Vasopressin: Mechanisms of action on the vasculature in health and in septic shock

Lucinda K. Barrett, MA, MBBS, MRCP; Mervyn Singer, MBBS, MD, FRCP; Lucie H. Clapp, PhD

LEARNING OBJECTIVES
On completion of this article, the reader should be able to:
1. Explain the effects of vasopressin on healthy patients.
2. Describe the effects of vasopressin in patients with septic shock.
3. Use this information in the clinical setting.

Dr. Singer has disclosed that he is/was the recipient of grant/funds from The Medical Research Council UK and that he is a consultant for Ferring. Dr. Clapp has disclosed that she is recipient of grant funds from The Medical Research Council.

Lippincott CME Institute, Inc., has identified and resolved all faculty conflicts of interest regarding this educational activity. Visit the Critical Care Medicine Web site (www.ccmjournal.org) for information on obtaining continuing medical education credit.

Background: Vasopressin is essential for cardiovascular homeostasis, acting via the kidney to regulate water resorption, on the vasculature to regulate smooth muscle tone, and as a central neurotransmitter, modulating brainstem autonomic function. Although it is released in response to stress or shock states, a relative deficiency of vasopressin has been found in prolonged vasodilatory shock, such as is seen in severe sepsis. In this circumstance, exogenous vasopressin has marked vasopressor effects, even at doses that would not affect blood pressure in healthy individuals. These two findings provide the rationale for the use of vasopressin in the treatment of septic shock. However, despite considerable research attention, the mechanisms for vasopressin deficiency and hypersensitivity in vasodilatory shock remain unclear.

Objective: To summarize vasopressin’s synthesis, physiologic roles, and regulation and then review the literature describing its vascular receptors and downstream signaling pathways. A discussion of potential mechanisms underlying vasopressin hypersensitivity in septic shock follows, with reference to relevant clinical, in vivo, and in vitro experimental evidence.

Data Source: Search of the PubMed database (keywords: vasopressin and receptors and/or sepsis or septic shock) for articles published in English before May 2006 and manual review of article bibliographies.

Data Synthesis and Conclusions: The pathophysiologic mechanism underlying vasopressin hypersensitivity in septic shock is probably multifactorial. It is doubtful that this phenomenon is merely the consequence of replacing a deficiency. Changes in vascular receptors or their signaling and/or interactions between vasopressin, nitric oxide, and adenosine triphosphate-dependent potassium channels are likely to be relevant. Further translational research is required to improve our understanding and direct appropriate educated clinical use of vasopressin. (Crit Care Med 2007; 35:33–40)

Key Words: vasopressin; septic shock; vasopressor agents; receptors; nitric oxide; potassium channels

Vasopressin (antidiuretic hormone) is a nonapeptide hormone synthesized in the magnocellular neurons of the paraventricular and supraoptic nuclei of the hypothalamus. Hormone precursors migrate via the supraoptic-hypophyseal tract to the posterior pituitary gland, where they are stored in neurosecretory vesicles (1). Under normal conditions, circulating levels are maintained at around 2 pg/mL (10^{-12} M) (1, 2). Only 10–20% of the hormone within the posterior pituitary can be rapidly released, and with sustained stimulation this occurs at a greatly reduced rate (1). Vasopressin is rapidly metabolized by liver and kidney vasopressinases and has a half-life of 10–35 mins (1).

Regulation of vasopressin release is complex. In health, secretion is primarily governed by changes in serum osmolarity (osmoregulation). This system is highly sensitive, such that a small (2%) increase in osmolarity is reversed by the antidiuretic effect of a small (~5 pg/mL) increase in vasopressin (2). In contrast, baroregulation of vasopressin secretion only plays a significant role in the context of a >10% decrease in blood pressure. Hormone levels can then increase more than ten-fold to help restore normotension, largely via vasoconstriction (2).

