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(54) Title: USE OF VASOPRESSIN-RECEPTOR AGONISTS FOR THE TREATMENT OF SEPTIC SHOCK

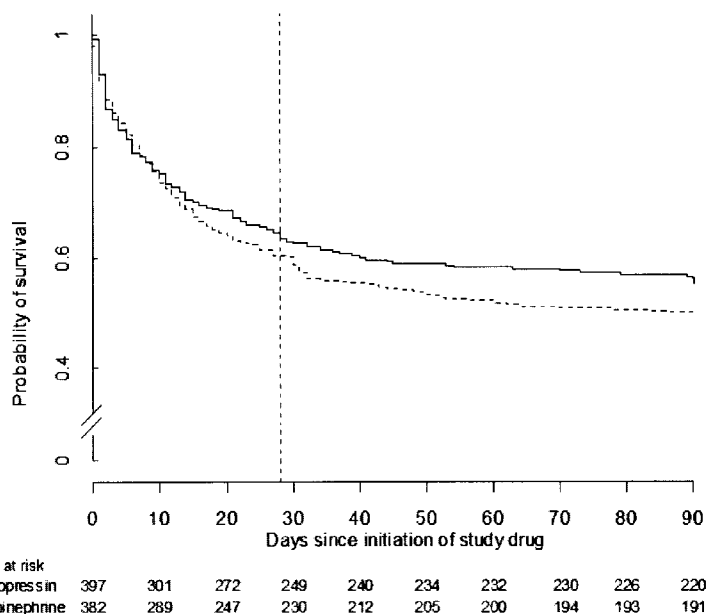


FIG. 2

(57) Abstract: A method of treating a subject having septic shock is provided, the method comprising administering to the subject vasopressin, or a vasopressin-receptor agonist. The method may further comprise administration of a corticosteroid, or a reduced dose of a vasopressor, such as norepinephrine. Methods for accessing the eligibility of a subject for treatment of septic shock with vasopressin are also disclosed.

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USE OF VASOPRESSIN-RECEPTOR AGONISTS FOR THE TREATMENT OF SEPTIC SHOCK

This application claims priority to U.S. Provisional Application No. 60/929,926, filed July 18, 2007.

5 FIELD OF INVENTION

[0001] The present invention relates to the field of septic shock. More specifically, the present invention relates to a method of using vasopressin-receptor agonists for the treatment of patients with septic shock.

BACKGROUND OF THE INVENTION

10 [0002] Septic shock remains the most common cause of death in intensive care units (ICU). Approximately 600,000 Americans develop septic shock annually, and roughly half of them die from complications of the condition. With a mortality rate of 40–60%^{1, 2} there is dissatisfaction with existing therapies, and the treatment for septic shock has not significantly improved since the advent of broad spectrum antibiotics in the early 1960s.
15 Improving treatment can save many lives of patients that are at risk of developing this condition. Furthermore, treatment in the ICU is very costly, and limiting the number of days and patients in need of such intensive intervention could save health care dollars.

[0003] Sepsis is a systemic infection that may be caused by primary bacteremia; however it is often triggered by events such as trauma, surgery, perforated bowel and burns, or by
20 conditions such as pneumonia, urinary tract infection, wound infection, peritonitis, cholangitis, cellulitis, intra-abdominal abscess and post-surgery infection. It may progress to more severe conditions such as septic shock and result in multiple organ failure and death. Septic shock is a serious medical condition characterized by hypotension, hyperventilation and eventually organ dysfunction. Refractory septic shock is very serious
25 with the patient not responding to increasing doses of vasopressors and often accompanied by multiple organ failures or near-failures. Death and organ dysfunction/failure are caused by poor tissue perfusion and reduced oxygen delivery.

[0004] The most common victims of sepsis are people whose immune system cannot cope with infection efficiently, such as children, the elderly and immunocompromised
30 individuals. A compromised immune system can be caused by for example surgery, tissue

transplantation and chemotherapy. Large scale inflammatory response results in massive vasodilation, increased capillary permeability, decreased systemic vascular resistance, and hypotension. Hypotension reduces tissue perfusion pressure and thus tissue hypoxia ensues. Finally, in an attempt to offset decreased blood pressure, ventricular dilatation and myocardial dysfunction may occur. The impact of sepsis is particularly devastating to patients with compromised cardiac, hepatic and immune-defense function.

