Na\(^{+}\)-K\(^{+}\)-ATPase caused by hypersecretion of aldosterone and other steroid hormones or by excessive activation of Na\(^{+}\)-K\(^{+}\)-ATPase by insulin, calcitonin gene-related peptide, or catecholamines (36).

Unlike [K\(^{+}\)], changes in plasma [Na\(^{+}\)] are relatively small and typically do not exceed 5% even with a sharp modulation of salt intake (44). For instance, mice with DOCA/salt-induced hypertension do not show any changes in plasma [Na\(^{+}\)] (188). After 4 wk of maintenance on a high-salt diet (8% vs. 0.5%), plasma [Na\(^{+}\)] in DS rats was increased by only 4% (52) and did not change in Sprague-Dawley rats (178). In both hypertensive and normotensive subjects, restriction of salt intake.
from 350 to 10 mmol/day decreased plasma [Na+] by only 2% (70). To our best knowledge, the highest impact of the high-salt diet on average [Na+] (154.3 ± 3.7 vs. 145.1 ± 3.4 mM in the control diet) was demonstrated in SHRs by de la Sierra and coworkers (42). Severe forms of hyponatremia caused by postoperative complications, pharmacological agents, acquired immune deficiency syndrome, and psychogenic polydipsia are out of the scope of this review and have been considered elsewhere (8).

Three hypothetical mechanisms of plasma [Na+] sensing involving different ion transporters may be proposed.

Mechanism 1. Transient elevation of plasma [Na+] may activate Na+ carriers leading to increases in intracellular Na+ (Na+i) concentration ([Na+]i) and modulation of cell functions via the activation of intracellular Na+ sensors (140). The list of relevant transporters includes Na+/K+-Cl- cotransporters (NKCCs), Na+/Cl- cotransporters (NCCs), Na+/H+ exchangers and (NHEs) as well as Na+o-coupled carriers mediating the symport of inorganic phosphate, bicarbonate, and small organic molecules. However, the EC50 values for Na+i of all major inwardly directed carriers are <30 mM (14, 31, 58, 84, 127, 143, 150, 153, 157). Therefore, the aforementioned Na+ carriers likely play a negligible role in [Na+]i regulation and sensing in the range of [Na+] detected in plasma (Fig. 1, curve 1).

Mechanism 2. Sensing of plasma [Na+] may be mediated by increases in [Na+]i due to changes in the inwardly directed Na+ current (INa-in) via Na+ channels. With the exception of Na+ channels, which are functionally important in the brain (see Na+ and Cl- Sensing in Organ-Specific Extracellular Fluids, CNS), Na+ permeability (PNa) of all known voltage-gated (Na+) and epithelial Na+ channels (ENaC) is not affected by modulation of [Na+]i. Thus, with the omission of Na+ channel-expressing cells of the brain, the INa-in value is in direct proportion to [Na+]o and follows the constant field equation:

\[ I_{Na-in} = P_{Na} zFE_{m} FRT ([Na+]_o \exp(-zFE_{m}/RT) / (1 - \exp(-zFE_{m}/RT)), \]

where the \( P_{Na} \) is the channel permeability for Na+, \( z \) is the charge of Na+, \( F \) is Faraday’s constant, \( R \) is the gas constants, and \( T \) is the absolute temperature. Therefore, even pathological changes in [Na+]o will limit changes in INa-in and [Na+]i to 5–10% (Fig. 1, curve 2). Moreover, even this negligible impact of “classic” Na+ channels on [Na+]i will be diminished due to feedback activation of Na+-K+-ATPase by Na+i (175).

Mechanism 3. [Na+]i modulation may affect membrane potential (Em) and E-m-coupled cellular functions. However, in an overwhelming number of cell types, the resting PNa of the plasma membrane is much lower than the permeability for K+ (PK) and Cl- (PCl). Moreover, even when \( P_{Na} > P_{K} \) or \( P_{Cl} \) and Em ~ Na+ potential = RT/F \( \ln([Na+]_o/[Na+]_i) \), and assuming that [Na+]i stays constant at ~10 mM, an extreme deviation of [Na+]i within 10% of its normal values leads to an Em alteration by only 2 mV.

Clinical handbooks have indicated that plasma Cl- levels in healthy subjects vary within the range from 98 to 108 mM. We failed to find any systematic analysis of this parameter in human pathologies. However, keeping in mind the principle of electroneutrality, the range of [Cl-] variation in plasma should be about the same as that reported for [Na+], i.e., <10% of mean physiological values. To the best of our knowledge, in this concentration range, [Cl-]o does not affect the PCl of any of the known Cl- channels. Half-maximal activation of inwardly directed Cl- carriers occurs at [Cl-]o <50 mM (56, 157), thus excluding their substantial contribution to plasma [Cl-] sensing.

Considered collectively, these data show that monovalent ion transporters are unlikely to be involved in sensing plasma [Na+] and [Cl-]. Therefore, two alternative mechanisms might be proposed. First, systems distinct from ion transporters may provide sensing of plasma [Na+] and [Cl-]. This is true in the case of extracellular Ca2+ concentration-sensing machinery, which is mediated by extracellular Ca2+ interactions with a member of the superfamily of G protein-coupled receptors predominantly expressed in parathyroid and renal epithelial cells (74). Alternatively, Na+ and Cl- handling occurs via their sensing in extracellular fluids distinct from blood plasma. Data supporting this latter hypothesis are considered below.