Vasopressin release is affected by other hormones. At low concentrations, catecholamines tend to exert stimulatory ef-
fects via central α₁ receptors but at higher levels may inhibit vasopressin release via α₂ and β receptors (3, 4). Secretion of vasopressin also stimulates release of adrenocorticotropic hormone from the anterior pituitary, with consequent negative feedback of glucocorticoids on the posterior pituitary (2). Additional factors are important in critical illness. Hypoxia and acidosis stimulate carotid body chemoreceptors to increase vasopressin release (1). Furthermore, both endotoxin and cytokines enhance vasopressin production (2), whereas nitric oxide (NO) plays a mainly inhibitory neuromodulating role on its secretion (5).

The actions of vasopressin are mediated via G protein-coupled receptors, classified by virtue of their location and second messenger pathways into V₁ (or V₁ₐ), V₂, and V₃ (formerly V₁₉) receptors (6). In addition, vasopressin has equal affinity with oxytocin for oxytocin receptors (OTRs) and may exert some of its actions via this route (7).

**V₁ Receptors (V₁Rs).** V₁Rs are found mainly on vascular smooth muscle in the systemic, splanchnic, renal, and coronary circulations. They are coupled through Gₛ to phospholipase C (PLC), and their activation produces vasoconstriction via the elevation of intracellular calcium (Ca²⁺) (Figs. 1 and 2). The emptying of stores within the sarcoplasmic reticulum transiently increases cytoplasmic Ca²⁺, whereas a sustained increase is produced by influx of extracellular Ca²⁺ (8, 9). The pathways leading to vasopressin-induced extracellular calcium entry are complex (Fig. 1). Store-operated channels probably play a minor role compared with voltage-gated calcium channels and receptor-operated channels (10, 11). Voltage-gated calcium channels are activated indirectly by cell membrane depolarization or directly by protein kinase C (PKC) (12) (Fig. 1). The opening of receptor-operated channels is G protein-dependent via PLC and its downstream second messenger systems. To date, the best characterized role of the V₁R is in the secretion of adrenocorticotropic hormone, which appears to be mediated via the activation of PKC (7).

**Oxytocin Receptors (OTRs).** Like V₁Rs, OTRs are coupled to PLC, the metabolism of phosphoinositides, and the consequent elevation of intracellular calcium (7). In myometrial and mammary myoepithelial cells, OTR stimulation produces smooth muscle contraction (7), and this may also occur in vascular smooth muscle (17, 18) (Fig. 2). In addition, OTRs are highly expressed in the vascular endothelium (19), where an increase in intracellular Ca²⁺ activates constitutive endothelial NO synthase to release NO and produce vasorelaxation (7) (Fig. 2). The lack of pressor response observed with oxytocin infusions in obstetrical practice may be consequent to the opposing effects of OTR stimulation on endothelial and smooth muscle cells.
Vasopressin produces vasodilation in some vascular beds, but the receptor subtype responsible is uncertain, may vary between blood vessels, and may depend on hormone concentration. The receptors mediating this effect may be situated on endothelial or smooth muscle cells or both. V2Rs on vascular smooth muscle could produce vasorelaxation via a cAMP-mediated drop in intracellular calcium; alternatively, generation of cAMP in the vascular endothelium, where an increase in intracellular Ca\(^{2+}\) activates constitutive endothelial nitric oxide synthase (eNOS) to release nitric oxide (NO). There is experimental evidence to suggest the existence of endothelial V1Rs and V2Rs. Stimulation of either subtype would activate eNOS, the former via calcium-calmodulin and the latter via an elevation in cAMP. Endothelial-derived NO causes vascular smooth muscle relaxation by a cAMP-mediated decrease in intracellular Ca\(^{2+}\) entry via voltage-gated calcium channels. This contributes to both hypotension and hyporesponsiveness to catecholamines (35–37). In addition to the effect of elevated NO, persistent KATP activation may result from tissue hypoxia, acidosis, reduced ATP, and changes in calcitomin gene-related peptide, adenosine, and atrial natriuretic factor levels (34). A third factor is adrenoreceptor desensitization and down-regulation due to high circulating levels of catecholamines (38, 39).

