Vasopressin regulation of blood pressure and volume: findings from V1a receptor–deficient mice

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[Arg[8]]-vasopressin (AVP) has several functions via its three distinct receptors, V1a, V1b, and V2. The V1a vasopressin receptor (V1aR) is expressed in blood vessels and involved in vascular contraction. Recently, we generated V1a receptor–deficient ($V1aR^{-/-}$) mice and found that they were hypotensive. In addition, $V1aR^{-/-}$ mice exhibited (1) blunted AVP-induced vasopressor response, (2) impaired arterial baroreceptor reflex, (3) decreased sympathetic nerve activity, and (4) decreased blood volume, all of which could contribute to the observed hypotension. In relation to their decreased blood volume, $V1aR^{-/-}$ mice had decreased plasma aldosterone levels, which could result not only from decreased activity of the renin–angiotensin system (RAS), but also from impaired AVP-stimulated aldosterone release in the adrenal glands. V1aR was found to specifically co-express at the macula densa cells with cyclooxygenase (COX)-2 and with neuronal nitric oxide synthase, which produces potent stimulators of renin, PGE_2, and NO. The expression levels of renin, COX-2, and nNOS were significantly decreased in $V1aR^{-/-}$ mice, which led to the suppression of RAS activity and consequent decreases in aldosterone and blood volume. Furthermore, V1aR is also expressed in collecting duct cells and involved in regulating water reabsorption by affecting V2/aquaporin 2 function. Thus, AVP regulates blood pressure and volume via V1aR by exerting diverse functions *in vivo*.

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AVP RECEPTORS AND KNOCKOUT MICE

The neurohypophyseal peptide [Arg[8]]-vasopressin (AVP) is involved in diverse functions including regulation of body fluid homeostasis, vasoconstriction, and adrenocorticotropic hormone release. These physiological effects are mediated by three subtypes of AVP receptors, designated V1a, V1b, and V2.[1] The V1a vasopressin receptor (V1aR) is widely expressed, whereas the V1b receptor (V1bR) is specifically expressed in the anterior pituitary glands and pancreatic islets; the V2 receptor (V2R) is predominantly expressed in the kidneys.[1,2] V1aR mediates vascular contraction, cellular proliferation, platelet aggregation, glycogenolysis, lipid metabolism, protein catabolism, and glucose tolerance.[1,2] V1bR mediates adrenocorticotropic hormone and insulin release.[2] Both V1aR and V1bR couple to the Gq protein and act through phosphatidylinositol hydrolysis to mobilize intercellular Ca^{2+}.[1] V2R is expressed in the thick ascending limbs (TAL) of Henle's loop and the collecting ducts in the kidney. Through V2R, AVP stimulates Gs protein and adenylate cyclase to increase cellular cAMP, which stimulates the translocation of aquaporin (AQP)-2 in the collecting duct, and thereby increases water resorption.[1] As V1aR and V2R are both detected in the juxtaglomerular apparatus, the TAL, and the collecting duct, both could be involved in regulating body fluid homeostasis.[1,3]

Recently, we generated mice deficient in either V1a ($V1aR^{-/-}$) or V1b ($V1bR^{-/-}$). As AVP exerts its various actions through V1aR and V1bR, as well as through V2R, mutant mice deficient in V1aR or V1bR exhibit a variety of phenotypes. Although $V1bR^{-/-}$ mice exhibited altered hypothalamic–pituitary–adrenal axis activity,[2] enhanced insulin sensitivity,[2] and altered psychological behavior,[4] $V1aR^{-/-}$ mice also exhibited alterations in endocrine activity, metabolism, and feeding and social behavior.[2,5,6] In addition, $V1aR^{-/-}$ mice showed hypotension accompanied with decreased plasma volume.[5,7]

In this review, we suggest possible mechanism(s) by which AVP/V1aR may regulate blood pressure based on our studies with $V1aR^{-/-}$ mice.

BLOOD PRESSURE IN $V1aR^{-/-}$ MICE

Blood pressure is regulated by many vasoactive factors such as epinephrine.[8] Among them, AVP is one of the most potent vasoconstrictors and is known to affect blood pressure by regulating vascular tonus and body fluid through V1aR and/or V2R.[1] Yet the role of AVP in blood pressure maintenance is ill-defined, as the cardiovascular effects of AVP are complex. We studied the role of V1aR by observing V1aR$^{-/-}$ mice, and found that the mutant mice are hypotensive.[7] Although vascular V1aR is well recognized, the hypotensive phenotype of V1aR$^{-/-}$ mice was unexpected, because previous pharmacological studies with selective V1aR antagonists showed that blocking V1aR did not induce any significant change in basal blood pressure level.[9] Therefore, participation of vascular V1aR in maintaining peripheral vascular tonus is unexpectedly small or nonexistent in normal animals and humans at resting state. To elucidate the possible mechanism(s) by which AVP/V1aR regulates blood pressure, we analyzed V1aR$^{-/-}$ mice and found several changes that could affect blood pressure homeostasis.