Na+ and Cl- Sensing in Organ-Specific Extracellular Fluids

Renal tubular and interstitial fluids. The kidneys reabsorb up to 90% of filtered salt via the coupled functioning of basolateral \( \alpha_{1}Na^{+}-K^{+}-ATPase \) and apical renal-specific NHE3 and NKCC2, which are expressed in the proximal tubules and thick ascending limb of Henle’s loop (TAHL), respectively (Fig. 2A). However, as considered below, salt-sensing and the regulation of salt excretion is provided mainly via tubuloglomerular feedback (TGF) in the juxtaglomerular apparatus (JGA) consisting of macula densa (MD), mesangial, granular, and vascular smooth muscle cells (VSMCs) and located in the cortical TAHL (Fig. 2B). Such a location is unique along the nephron and plays a key role in JGA function. Indeed, [NaCl] in renal fluid delivered to the MD is in the range of 20–60 mM. This is in sharp contrast to the proximal tubule, where concentrations of most solutes deviate modestly

![Fig. 1. Dependencies of the activity of electroneutral Na+ carriers (curve 1) and Na+ currents (I_{Na-in}) mediated by “classical” Na+ channels (curve 2) and Na+ channels (curve 3) on the extracellular Na+ ([Na+]o) concentration ([Na+]o). Values of maximal activities of ion carriers and I_{Na-in} at [Na+]o ~ 200 mM were taken as 100%. The range of [Na+] detected in plasma and cerebrospinal fluid (CSF) is shown by the black bar at the bottom.](http://ajpheart.physiology.org/)}
from those in plasma. Importantly, unlike the JGA, salt concentrations in the medullary segments of Henle’s loop are governed by osmotic gradients between mucosal fluid and the interstitium (for more details, see Ref. 26).

TGF is triggered immediately after an elevation of salt concentration in the tubular fluid delivered to the JGA and results in the contraction of VSMCs of afferent arterioles, thus causing increases in the exposure of proximal tubules to high-salt fluid via the attenuation of glomerular capillary pressure and the glomerular filtration rate (GFR) (Fig. 3A). As a consequence of this negative feedback loop, salt delivery to the distal nephron is kept within a narrow range. This process facilitates the fine adjustment of salt handling in the distal tubules by corticosteroids and peptide hormones, such as aldosterone and arginine vasopressin (AVP). Aldosterone via tubular mineralocorticoid receptors stimulates apical ENaCs and basolateral Na\(^+\)-K\(^+\) pumps, which results in the reabsorption of Na\(^+\) and osmotically obliged water into the blood and secretion of K\(^+\) into the urine (183). AVP, acting primarily via AVP receptor 2, promotes insertion of the aquaporin -2 water channel into apical membranes, also allowing water reabsorption (21, 90).

The second mechanism of JGA involvement in salt sensing results from strong downregulation of renin production by MD cells after several hours of exposure to apical NaCl in the range of \(-50\) to \(-60\) mM (47, 107) (Fig. 3A). This delayed mechanism produces the shift in the TGF operating point and changes the salt sensitivity of GFR regulation (Fig. 3B). The shift in the TGF operating point, in turn, influences salt reabsorption via decreased exposure of the proximal tubules to high salt, at the expense of its augmented delivery to distal tubules, which possess a lower salt reabsorption capacity. For more details, see Ref. 160.

Early studies by Wilcox and coworkers (128, 129, 194) demonstrated that changes in plasma [NaCl] affect renal blood flow in dogs mainly via modulation of plasma [Cl\(^-\)]. Later, it was shown that regulation of renal blood flow is mediated by activation of [Cl\(^-\)_o]-sensitive, osmolality-independent TGF (25, 161, 164). A key role of [Cl\(^-\)_o] rather than [Na\(^+\)_o] was also documented in studies on the salt-dependent release of renin, angiotensin II, and vasodilator prostaglandins (94, 107, 195, 196, 197). Cheng and coworkers (33) found a sharp elevation in the expression of cyclooxygenase (COX)-2 after exposure of cultured cortical TAHL cells to high-[Cl\(^-\)_o] solutions but not high-[Na\(^+\)_o] solutions. Further evidence for Cl\(^-\)_o-mediated signaling in the JGA came from identification of ion transporters involved in salt sensing by the MD.

The apical membrane of the MD, as well as that of TAHL epithelial cells, is abundant with NKCC2 (Fig. 4), which provides up to 80% of apical NaCl entry in these cells (11, 98, 130). In contrast to ubiquitously expressed NKCC1, NKCC2 has not been detected in any other cell types studied so far, including epithelial cells of proximal and distal tubules. Numerous studies have demonstrated that acute administration of furosemide, bumetanide, and other loop diuretics augments renin production, abolishes the TGF, and prevents the down-regulation of renin secretion triggered by high salt intake (65, 71, 160, 182, 198). Since these compounds inhibit NKCC1 and NKCC2 with the same potency (56), NKCC1\(^{-/-}\) mice were utilized for dissecting the relative contribution of NKCC isoforms in JGA function (32). NKCC1 deficiency causes approximately twofold elevation of plasma renin concentrations but slightly attenuates the renin production triggered by furosemide administration. These data strongly suggest that NKCC2 is a major component of salt-sensing machinery in the JGA. Possible complementary mechanisms underlying...
NKCC1 involvement in baseline renin production and TGF resetting are considered below.

Because of its stoichiometry (1Na⁺:1K⁺:2Cl⁻), [Cl⁻]₀ sensing by NKCC has an advantage compared with monovalent cations. Indeed, unlike Michael-Menten’s pattern for Na⁺ and K⁺ dependencies, binding of Cl⁻ to NKCC is a cooperative process with a Hill coefficient of 2 (156), providing high-efficiency regulation of carrier activity in the range of [Cl⁻]₀ existing in the JGA and close to the EC₅₀ value of the transporter (see Fig. 3C). Using isolated rabbit cortical TALH

Fig. 3. Salt-sensing mechanisms in the kidney. A: relationship among salt delivery, glomerular filtration rate (GFR), and renin production in the JGA (160). B: effect of long-term alteration of salt delivery to the JGA on the tubulogloberular feedback (TGF) set point. Dashed-dotted lines show TGF EC₅₀ values. C: dose-dependent effect of Na⁺ and Cl⁻ on NKCC activity in MD cells measured at an extracellular Cl⁻ (Clᵢ) concentration ([Clᵢ]₀) = 25 mM and [Na⁺]₀ = 25 mM, respectively (98). The range of salt concentration detected in distal tubules (107) is shown by the black bar on the bottom. The maximal values of GFR, renin production, and NKCC activity were taken as 100%.