The finding that patients with severe, refractory septic shock were exquisitely sensitive to the pressor effects of exogenous vasopressin led to the investigation of its endogenous profile (40). In acute septic shock, an early increase (approximately ten-fold) in plasma vasopressin occurs in both patients (41) and animal models (42, 43). When prolonged (>24 hrs), however, levels fall back toward baseline (40, 41), a pattern mimicking that observed for other hormones in advanced critical illness (44). Hence, a relative deficiency of vasopressin may also be crucial to the altered functional status of vascular smooth muscle. Indeed, in endotoxemic models, V1R blockade worsens hypotension (45), whereas survival was decreased in vasopressin-deficient Brattleboro rats (46).
Inappropriately low hormone levels are not explained by increased vasopressin breakdown but may be caused by depletion of neurohypophysial stores or inhibition of synthesis or release (47). Either osmoregulation or baroregulation may be abnormal, and baroreflex dysfunction could underlie the apparent loss of correlation between blood pressure and vasopressin levels in septic shock (48). Impaired vasopressin release has been documented in patients with autonomic insufficiency (49, 50), a phenomenon well recognized in sepsis (51). Elevated levels of NO may contribute to autonomic dysfunction (52) and have direct inhibitory effects on vasopressin secretion (5). Sustained elevation of hormone levels following endotoxin challenge in mice was seen in iNOS knockouts or after pharmacologic NO inhibition (53–55).

**EVIDENCE FOR HYPERSENSITIVITY TO VASOPRESSIN IN SEPTIC SHOCK**

Exogenous administration of vasopressin in health does not elevate blood pressure, and hypertension is not characteristic of the syndrome of inappropriate antidiuretic hormone. Landry and colleagues (56) reported marked pressor sensitivity to a low dose of vasopressin in five patients with sepsis-related refractory hypotension. The pressor effect occurred within minutes and enabled catecholamines to be discontinued. Our group has published comparable results in a cohort of eight similar patients in whom terlipressin, a long-acting synthetic vasopressin analogue, was administered (57). Despite an increasing number of related studies, most have only included small numbers of patients and have been retrospective or nonrandomized (58). Ongoing is a large, multiple-center Canadian trial of vasopressin vs. norepinephrine in septic shock (VASST), which will examine 28-day mortality as the primary end point. Current consensus opinion is that low-rate constant infusion (0.01–0.04 units/min) of vasopressin is preferable to a higher, blood pressure-titrated dose if coronary, mesenteric, and skin ischemias are to be avoided (59, 60). There is increasing evidence to suggest neutral or beneficial effects on renal blood flow and urine output at these low doses (58, 61).

Several animal models of septic shock have demonstrated hypersensitivity to vasopressin. In anesthetized endotoxic rats, a heightened contractile response of cremaster muscle microvessels to topical vasopressin was coincident to hyporeactivity to norepinephrine (62, 63). In our laboratory’s conscious, fluid-resuscitated model of rat fecal peritonitis (64), septic animals show a marked pressor response to terlipressin 24 hrs post insult that is not seen in paired sham controls (65). Hypersensitivity to terlipressin was also seen in conscious ewes after 16 hrs of endotoxemia (66). Other in vivo setups have, however, produced discordant results (67–69), most likely related to the wide experimental variation in terms of duration, insult, severity, and fluid resuscitation.

Although ex vivo reproduction of the vascular hyporeactivity to catecholamines is well described in septic models (70–72), there has been relatively little work examining vascular reactivity to vasopressin. One study showed increased potency of vasopressin to constrict isolated mesenteric vessels from endotoxemic compared with control rats (73). In contrast, attenuated responses to vasopressin were observed in human gastroepiploic arteries after endotoxin treatment, but vasopressin significantly enhanced norepinephrine-induced contractions in the same tissue (74). Decreased sensitivity to both vasopressin and norepinephrine was found in isolated rat mesenteric arteries pretreated with an NO donor to simulate septic shock (75).