Blunted AVP-induced vasopressor response

V1aR$^{-/-}$ mice exhibited remarkable hypotension without a significant change in heart rate during rest.[7] An echocardiogram study revealed no difference in cardiac function between V1aR$^{-/-}$ and wild-type (WT) mice.[7] As AVP is a potent vasoconstrictor, pressor responses to AVP were assessed in perfused mesenteric arterial beds. Whereas isolated perfused mesenteric arterial beds from WT and V1aR$^{-/-}$ mice exhibited comparable responses to KCl, the pressor response to AVP seen in WT mice was absent in V1aR$^{-/-}$ mice, indicating that the AVP-induced vascular contraction was caused primarily through V1aR.[7] In addition to this *in vitro* finding, *in vivo* AVP-induced pressor response was abolished in non-anesthetized V1aR$^{-/-}$ mice, which seemed at first to imply that a blunted blood pressure response to AVP could cause the hypotension. However, vasodilatation and depressor responses were induced in V1aR$^{-/-}$ arterial beds and non-anesthetized animals, respectively, by high concentrations of AVP far exceeding the physiological range.[1,7,10] Considering our finding and other reports on V1aR antagonists, it seems that other cause(s), rather than the blunted AVP-induced pressor response, could contribute to the decreased blood pressure observed in V1aR$^{-/-}$ mice.

Impaired arterial baroreceptor reflex

Functional relationships between blood pressure and reflex changes in heart rate were examined in non-anesthetized mice. We found that baroreceptor reflexes in V1aR$^{-/-}$ mice were significantly impaired. When blood pressure was increased by phenylephrine, an α1 adrenergic receptor agonist, or decreased by nitrates, markedly attenuated heart rate changes were evident in V1aR$^{-/-}$ mice and in WT mice treated with a selective V1aR blocker, although blood pressure changed by similar amounts in the mutants and the control mice.[7] Our observation was in line with the attenuated baroreflex sensitivity seen in AVP-deficient rats, and indicates involvement of V1aR in that reflex.[11] In addition, the bradycardia induced by electrical stimulation of the hemilateral vagus nerve, which was exposed and cut at the cervical level, was markedly attenuated in V1aR$^{-/-}$ mice, suggesting that their central baroreflex arc is impaired.[12] When V1aR expression in the nucleus of the solitary tract where the vagal afferents terminate was examined through *in situ* hybridization analysis, V1aR was highly expressed in the nucleus of the solitary tract and area postrema in WT mice, but not in V1aR$^{-/-}$. Thus, V1aR in the central nervous system could have a crucial role in maintaining the baroreflex control of heart rate and blood pressure homeostasis.

Decreased sympathetic nerve activity in central nervous system

Although previous pharmacological studies with selective V1aR antagonists did not induce any significant change in basal blood pressure level, such drugs can exert a hypotensive effect in hypertensive animals.[9,13] This suggests that the AVP/V1aR signal could be involved in developing or maintaining hypertension. To clarify the underlying mechanisms, we generated a salt-induced hypertension model. In WT mice, blood pressure was significantly increased during salt loading after subtotal nephrectomy. In V1aR$^{-/-}$ mice, in contrast, blood pressure increased during salt loading, but the final blood pressure after salt loading was significantly lower, indicating that V1aR deficiency leads to resistance to salt-induced hypertension (Koshimizu T *et al.*, manuscript in preparation). Plasma norepinephrine levels were increased by the salt loading in both WT and V1aR$^{-/-}$ mice, but the increase in norepinephrine was smaller in V1aR$^{-/-}$ mice, suggesting that sympathetic nerve activity was suppressed in V1aR$^{-/-}$ mice. Furthermore, V1aR antagonists markedly lowered the blood pressure in salt-loaded WT mice, but not in control WT mice or in salt-loaded or control V1aR$^{-/-}$ mice. Related to the salt-loaded hypertension model are several previous studies involving crosses of genetically hypertensive rat strains with vasopressin-deficient Brattleboro rats, intended to evaluate the importance of AVP in the development of hypertension. These studies revealed that genetic background contributes to salt- and water-retention ability.[14,15] SHRs (stroke-prone spontaneously hypertensive rats) have normal extracellular fluid volumes and become hypertensive on salt loading,[16,17] whereas New Zealand genetically hypertensive (NZGH) rats have reduced extracellular fluid volumes and are insensitive to salt loading.[18,19] When crossed with Brattleboro rats, vasopressin-deficient SHR developed full hypertension,[14] whereas the blood pressures of vasopressin-deficient NZGH rats were lower than those of the original NZGH strain.[15] It seems that the blood pressure phenotype of V1aR$^{-/-}$ mice is more similar to that of NZGH than to that of SHR, because AVP action is indispensable for the basal blood pressure seen in V1aR$^{-/-}$ mice, as it is for the hypertensive response seen in NZGH rats.

We also examined the role of central V1aR in blood pressure control by administering AVP into the cerebral