Fig. 4. Role of the interstitial fluid [Cl⁻] and osmolality in the regulation of JGA function. Channel 1, Ca²⁺-sensitive Cl⁻ channels and other members of the Cl⁻ channel superfamily; channel 2, L-type Ca²⁺ channels. Nonidentified ion transporters in the basolateral surface of the macula densa are indicated by question marks. [Ca²⁺]₀, extracellular Ca²⁺ concentration; [K⁺]₀, extracellular K⁺ (Kᵢ) concentration; [Na⁺]₀, [Clᵢ], and [Kᵢ], intracellular Na⁺, Cl⁻, and K⁺ concentration, respectively; Eₘ, membrane potential.
with attached glomeruli, Laamerti and co-workers (98) determined that NKCC in MD cells was activated by Na\(^+\) and Cl\(^-\) with an EC\(_{50}\) of 1.0 and 17 mM, respectively. These values are very similar to the affinities for monovalent cations of all three alternatively spliced NKCC2 isoforms (NKCC2A, NKCC2B, and NKCC2F) cloned from rabbit (145) and mouse (83) cDNA libraries. These splice variants are differently distributed along the nephron: NKCC2B and NKCC2A are coexpressed in the MD, whereas NKCC2F is prevalent in the medullary TAHl (28, 145, 201). In Xenopus oocytes, EC\(_{50}\) values of mouse NKCC2B for K\(^+\), Na\(^+\), and Cl\(^-\) are 0.8, 3.0, and 12 mM, respectively (150). Very similar EC\(_{50}\) values for Na\(^+\) and Cl\(^-\) were obtained in a study of rabbit NKCC2B (58). In Xenopus oocytes injected with NKCC2A, EC\(_{50}\) values for Na\(^+\) are very close to those for NKCC2B, whereas the affinity for Cl\(^-\) is two- to fivefold lower (58, 150). Recently, NKCC2A/−/− and NKCC2B/−/− mice were generated by introduction of premature stop codons. Experiments performed in gene knockout animals demonstrated that NKCC2B and NKCC2A contribute to salt absorption and MD function in low and high [NaCl] ranges, respectively (136, 137). Importantly, the range of NKCC activation by Cl\(^-\) (Fig. 3C) is consistent with the range of modulation of TGF and renin production by apical NaCl (Fig. 3A) and was similar to the [Cl\(^-\)] in tubular fluid delivered to rat distal tubules (107) (Fig. 3C).

Downstream signaling events triggered by acute changes in apical [Cl\(^-\)] probably involve Cl\(^-\)-induced depolarization detected in the MD (147). Such depolarization causes activation of voltage-gated Ca\(^{2+}\) (Ca\(_v\)) channels and intracellular Ca\(^{2+}\)-mediated release of potent regulators of VSMC tone, such as nitric oxide (NO), eicosanoids, and others (11). Alternatively, MD function may be modulated by changes in cellular volume. Because Na\(^+\)-K\(^+\)-ATPase activity in MD cells is much lower than in other tubule segments (163), NKCC2 activation results in a sharp elevation of [Na\(^+\)], and osmotic load (10) that might be sufficient for swelling of MD cells and the release of ATP and other potent vasoconstrictors. The mechanism of ATP release in swollen cells has been extensively investigated (22, 132), and its role in the TGF has been subjected to detailed analysis in several recent reviews (11, 92, 93, 179).

It should be noted that the hypothesis that swelling-induced ATP release plays a key role in MD signaling is at odds with recent data on cell volume changes in MD cells obtained with fluorescent dyes (91). The authors of this study (91) found that MD cells swell only when apical [NaCl] is elevated isosmotically. In contrast, under physiological relevant conditions, when elevation of NaCl was accompanied by increases in medium osmolality, MD cells underwent rapid shrinkage, which was probably due to water efflux via the apical membrane. Interestingly, unlike the majority of other cell types studied so far (121), MD cells lack effective cell volume regulatory mechanisms (16, 91). Therefore, these cells behave as a perfect osmosensor, and the idea of their contribution to [Cl\(^-\)] sensing via osmosensitive mechanisms should be further explored.

Much less is known about the second arm of TGF resetting triggered by chronic exposure of the MD to high-salt fluid. A priori, attenuated renin production, which has been observed after chronic exposure of the JGA to high-salt fluid, should lead to relaxation of afferent arterioles via decreases in local levels of angiotensin II. However, the renin-angiotensin-aldosterone system may also affect renal function via modulation of salt reabsorption in the proximal tubule (183). Moreover, administration of inhibitors of COX and thromboxane synthase (30, 195), rather than angiotensin-converting enzyme inhibitor (195), affected the Cl\(^-\)-dependent modulation of renal blood flow in anesthetized dogs. Thomson and coworkers (176) found that sustained inhibition of proximal reabsorption with the carbonic anhydrase inhibitor benzolamide increased the levels of neuronal NO synthase in the MD and that inhibition of this enzyme abolished TGF resetting. In cultured MD-like cells, decreases in [NaCl] trigger phosphorylation of p38 MAPK and activation of COX-2 (33). The nature of upstream signals in this signaling cascade(s) and their relative contribution to TGF resetting are currently disputed (177) and have to be further investigated.

Bell and coworkers (10) demonstrated a fivefold elevation of [Na\(^+\)], in MD cells triggered by high-salt fluid, which should be accompanied by massive Cl\(^-\) uptake as a major charge-balancing anion. In all types of cells studied so far (156), including renal epithelial cells (3), [Cl\(^-\)] elevation from 5 to 80 mM inhibited NKCC activity by >10-fold. These data suggest that highly efficient regulation of JGA function by apical NaCl is impossible without rapid normalization of [Cl\(^-\)] via basolateral transporter(s). The outward-directed ion fluxes should affect the ionic composition of the interstitial fluid (Fig. 4). Indeed, increasing perfusion rates in Amphiuma kidneys lead to [Cl\(^-\)] increases in the intestinal fluid to values that are much higher than in plasma (146). Viewing these data collectively, we hypothesize that sustained changes in the ionic composition of interstitial fluid contribute to TGF resetting via modulation of function of VSMCs and mesangial cells exposed to this fluid (Fig. 2B). Experimental evidence in support of this hypothesis is considered below.