[0005] The Acute Physiology and Chronic Health Evaluation (APACHE) II³ scoring system is used to predict morbidity for ICU patients 15 years of age or older. The score is based on age, routine physiological measurements, information about immunocompromisation or chronic organ failure and the Glasgow Coma Score (a neurological measure of consciousness). The Apache II score ranges from 0-71; 71 indicating the highest risk of death. The physiological measurements include: body temperature, heart rate, blood pressure, mean arterial pressure, respiratory rate, pulmonary function ($\text{PaO}_2/\text{FiO}_2$), white blood cell count, arterial pH, blood oxygenation and serum levels of creatinine, sodium, potassium and HCO_3 . The APACHE II score can be used to determine the severity of septic shock.

[0006] One of the outcomes that can cause or contribute to death in septic shock is renal failure. The RIFLE criteria (Risk/ Injury/ Failure/ Loss/ End-stage) were recently proposed to describe acute renal failure⁴ and can be used to distinguish septic shock patients at increased risk of renal failure or death.

[0007] Resuscitation strategies include intravenous fluid and the use of vasopressors including catecholamines such as norepinephrine, epinephrine, dopamine and dobutamine. Although largely successful in re-establishing minimally acceptable mean arterial pressures to maintain organ perfusion, catecholamines have important adverse effects and may even increase mortality. For example, norepinephrine, a potent, commonly-used α -adrenergic agent in septic shock may decrease cardiac output, oxygen delivery, and blood flow to vulnerable organs despite adequate perfusion pressure.

[0008] Corticosteroids may also be used in resuscitation strategies for subjects with septic shock. Reducing the subject's inflammatory response may be beneficial and aid in restoring cytokine homeostasis, however a review of various clinical studies (Delliner,

2004), suggests these may be several opinions on the matter, with respect to dose and timing of administration.

[0009] Arginine vasopressin (AVP) is a human peptide hormone that is released in response to hypotension or when the body is low on water. AVP causes the kidneys to conserve water, but not salt, by concentrating the urine and reducing urine volume. It also raises blood pressure by inducing moderate vasoconstriction. Ethanol and caffeine increase urination by reducing vasopressin secretion, whereas angiotensin II increases water reabsorption by stimulating the secretion of vasopressin.

[0010] Vasopressin is currently used as a second-line and add-on therapy to treat severe vasodilatory hypotension that is unresponsive or refractory to one or more adrenergic agents (eg, high-dose dopamine, epinephrine, norepinephrine, phenylephrine)⁵. Its use has been associated with side-effects such as bradycardia, gut ischemia, digital ischemia and myocardial ischemia⁶.

[0011] Vasopressin binds to vasopressin-specific membrane-bound V1 receptors (also known as AVPR1A) on vascular smooth muscle⁶, stimulating vasoconstriction because of contraction of vascular smooth muscle. Furthermore, vasopressin binds V2 receptors on the distal convoluted tubule and collecting ducts in the kidney, and V3 receptors in the pituitary gland that modify adrenocorticotropin hormone synthesis and release.

[0012] Vasopressin is synthesized in the hypothalamus. Most of it is stored in the posterior part of the pituitary gland to be released into the blood stream in response to a reduced plasma volume or an increase in plasma osmolality. Steady state normal plasma levels of vasopressin is about 5 – 10 pmol/L. The half-life of vasopressin is approximately 5-15 minutes.

[0013] Exogenous vasopressin and other vasopressin-receptor agonists are used to prevent or control polyuria, polydipsia, and dehydration in patients with central diabetes insipidus. It is also used to prevent nocturnal enuresis, or nocturnal involuntary emptying of the bladder (Nocturia), mostly affecting children. Intravenous vasopressin is included in the Advanced Cardiac Life Support (ACLS) algorithm as an alternative to epinephrine for the treatment of cardiac arrest associated with asystole or pulseless electrical activity.