**MECHANISMS UNDERLYING VASOPRESSIN HYPERSENSITIVITY**

These are summarized in Figure 3.

**Interaction With Other Factors Contributing to Vasodilatory Shock**

**Nitric Oxide.** As discussed previously, elevated levels of NO in septic shock may contribute to relative vasopressin deficiency. Another consideration is a possible reciprocal negative effect of vasopressin on the NO cascade. Vasopressin inhibited interleukin-1 stimulated iNOS messenger RNA expression and nitrite and cyclic guanosine monophosphate production in cultured rat vascular smooth muscle cells (76). Basal NO production was unaltered, suggesting an effect specific to iNOS and hence states of
inflammatory activation. The hypothesis that heightened sensitivity to exogenous vasopressin in septic shock may be consequent to iNOS inhibition is further supported by the findings of an in vivo study where administration of terlipressin to endotoxic rats resulted in recovery of arterial blood pressure associated with decreased iNOS expression in isolated aortic tissue (77). However, no decrease in serum nitrite/nitrate concentrations was demonstrated in patients with vasodilatory shock after vasopressin infusion (78).

**K\textsubscript{ATP} Channels.** In septic shock K\textsubscript{ATP} channels are persistently open, resulting in a sustained hyperpolarized state and vasorelaxation. Inhibition of these channels could therefore help to restore normal vascular reactivity (37, 79). In vitro work in cultured porcine vascular smooth muscle cells and isolated cardiac myocytes has demonstrated the ability of vasopressin to close K\textsubscript{ATP} channels (80, 81). This effect was blocked by selective inhibition of PKC (81), which may act by direct phosphorylation of the channel (82) or by increasing sarcolemmal ATP (81). Other non-PKC-mediated mechanisms are also possible. An increase in intracellular calcium evoked by V\textsubscript{1}R stimulation could activate the calcium-dependent phosphatase, calcineurin, to promote channel inhibition (79, 83). In addition, calcineurin regulates gene transcription via the nuclear transcription factor nuclear factor of activated T cells; this in turn may down-regulate genes encoding K\textsubscript{ATP} channel subunits, as has been shown for delayed rectifier potassium channels (84).

**Catecholamine Sensitivity.** Clinical experience with vasopressin and terlipressin in patients with refractory septic shock suggests that vasopressin and terlipressin restore vascular reactivity to both endogenous and exogenous catecholamines (56, 57, 85). In vivo potentiation of the vasoconstrictor actions of endogenous norepinephrine by physiologic doses of exogenous vasopressin was first reported >40 yrs ago (86). Parallel ex vivo studies with rat aortic strips suggested a direct vascular rather than central mechanism of vasopressin action (86). This potentiation has been confirmed in rat and human resistance arteries, seen in both normal vessels (87, 88) and those exposed to experimental sepsis (74, 75). Similarly, constriction evoked by stimulation of periartrial nerves is also enhanced and observed at concentra-

**Changes in Vasopressin Receptor Behavior.** The opposing effects of vasopressin on vascular tissue are consequent to the stimulation of different vasopressin receptor subtypes located on smooth muscle and/or endothelial cells. Differential changes in the regulation of these subtypes could therefore explain the hypersensitivity seen in septic shock.

**V\textsubscript{1} Receptors.** In contrast to high norepinephrine levels and the resultant \(\alpha_1\) receptor changes, relatively low circulating concentrations of vasopressin in prolonged septic shock would leave V\textsubscript{1}Rs available for occupancy by exogenous hormone and decrease the endogenous stimulus for receptor desensitization (6, 32). Sepsis may also induce specific changes in receptor populations. Endotoxin can alter receptor function directly (91) or indirectly via cytokines, NO, and PKC. No change in either the number or affinity of V\textsubscript{1}Rs was seen in cultured aortic smooth muscle cells exposed to lipopolysaccharide for 24 hrs, however (92).