Since P\(_K\) and P\(_{Cl}\) values in VSMCs are about the same (34), transmembrane Cl\(^-\) gradients are important for the regulation of E\(_m\) in these cells. Brown and coworkers (27) were the first to suggest that NKCC1, the only NKCC isoform expressed in VSMCs, contributes to the maintenance of vascular tone via adjustment of the transmembrane Cl\(^-\) gradient above the values predicted by its Nernst equilibrium potential. Accordingly, inhibition of NKCC by bumetanide decreases [Cl\(^-\)]\(_o\), hyperpolarizes VSMCs, and abolishes activation of L-type Ca\(^{2+}\) channels by modest K\(^+\)-induced depolarization (7, 40). These data are consistent with the reduced SMC contraction demonstrated in the rat aorta and guinea pig urethra treated with bumetanide and subjected to modest depolarization, α-adrenergic agonists, or electrical stimulation (1, 2, 7, 96). Importantly, NKCC inhibitors suppress contraction in endothelium-denuded renal vasculature (190) as well as myogenic responses of the renal afferent arteriole in the in vitro perfused hydropnephrotic rat kidney (189). This preparation has no tubules, eliminating potential actions of diuretics on TGF via NKCC2 inhibition. In our early study (142), we observed that NKCC in VSMCs exhibited maximal activity at [K\(^+\)]\(_o\) and [Na\(^+\)]\(_o\) of ~5 and 40 mM, respectively, whereas Cl\(^-\) activated this carrier in the range from 10 to 140 mM. These data demonstrate that modulation of [Cl\(^-\)]\(_o\), rather than the concentration of monovalent cations in the interstitial fluid, may contribute to the NKCC1-mediated regulation of VSMC contraction.
A number of early studies proposed that because of the unique position of mesangial cells (Fig. 2B), their role in JGA function is not limited to secretion of cytokines, NO, and other vasoactive compounds but may additionally involve the direct action of these cells on the capillary surface area. The latter idea was supported by the similar morphology of VSMCs and mesangial cells as well as by observations of whole glomeruli and isolated mesangial cell contraction when cells were treated with vasoconstricting agents (171). Importantly, like in VSMCs, [Cl\textsuperscript{−}] in mesangial cells was higher than that predicted from the Nernst equilibrium potential, and these differences were completely abolished by NKCC inhibitors (111). Furthermore, Ca\textsuperscript{2+} influx via L-type Ca\textsubscript{v} channels causes mesangial cell contraction due to activation of Ca\textsuperscript{2+}-sensitive Cl\textsuperscript{−} channels (110, 171). Consistently, reductions in [Cl\textsuperscript{−}] inhibit the angiotensin II-induced contraction and intracellular Ca\textsuperscript{2+} responses (for a review, see Ref. 26). These data suggest that long-term modulation of [Cl\textsuperscript{−}] in the interstitial fluid may contribute to TGF resetting via altered mesangial cell function, side by side with the more recognized regulation of VSMC contraction.

Cerebrospinal fluid. In the central nervous system (CNS), neural tissue is isolated from the circulation by the blood-brain barrier. In addition to variety of other functions, the blood-brain barrier allows for setting local ion concentrations in the cerebrospinal fluid (CSF). [K\textsuperscript{+}] in the CSF is normally between 2.7 and 3.5 mM, which is substantially lower than [K\textsuperscript{+}] of ~4.5 mM found in plasma (170). An additional limitation that strongly influences the physiology of the neural tissue stems from the fact that the total volume of the brain’s intracellular and extracellular fluids is restricted by a rigid skull. It is well documented that neuronal activity affects CSF ion composition and causes changes in the volume of extracellular and intracellular compartments (170, 172). Thus, physiological levels of neuronal activity increase [K\textsuperscript{+}] by ~0.5 mM, as measured in a number of studies using ion-selective electrodes (169, 170) or up to 1.5 mM as deducted from depolarization of glial cells in the cat cortex (6). The authors of the latter study (6) have also found transient increases in the volume of glial cells with a magnitude of ~5%.

Much stronger changes in ionic composition and cell volume have been observed under pathological conditions. For example, anoxia/ischemia is accompanied by elevation of CSF [K\textsuperscript{+}] from ~3 to 70 mM and decreases in CSF [Na\textsuperscript{+}] from 140 to 60 mM and CSF [Cl\textsuperscript{−}] from 125 to 75 mM. In ischemia, the fractional volume of the extracellular space is reduced from ~20% to ~10%, while in the terminal anoxia, its fractional volume is decreased to 4% (172). Such volume changes themselves strongly affect CSF ionic composition (by concentrating the extracellular ions), thus complementing the effects of ionic shifts between extra- and intracellular compartments.

In mammals, the CNS-mediated regulation of water and salt intake is mainly governed via the release of the antidiuretic hormone AVP, which regulates water transport in the distal tubule (21, 23, 90). AVP is produced by the magnocellular neurosecretory cells of the hypothalamic supraoptic nucleus (SON) and paraventricular nucleus (PVN), which both project to the median eminence in the neurohypophysis (Fig. 5). However, CNS [Na\textsuperscript{+}] sensors are primarily located in the circumventricular organs, separate structures that line brain ventricles and consist of the subfornical organ (SFO), median preoptic nucleus (MnPO), and organum vasculosum lamina terminalis (OVLT) (Fig. 5). In these brain areas, the blood-brain barrier is partially open, allowing for direct sensing of the ionic composition of the plasma. OVLT and SFO neurons also project to the MnPO. The function of the PVN and SON is additionally affected by the osmolality of the CSF. OC, optic chiasm. [Modified with permission from Ref. 81.]

Fig. 5. Cellular mechanisms of salt sensing in the central nervous system (CNS). The magnocellular neurons, whose bodies are localized in the supraoptic nucleus (SON) and paraventricular nucleus (PVN), project their axons to the posterior lobe of the pituitary, where they secrete arginine vasopressin (AVP) to the blood. Magnocellular neurons receive synaptic inputs from central Na\textsuperscript{+}-sensitive neurons located in the organum vasculosum lamina terminalis (OVLT), subfornical organ (SFO), and median preoptic nucleus (MnPO). In these latter areas, the blood-brain barrier is partially open, allowing for direct sensing of the ionic composition of the plasma. The involvement of circumventricular organs in central [Na\textsuperscript{+}] and osmosensing has been confirmed in numerous lesion studies (23, 46). However, the molecular mechanisms responsible for [Na\textsuperscript{+}] and osmosensitivity in the CNS have been uncovered only recently.