Vasopressin also has been used in the treatment of intestinal paresis, postoperative abdominal distention, and upper GI tract hemorrhage, although this practice has been abandoned.

[0014] Terlipressin is a long-acting synthetic vasopressin analogue. When the term vasopressin or AVP is used (herein and elsewhere), it may refer to terlipressin or any other vasopressin-receptor agonist.

[0015] The use of vasopressin for the treatment of severe septic shock has been discussed, for example by Dünser et al⁷, Patel et al⁸, Landry et al^{9, 10} and Russell & Landry¹¹, and the opinions, comments and election of whether or not to use vasopressin and when, and under what conditions, are many and varied. Russell & Landry¹¹ have indicated that vasopressin may be beneficial in extreme cases of sepsis, but should not be used routinely, and there is suggestion that vasopressin may be harmful.

[0016] The use of corticosteroids in combination with vasopressin has recently been described by Ertmer et al¹².

[0017] Furthermore, the following patents relate to the use of vasopressin for related indications:

[0018] **US7183255: Method for stabilizing blood pressure in hemodialysis subjects**

[0019] **US5827893: Mechanical and pharmacological therapies to treat cardiac arrest**

[0020] The present invention provides, in part, improved uses of vasopressin or vasopressin-receptor agonists for the treatment of septic shock.

SUMMARY OF THE INVENTION

[0021] The present invention relates to a use of vasopressin, or a vasopressin-receptor agonist for the treatment of septic shock. Vasopressin may be used as an adjunct to catecholamines, corticosteroids or norepinephrine to support blood pressure in refractory septic shock. Infusion of low-dose vasopressin as a first-line of treatment significantly decreases mortality in patients with less severe septic shock, and in septic shock-patients treated with steroids.

[0022] In accordance with one aspect of the invention, there is provided a method of preventing organ failure in a subject having septic shock, the method comprising administering to the subject vasopressin, or a vasopressin-receptor agonist, for sufficient time to prevent organ failure. The organ failure may be renal failure. The septic shock may be less severe septic shock.

[0023] In accordance with another aspect of the invention, there is provided a method of treating a subject having less severe septic shock, the method comprising administering to the subject vasopressin, or a vasopressin-receptor agonist, for sufficient time to treat septic shock.

[0024] In accordance with another aspect of the invention, the methods may further comprise administering to the subject one or more corticosteroids.

[0025] In accordance with another aspect of the invention, the vasopressin or vasopressin-receptor agonist may be administered in a dose of 0.01 to 0.03 U / min.

[0026] In accordance with another aspect of the invention, there is provided a kit for use in treating a subject having less severe septic shock, comprising vasopressin or a vasopressin-receptor agonist, and instructions for use. The instructions may further comprise dosing information, or instructions for selecting a subject having less severe septic shock.

[0027] In accordance with another aspect of the invention, there is provided a method of determining the eligibility of a subject for treatment of septic shock with vasopressin or a vasopressin-receptor agonist, the method comprising: (i) determining the subject's requirement for vasopressor support; (ii) determining the subject's requirement for one or less vasopressors at baseline; (iii) determining the subject's baseline lactate levels; (iv) determining the subject's APACHE II score at baseline; and (v) determining the subject's signs of renal dysfunction at baseline; wherein the subject is eligible for treatment of septic shock with vasopressin or a vasopressin-receptor agonist if the subject meets one or more of the following criteria: (i) requiring less than or equal to 14µg norepinephrine /min); (ii) requiring one or less vasopressors at baseline; (iii) a baseline lactate level ≤ 1.4 mmol/L; (iv) an APACHE II score at baseline ≤ 25 ; and (v) an absence of signs of renal dysfunction at baseline.

[0028] In accordance with another aspect of the invention, there is provided a use of vasopressin as a catecholeamine-sparing agent for the treatment of less severe septic shock, wherein the catecholeamine is norepinephrine, and administered at a dose of less than 14 ug/minute.