Another group reported a cytokine-mediated decrease in V\textsubscript{1}Rs in liver, lung, kidney, and heart tissue isolated from endotoxic rats exposed for a similar period (93). This model was not fluid resuscitated and did not demonstrate hypersensitivity to in vivo administration of a vasopressin agonist. Comparable results were seen in nonshocked rats who received continuous endotoxin infusion for 30 hrs (94). Further studies are required to examine changes in receptor binding in tissues from models more representative of human septic shock.

**V\textsubscript{2} and Oxytocin Receptors.** Whereas vasorelaxation can be mediated via both V\textsubscript{2}Rs and OTRs, changes in the expression or function of these receptors in sepsis remain unknown. Increasing availability of specific agonists and antagonists now makes this a realistic proposition. V\textsubscript{2}R recycling and resensitization are slow compared with V\textsubscript{1}Rs (6, 7). This may well be of relevance in the context of exogenous vasopressin administration and could be one explanation for the observation that rebound hypotension on cessation of vasopressin treatment in septic shock is often prevented with the use of terlipressin (57, 95), an analogue with greater selectivity for V\textsubscript{1}Rs over V\textsubscript{2}Rs (2.2 vs. 1) (57, 96).

**Autonomic Nervous System Dysfunction.** Vasopressin release is under the control of the autonomic nervous system, with baro- and chemoreceptor afferents projecting to the brainstem and efferents from the brainstem to the paraventricular and supraoptic (3). By virtue of its neurotransmitter role, autonomic nervous system output is also modulated by vasopressin (15). Therefore, the autonomic and vasopressinergic system abnormalities seen in sepsis may well be related. Further complexity is added by the apparent negative correlation between NO levels and sympathetic cardiovascular output (52, 97) and the known interactions between vasopressin and NO described previously. In patients who died from septic shock, iNOS expression was linked to apoptosis in the paraventricular and supraoptic nuclei (52). Primary autonomic failure is associated with hypersensitivity to vasopressin’s pressor effects (98) as well as with abnormalities of its release (49). The former has also been reported in dogs with baroreceptor denervation (99). Moreover, cirrhotic pa-
tients show an abnormally prolonged blood pressure response to vasopressin, and this has been ascribed to abnormal autonomic cardiovascular regulation (100). Vasopressin administration in septic shock does not produce the degree of bradycardia seen in normal individuals (40), suggesting impairment of normal baroreflexes.

Interaction With Other Vasocostrictrors

Elevated levels of endothelin-1 and thromboxane A2 are found in septic shock and contribute to the heterogeneity in tone observed across different vascular beds (101). Vasopressin administration may increase the synthesis of these vasocostrictrors. In vitro work with human platelets showed that V1R stimulation acts despite no hemodynamic response to corticotropin releasing hormone (41). Furthermore, hypersensitivity to vasopressin is likely to have a relative deficiency of vascular sensitivity to exogenous vasopressin in spontaneously hypertensive rats was abolished by pre-treatment with the endothelin antagonist bosentan (105).

Interactions With the Hypothalamic-Pituitary-Adrenal Axis

Vasopressin stimulates adrenocorticotrophic hormone and hence cortisol secretion (2). Relative adrenal insufficiency is recognized in severe sepsis (106), and “low-dose” steroid replacement may provide outcome benefit in such patients (107). Steroid administration is thought to restore vascular sensitivity to catecholamines via an increase in adrenergic receptor gene expression (39). Although a similar effect on vasopressin receptor expression is feasible (108), vasopressin replacement may increase cortisol levels, thus acting synergistically to restore reactivity. However, septic patients with relative adrenal insufficiency were less likely to have a relative deficiency of vasopressin than those with normal adrenal function (41). Furthermore, hypersensitivity to vasopressin may still occur despite no hemodynamic response to corticosteroids (57).

REFERENCES

22. Tagawa T, Imaiuzumi T, Shiramoto M, et al: V2 receptor-mediated vasodilation in