Data obtained by Noda and coworkers strongly indicated that Na\textsubscript{v}-dependent responses in the CNS are triggered by activation of Na\textsubscript{v} channels, specialized Na\textsubscript{v}-sensitive channels. The Na\textsubscript{v} channel, formerly called Na\textsubscript{G}/SCL11 or voltage-sensitive Na\textsuperscript{+} channel (Na\textsubscript{v}) in rats, mice, and humans, respectively, possesses ~50% identity with other Na\textsubscript{v} channels cloned so far (73, 191). It is insensitive to modulation of $E_{m}$ from ~90 to ~40 mV as well as to the application of inhibitors of classic Na\textsubscript{v} channels and ENaCs, such as tetrodotoxin and amiloride (131). In contrast to classic Na\textsuperscript{+} channels, the Na\textsubscript{v} channel is sharply activated by elevations in [Na\textsuperscript{+}], from 145 to 170 mM with an EC\textsubscript{50} of 157 mM (Fig. 1, curve 3). Although Na\textsuperscript{+}-dependent responses are linked to changes in neuronal activity, an immunohistochemical study (192) has established that Na\textsubscript{v} channels are primarily expressed in the processes of glial cells in circumventricular organs of the brain, including the SFO, OVLT, MnPO, and posterior pituitary. How the [Na\textsuperscript{+}] signal is transferred from glial cells to neurons is not known.

In situ experiments in the SFO demonstrated that a 20-min elevation of CSF [NaCl] from 145 to 170 mM increased [Na\textsuperscript{+}], from 10 to 32 mM, and this effect was restricted to glial cells (192). Importantly, this signal was absent in the experiments.
employing an equimolar substitution of NaCl with choline chloride or mannitol (131). These data reveal a key role for Na\(^+\), rather than Cl\(^-\) or augmented osmolality, in salt sensing by Na\(_o\) channels. The role of Na\(_o\) channels in Na\(^+\)\_sensing was further confirmed in Na\(_o\) channel knockout mice (72, 131). In contrast to wild-type mice, Na\(_o\)\_null animals are constantly dehydrated, do not stop ingesting salt, and manifest abnormal salt intake behavior during NaCl microinjection into the cerebral ventricle. These abnormalities were rescued by a transduction of the Na\(_o\) gene into the SFO (72).

SFO and OVLT control electrical activity of the magnocellular neurons in the SON and PVN, both of which secrete AVP into the circulation (46, 81). Such regulation occurs via direct excitatory projections to the SON and PVN or indirectly via projections to the MnPO, a nucleus that also innervates both the SON and PVN (Fig. 5) (81). For a detailed discussion of related anatomic features and neurotransmitter systems, see also Ref. 43. Interestingly, the PVN and SON possess their own intrinsic [NaCl]-sensing mechanism, which adds additional complexity to the CNS regulation of salt intake and secretion. This mechanism involves sensing changes in CSF osmolality rather than alterations in [Na\(^-\)]\(_o\) (114, 134). The osmosensitivity of magnocellular neurons is largely mediated by mechanosensitive nonselective cation channels (135), which are activated by cell shrinkage and have been recently identified as a NH\(_2\)-terminal splice variant of the transient receptor potential vanilloid type 1 (TRPV1) channel (167).

TRPV channels belong to a large superfamily of proteins homologous to the Drosophila TRP channel. Mammalian TRP channels include >20 individual proteins, which are expressed in all tissues and regulated by various factors stimulating phosphatidylinositol signal transduction pathways (35). Biological effects of TRP channels are typically mediated by Ca\(^{2+}\) influx. Two TRP channels that expressed in the brain are osmosensitive. These include the above-mentioned NH\(_2\)-terminal splice variant of TRPV1, which is activated by cell shrinkage (high NaCl) and inactivated by cell swelling. The second osmosensitive TRP channel is the TRPV4 channel, which is activated by cell swelling (low NaCl) and inactivated by cell shrinkage (103, 167, 186). The TRPV4 channel is expressed in neuronal populations of the SFO and OVLT and has been reported to modulate osmosensing on a whole organism level (104, 118). However, the data on the impairment of water-electrolyte homeostasis in TRPV4 knockout animals are somewhat conflicting (102). Furthermore, swelling-induced activation of TRPV4 channels in the SFO and OVLT, and shrinkage-induced activation of TRPV1 channels in the SON and the PVN, should produce opposite effects on AVP release, which is difficult to reconcile from the physiological point of view.

Similar to [Na\(^+\)]\(_o\) sensing in the SFO and OVLT, the osmosensitivity of magnocellular neurons in the SON and PVN is strongly aided by adjacent glial cells. In the SON, a specialized population of astrocytes contain high cytoplasmic concentration of the amino sulfonic acid taurine (Fig. 6). Taurine release from glial cells is tonically regulated by physiologically relevant changes in CSF osmolality and is mediated by volume-sensitive anion channels (VRACs), the molecular identity of which remains unclear (24, 45, 81). Extracellular osmolality reductions as small as 5% and 7% increase tonic taurine release by ~10% and 25%, respectively, whereas a 15% osmolality reduction more than doubles the taurine release rate. Conversely, 10–15% increases in extracellular osmolality strongly reduce tonic taurine release (45). Once released, taurine potentially inhibits electrical activity and the release of AVP in magnocellular neurons via its action on neuronal glycine receptors (81, 82, 198). Interestingly, intracerebroventricular or systemic application of taurine reduces blood pressure in rats, and taurine has been reported to decrease blood pressure in hypertensive individuals (for a review, see Ref. 117).