5 [0029] Such methods of treatment or prevention, or uses of one or more aspects of the invention may provide for an advantageous reduction in the subject's requirement for corticosteroids relative to a subject not receiving vasopressin or a vasopressin-receptor agonist. As an example, a subject receiving vasopressin or a vasopressin-receptor agonist may demonstrate a "Days Alive and Free" (DAF) score (corticosteroid) of >16), while a
10 subject not receiving vasopressin or a vasopressin-receptor agonist may demonstrate a DAF score (corticosteroid use) of 16 or less.

[0030] Such methods of treatment or prevention, or uses of one or more aspects of the invention may provide for an advantageous reduction in the subject's requirement for mechanical ventilation relative to a subject not receiving vasopressin or a vasopressin-receptor agonist. As an example, a subject receiving vasopressin or a vasopressin-
15 receptor agonist may demonstrate a "Days Alive and Free" (DAF) score (mechanical ventilation) of >14), while a subject not receiving vasopressin or a vasopressin-receptor agonist may demonstrate a DAF score (mechanical ventilation) of 14 or less.

[0031] Such methods of treatment or prevention, or uses of one or more aspects of the invention may provide for an advantageous reduction in the subject's requirement for mechanical ventilation relative to a subject not receiving vasopressin or a vasopressin-receptor agonist. As an example, a subject receiving vasopressin or a vasopressin-receptor agonist may demonstrate a "Days Alive and Free" (DAF) score (renal failure) of >4), while a subject not receiving vasopressin or a vasopressin-receptor agonist may
20 demonstrate a DAF score (renal failure) of 4 or less.
25

[0032] In accordance with another aspect of the invention, the vasopressin-receptor agonist is selected from the group comprising, arginine vasopressin (AVP), lysine vasopressin (LVP), triglycil-lysine vasopressin, octopressin, ornipressin, and desmopressin (DDAVP).

[0033] In a first embodiment, a method is provided for treating patients with septic shock, wherein the method may involve the use of vasopressin-receptor agonists on a selected subset of patients with septic shock.

[0034] In a further embodiment, a method is provided whereby a vasopressin-receptor agonist may be used for the treatment of patients with less severe septic shock.

[0035] In yet another embodiment, a method is provided whereby a vasopressin-receptor agonist may be used for the treatment of patients with septic shock and requiring low dose vasopressor support (such as $\leq 14\mu\text{g}$ norepinephrine /min).

[0036] In a further embodiment, a method is provided whereby a vasopressin-receptor agonist may be used for the treatment of patients with septic shock and low baseline lactate levels (such as $\leq 1.4\text{mmol/L}$).

[0037] In another embodiment, a method is provided whereby a vasopressin-receptor agonist may be used for the treatment of patients with septic shock treated with one or less vasopressors at baseline.

[0038] In one embodiment, a method is provided whereby a vasopressin-receptor agonist may be used for the treatment of patients with septic shock that show no signs of renal dysfunction at baseline.

[0039] In yet another embodiment, a method is provided whereby a vasopressin-receptor agonist may be used for the treatment of patients with septic shock at increased risk of developing renal failure or renal dysfunction.

[0040] In another embodiment, a method is provided whereby a vasopressin-receptor agonist may be used for the treatment of patients with septic shock in conjunction with treatment using corticosteroids, including but not limited to the administration of one or more doses of cortisone, hydrocortisone, prednisone, and methylprednisolone.

[0041] In a further embodiment, the dose of vasopressin-receptor agonist that is used may be low, such as corresponding to 0.01-0.03U of vasopressin /min.

[0042] In another embodiment, the vasopressin-receptor agonist may be used in combination with other vasopressors and/or any agents that may improve the outcome for the patient.

[0043] This summary of the invention does not necessarily describe all features of the invention. Other aspects, features and advantages of the present invention will become apparent to those of ordinary skill in the art upon review of the following description of specific embodiments of the invention.

BRIEF DESCRIPTION OF THE DRAWINGS

[0044] These and other features of the invention will become more apparent from the following description in which reference is made to the appended drawings wherein:

[0045] **FIGURE 1.** Flowchart of subject enrollment and outcomes. “NYHA” denotes New York Heart Association classification. “OR” denotes operating room. “MI” denotes myocardial infarction.