It should be noted that the osmosensitivity of the glial release of amino acids may be strongly enhanced by extracellular ATP, bradykinin, AVP, and other physiologically relevant neurotransmitters and neuromodulators. In cultured cortical astrocytes, low micromolar levels of extracellular ATP cause severalfold increases in astrocytic glutamate and taurine release via VRACs, due to activation of P2Y metabotropic purinoceptors (119, 120). Downstream intracellular signaling events that mediate the increased osmosensitivity of glial VRACs include intracellular Ca\(^{2+}\) increases and activation of calmodulin, protein kinase C, and Ca\(^{2+}\)/calmodulin-dependent kinase II (120). Similar potentiation of osmosensitive taurine release has been found in cultured neurohypophysial astrocytes (pituicytes) challenged with bradykinin or AVP, suggesting a potential negative feedback for hormone secretion (155).

The salt-sensing (in the OVLT, SFO, and MnPO) and osmosensing (in the SON and PVN) mechanisms described above likely complement each other because of the extensive neural connections between these brain formations (Fig. 5). To present a complete picture, it must also be mentioned that magnocellular neurons in the SON and PVN receive electrical inputs from peripheral osmoreceptors (Figs. 5 and 6). These receptors predominantly reside in the mesenteric vasculature of the upper small intestine and hepatic portal vein. The effects of gastric infusion of salt or water on blood levels of AVP can be prevented by lesion of splanchnic and hepatic vagal nerves, which directly or indirectly innervate the SON and PVN (81). The mechanisms of hepatic osmosensing are not completely understood. An early study (123) using pharmacological inhibitors demonstrated the functional significance of the NKCC1 transporter in the stimulation of hepatic afferent nerve activity. Such an effect can be induced by hyperosmotic NaCl or NaHCO\(_3\), but not LiCl or mannitol, suggesting a primary role for [Na\(^+\)] in hepatoportal salt sensing (124).

In summary, the salt sensitivity in the brain is mediated by coordinated work of the Na\(^+\)-sensitive Na\(_o\) channels, osmosensitive nonselective cation TRPV1 channels, and osmosensitive anion VRACs. Na\(_o\) channels are primarily expressed in glial cells in the circumventricular organs, most importantly in the SFO, MnPO, and OVLT. TRPV1 channels are mechanosensitive channels of magnocellular neurons in the SON and PVN. VRACs are expressed and functionally important in glial cells in the SON, PVN, and pituitary (Figs. 5 and 6). Unlike other players in the brain [Na\(^+\)]/osmosensing, VRACs have not yet been identified on the molecular level (121, 133).

Cross Talk of Salt Sensors in Blood Pressure Regulation and Hypertension

Studies considered in the previous sections strongly indicated that the kidney and CNS are involved in salt sensing via at least three different mechanisms: [Cl\(^-\)], sensing in the tubular fluid and separate sensing of [Na\(^+\)]\(_o\) and osmolality in
the CSF. Such plurality of salt-sensing mechanisms allows for a more precise control of blood pressure and likely explains the differential effects of Na\(^{+}\) and Cl\(^{-}\) that have been reported in different animal models of hypertension (39, 41, 97, 109, 193, 199). One possible scheme of the cross talk between the CNS and kidney in salt sensing is presented in Fig. 7.

As mentioned above, TGF is triggered immediately after the delivery of high-salt fluid to the JGA and involves the Cl\(^{-}\)-mediated activation of NKCC2. This results in the attenuation of GFR and prolonged exposure of proximal tubules to high-salt fluid. Such a rapid mechanism for Cl\(^{-}\) sensing largely mediates the rise in salt reabsorption in the proximal tubule (Fig. 7). Long-term delivery of Cl\(^{-}\)-enriched tubular fluid to the MD leads to TGF resetting (Fig. 3B). TGF resetting results in augmented GFR and salt delivery to the distal tubules and collecting ducts, whose reabsorptive capacity is ~10-fold less than that of the proximal tubules. This feature of the distal tubule determines augmented salt excretion and partial normalization of plasma [NaCl], extracellular fluid volume, and blood pressure.

In contrast to the Cl\(^{-}\)-induced NKCC2-mediated signaling in the kidneys, salt sensing in the CNS involves Na\(^{+}\)-sensitive Na\(_{\text{ax}}\) channels in the SFO, MnPO, and OVLT and mechano/osmosensitive TRPV1 channels in the SON and PVN, along with osmosensitive VRACs in adjacent glial cells in the same nuclei, as schematically presented in Fig. 6. Overall, increases in [Na\(^{+}\)] and osmolarity in the above-mentioned brain areas trigger the release of AVP by magnocellular neurosecretory cells, whose bodies reside in the SON and PVN. AVP increases the permeability of the apical membrane of the distal tubule for water via insertion of aquaporin-2 water channels, thus facilitating the reabsorption of salt in this nephron segment. Excitation of neuronal cells also augments sympathetic outflow, which represents the most potent servo-mechanism for increased vascular tone and long-term maintenance of elevated blood pressure (Fig. 7). It should be stressed that the scheme presented in Fig. 7 focuses predominantly on the cross talk between the brain and kidney in salt sensing and, therefore, lacks many important details, including the feedback regulation of the JGA by blood pressure (162).
How the above-mentioned fine-tuning mechanisms are disrupted or deregulated in hypertension is not entirely clear. Nonetheless, a substantial amount of experimental data strongly suggest that abnormal $[\text{Cl}^-]_{\text{i}}$ sensing in the kidney and, perhaps, deficiencies in $[\text{Na}^+]_{\text{i}}$ sensing in the brain may provide a partial explanation for long-term increases in blood pressure in salt-sensitive forms of hypertension. Below, we briefly summarize some critical published data.

**Kidney.** In accordance with the Guyton concept, kidney resetting (i.e., normal salt excretion despite elevated blood pressure) is a major mechanism for the long-term maintenance of hypertension independently of its origin (64), including salt-sensitive hypertension (115). As considered in the preceding sections, altered activity of NKCC2 and NKCC1 is involved in the sensing of $[\text{Cl}^-]_{\text{i}}$ in JGA tubular and interstitial fluids, respectively, and represents the most prominent event contributing to kidney resetting. Alvarez-Guerra and Garay (5) demonstrated that NKCC2 activity is increased in the TAHL isolated from DS rats. Renal NKCC2 hyperactivity in DS rats was also confirmed by in vivo experiments showing an increased saliduretic response to bumetanide (5). Although NKCC2 activity may be caused by the overexpression of this carrier, to our best knowledge, there are no data on changes in the abundance of alternatively spliced NKCC2A, NKCC2B, and NKCC2F variants in the kidney of DS rats. On the contrary, in DS rats, increased NKCC activity in the TAHL was accompanied by a modest attenuation of the total immunoreactivity of NKCC2 protein (5). Alternatively, NKCC2 activation may be evoked by decreases in $[\text{Cl}^-]_{\text{i}}$, that would relieve negative regulation of NKCC activity by $\text{Cl}^-$. However, $[\text{Cl}^-]_{\text{i}}$, was increased rather than decreased in the TAHL from DS rats (5).

NKCC2 knockout mice showed severe polyuria and metabolic acidosis and died before weaning, making it impossible to use these animals for evaluation of the collective impact of NKCC2A, NKCC2B, and NKCC2F splice variants on the maintenance of blood pressure (173). Splice variant-specific NKCC2B and NKCC2A deletion did not significantly change mean blood pressure or the heart rate in knockout mice compared with wild-type animals (136, 137). Thus, additional experiments are needed to identify the mechanism of NKCC2 hyperactivity in salt-induced hypertension and its relation to altered NKCC2-mediated $[\text{Cl}^-]_{\text{o}}$ sensing in the MD and blood pressure regulation.

The augmented NKCC1 activity in hypertension was demonstrated in numerous early studies using erythrocytes and VSMCs from SHRs and rats of the Milan hypertensive strain (MHS) (for comprehensive reviews, see Refs. 139 and 151). Positive correlation between erythrocyte NKCC and blood pressure was found in F2 hybrids of MHS rats, SHRs, and normotensive rats (13, 95). Mice lacking NKCC1 have lower blood pressure (55, 116). Furthermore, an acute blood pressure-lowering effect of bumetanide was seen in wild-type mice but not in NKCC1−/− animals (57). All these data strongly suggest the involvement of NKCC1 in blood pressure regulation (for more details, see Ref. 138). Gordon and coworkers (60) were the first to propose that metabolic acidosis triggered by acute salt loading contributes to TGF in anesthetized dogs. This phenomenon is probably caused by inhibition of neuronal NO synthase, whose activity is strongly pH dependent, reaching a maximum at pH of ~7.7 (61). Indeed, Garvin and coworkers (106) demonstrated that an elevation of apical $[\text{NaCl}]_{\text{i}}$ causes alkalinization of the MD from pH 7.0 to 7.8 and elevation of local NO production. Importantly, apical application of dimethylamiloride augmented the TGF response and completely abolished the salt loading-evoked increments in intracellular pH and NO production in the MD (106, 187). These results point to the involvement of apical $[\text{NaCl}]_{\text{o}}$-induced NHE activation in TGF regulation. Numerous studies (139, 154) have found augmented activity of NHE1 in blood cells and VSMCs from SHRs and patients with essential hypertension. Increased NHE activity was also de-
tected in renal epithelial cells isolated from the SHR outer medulla (141), which may be due to an increased expression of basolateral NHE1 and apical NHE3 (89, 99). It should be stressed that, in contrast to proximal and distal tubules, MD cells express high levels of basolateral NHE4 and apical NHE2 isoforms (148). The activity of these isoforms in hypertension has not been yet explored. Superficially, NHE involvement in the TGF implies that [Na\(^+\)]\(_{\text{e}}\), rather than [Cl\(^-\)]\(_{\text{e}}\), serves as the primary signal, which is at odds with the bulk of the data discussed in this review. This, however, may not necessarily be the case, as [Cl\(^-\)]\(_{\text{e}}\) may regulate NHE activity indirectly via changes in intracellular [HCO\(_3\)]\(_{\text{e}}\) and intracellular pH (140). Furthermore, the direct effects of [Cl\(^-\)] on NHE have also been reported for an NHE isoform cloned from the rat distal colon (158).

TGF resetting occurs in the response to chronic exposure of the JGA to high-salt fluid. This suggests that, along with changes in NKCC2 and NKCC1 activity, salt-sensitive hypertension may be caused by modified activity of NCCs and ENaCs, both of which contribute to salt reabsorption in the distal tubules (Fig. 2A). This hypothesis is consistent with data on gain or loss of function mutations in these carriers or their major regulators found in the Mendelian forms of hyper- and hypotension (for comprehensive reviews, see Refs. 56, 105, and 115). Further studies are needed to explore the relative contribution of altered [Cl\(^-\)]\(_{\text{e}}\), sensing by NKCC2 and NKCC1 channels in the JGA, weighed against altered salt reabsorption in the distal tubule by NCCs and ENaCs, in the pathogenesis of salt-induced hypertension.

The downstream events contributing to the development of salt-induced hypertension likely involve elevated secretion of endogenous cardiotonic steroids, which are analogous to plant glycoside ouabain and amphibian bufodienolides and act as very potent inhibitors of the Na\(^+\)-K\(^+\) pump (17, 67, 68). Such a hypothesis is strongly supported by the substantial amount of experimental and clinical data collected by several laboratories. First, endogenous cardiotonic steroids and endogenous ouabain, as well as several other ouabain-like compounds, were purified from human plasma by Hamlyn and coworkers (69, 108). These findings were soon followed by the identification of human bufodienolide by Bagrov et al. (9). Endogenous cardiotonic steroids are synthesized mainly by the adrenal cortex in a synthetic pathway shared with aldosterone (68, 100). Their plasma content is strongly elevated in ~50% patients with essential hypertension, and their levels positively correlate with blood pressure (113, 149). The association between levels of endogenous ouabain-like compounds and hypertension is supported by the findings that long-term delivery of low dosages of ouabain causes sustained elevation of blood pressure in rats (112) and mice (49). Hypertensive actions of ouabain and its endogenous analogs are mediated by tonic inhibition of the \(\alpha\)-isoform of the Na\(^+\)-K\(^+\) pump, prompting changes in the transmembrane [Na\(^+\)] gradient and elevated Ca\(^2+\) uptake via NCX1 (49, 85, 202). As a result, cardiotonic steroids induce long-term increases in myogenic tone and vascular resistance. For more detailed coverage of this topic and its therapeutic applications, see the recent excellent overview by Hamilton and Blaustein (68).

It is important to stress that the actions of endogenous cardiotonic steroids are not restricted to the vasculature. Sub-nanomolar concentrations of endogenous ouabain and other ouabain-like compounds stimulate renal hypertrophy (54) and have potent actions in the brain (for details, see below). Since the effects of exogenous and endogenous cardiotonic steroids on blood pressure take at least 1–2 wk to develop, they are secondary to more immediate [Na\(^+\)]- and [Cl\(^-\)]-sensing mechanisms, which are discussed in this review. It is not entirely clear what causes the elevated production of endogenous cardiotonic steroids in animal models of salt-dependent hypertension and in humans (66, 68). In vivo and in vitro data have suggested that such production is triggered by acute plasma volume expansion and may be related to elevated levels of ACTH and angiotensin II (51, 144, 166).

CNS. The understanding of the role of CNS-related mechanisms of salt sensing in the pathogenesis of hypertension is greatly complicated by the existence of multiple, and partially overlapping, mechanisms of blood pressure control in the mammalian brain. In addition to the CNS salt sensing machinery that has been described in previous sections, the CNS (brain and spinal cord) has numerous neural inputs responding to changes in arterial pressure (arterial volume receptors) and peripheral changes in osmolarity (hepatic and gut osmoreceptors). Two brain nuclei, the rostral ventrolateral medulla and nucleus of solitary tract, are responsible for receiving these inputs and provide coordinated control of blood pressure together with the already-mentioned hypothalamic SON and PVN. This sympathetic control of blood pressure has been analyzed in a recent comprehensive review by Guyenet (63). Here, we limit our discussion to the pathological significance of “local” salt-sensitive events.

Leenen and coworkers (29, 75) were the first to propose that blood pressure elevation in animal models of hypertension is triggered by augmented salt delivery to the CNS. This hypothesis is supported by the following observations. First, in DS rats and SHRs, a high-salt diet augments CSF [Na\(^+\)] from 147 to 152 mM (79), and approximately the same CSF [Na\(^+\)] elevation was found in hypertensive subjects (88). Second, the long-term changes in CSF [Na\(^+\)] found in DS rats and SHRs fed a high-salt diet have not been detected in Dahl salt-resistant (DR) rats and normotensive Wistar rats (79). Third, the elevation in CSF [Na\(^+\)] precedes the development of salt-dependent hypertension (79). Finally, in DS but not DR rats, intracerebroventricular administration of NaCl increases blood pressure and sympathetic outflow (76, 80, 86, 184).

As stated in Na\(^+\) and Cl\(^-\)-Sensing in Organ-Specific Extracellular Fluids, CNS, increases in CNS [Na\(^+\)] are primarily detected by Na\(_{\text{v}}\) channels in glial cells located in the circumventricular organs. The [Na\(^+\)]\(_{\text{i}}\) signal in glial cells is then translated to increased activity of local glutamatergic neurons. The precise mechanism of such intercellular communication is presently unknown. As indicated in Fig. 7, the following possibilities may be considered: 1) transient increases in [K\(^+\)]\(_{\text{i}}\), due to depolarization of glial cells, 2) local overproduction of cardiotonic steroids (77, 181), or 3) increases in local renin-angiotensin II signaling (44, 122). It is hard to envision that [Na\(^+\)]\(_{\text{i}}\) signaling may produce long-lasting changes in [K\(^+\)]\(_{\text{i}}\). An alternative mechanism for glial regulation of neuronal activity may involve the expression and release of intercellular signaling molecules via a Na\(^+\)-ATPase-mediated mechanism of excitation-transcription coupling (174). Indeed, salt delivery (131) as well as chronic administration of Na\(^+\)-K\(^+\)-ATPase inhibitors (185) lead to a sharp activation of the expression of
c-fos and other early response genes in several circumventricular
compartment, including the SFO. As a working hypothesis, we propose that Na\(^+\) -mediated gene expression triggered by Na\(_{\text{a}}\) channel activation in glial cells may lead to the local production of endogenous cardiotoic steroids, such as ouabain and marinobufagenin, as seen in hypertensive rat strains after salt loading (18, 48, 76, 165, 53, 101). Endogenous ouabain-immunoreactive neurons are particularly abundant in the PVN and SON (200). High local levels of cardiotoic steroids may suppress the activity of neuronal Na\(^+\)-K\(^+\)-ATPase, causing long-lasting shifts in neuronal excitability and blood pressure. Local administration of neutralizing antidiotxin antibodies (Digibind) into the lateral ventricle lower the blood pressure in SHRs kept on a high-sodium diet (78).

Local CNS changes in renin-angiotensin production have also been implicated in the development of hypertension. Targeted to glia expression of angiotensinogen causes elevated blood pressure in transgenic mice (122). Interestingly, several reports (50, 76, 180) have suggested extensive interactions between endogenous ouabain-like substances and the renin-angiotensin system in the brain and proposed a key role for such regulation in CNS salt sensing. These and other mechanisms of normal and pathological salt sensing in the brain are awaiting further experimental exploration.

Even with the limited information that presently available, it is clear that several brain structures are critically important for normal and pathological salt sensing. Pathological changes in hypothalamic function have to be integrated into our current understanding of salt-sensitive hypertension. One potential hypothesis incorporating both hypothalamic changes and impairment of sodium excretion in the kidney has been put forward by de Wardener (43). According to his model, initial changes in kidney salt retention cause a persistent decrease in venous compliance; the increased venous pressure is then communicated to the hypothalamus via cardiopulmonary afferents. This resets hypothalamic neuronal activity and, via modulation of of midbrain medulla activity, causes the increased sympathetic activity and persistent arteriolar constriction that underlie elevated blood pressure. Nonetheless, as both blood pressure and hypothalamic function are sensitive to local changes in sodium concentration and osmolality within the hypothalamus, this strongly suggests the existence of brain-specific mechanisms responsible for the long-term maintenance of (elevated) blood pressure.

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**REFERENCES**

30. Buxton RL, Watson 10.220.33.5 on October 23, 2017 http://ajpheart.physiology.org/ Downloaded from


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