[0046] **FIGURE 2.** 90-day Kaplan Meier survival curves for all randomized and infused patients, ($P = 0.27$ at day 28 and $P = 0.10$ at day 90). (See Fig. 6 for subgroup results.) solid black line is the vasopressin treated group, the dotted line is the norepinephrine treated group, and the vertical line marks day 28. P values were calculated using the log rank statistic.

[0047] **FIGURE 3.** Comparison of mean arterial pressure (A) and heart rate (B) in the norepinephrine group (grey circles) and vasopressin group (black squares). Values are mean $\pm$ standard deviation.

[0048] **FIGURE 4.** Rates of total norepinephrine infusion (open-label and study drug) in the vasopressin treated group (black squares) and the norepinephrine treated group (grey circles) amongst all patients who were treated with open-label norepinephrine at baseline. Values are median + interquartile range.

[0049] **FIGURE 5.** Plasma vasopressin levels over time in patients receiving a vasopressin infusion, $N = 54$, (black squares), patients in the vasopressin group once

vasopressin infusions had stopped (open squares) and patients in the norepinephrine group, N = 53, (grey circles). Values are median + interquartile range.

[0050] **FIGURE 6.** 90-day Kaplan Meier survival curves, A) patients in more severe stratum, (P = 0.77 at day 28 and P= 0.92 at day 90), B) patients in less severe stratum (P = 0.05 at day 28 and P= 0.03 at day 90). Solid black line is the vasopressin treated group, the dotted line is the norepinephrine treated group, and the vertical line marks day 28. P values were calculated using the log rank statistic.

[0051] **FIGURE 7.** Renal function at enrollment for all VASST patients according to RIFLE criteria.

[0052] **FIGURE 8.** 28-day mortality according to baseline RIFLE criteria. The presence of any renal dysfunction was associated with increased mortality compared to no renal dysfunction (44.3% vs 27.0%, respectively, * P<0.001).

[0053] **FIGURE 9.** The rate of progression to renal failure or loss in patients at risk according to treatment (* P=0.028).

DETAILED DESCRIPTION

[0054] In the description that follows, a number of terms are used extensively, the following definitions are provided to facilitate understanding of various aspects of the invention. Use of examples in the specification, including examples of terms, is for illustrative purposes only and is not intended to limit the scope and meaning of the embodiments of the invention herein.

[0055] In order that the invention herein described may be more fully understood, the following description is set forth.

[0056] As used herein a 'subject' refers to an animal, such as a bird or a mammal.

Specific animals include rat, mouse, dog, cat, cow, sheep, horse, pig or primate. A subject may further be a human, alternatively referred to as a patient. A subject may further be a transgenic animal. A subject may further be a rodent, such as a mouse or a rat.

[0057] A 'vasopressin-receptor agonist' is any compound that has the ability to bind and activate any vasopressin-receptor and induce constriction of smooth muscle and blood

vessels and/ or returning a reduced blood pressure to normal levels. Examples of vasopressin-receptor agonists or vasopressin-like peptides include, but are not limited to: arginine vasopressin (also known as AVP, argipressin, antidiuretic hormone or ADH), lysine vasopressin triglycyl-lysine vasopressin (also known as glycopressin or terlipressin), octopressin, ornipressin, and 1-deamino-8-D-arginine vasopressin (also known as desmopressin or DDAVP).

[0058] Furthermore, vasopressin-receptor agonists includes analogues of arginine vasopressin, including but not limited to such analogues extended by 1-3 amino acids such as Ala-AVP, Ser-Ala-AVP and Thr-Ser-Ala-AVP as well as other compounds including but not limited to 3-beta-(2-thienyl)-L-alanine-8-lysine-, N-alpha-glycyl-glycyl-glycyl-[8-lysine]-vasopressin 1-deamino-6-carba-[8-arginine]-vasopressin and analogues thereof. Other examples of vasopressin-receptor agonists are described in, for example, PCT Publication WO 2006/020491, incorporated herein by reference.

[0059] When the term 'study drug' or 'drug' or 'AVP' or 'vasopressin' is used (herein and elsewhere), it may refer to terlipressin or any other vasopressin-receptor agonist.

[0060] 'Baseline' refers to the level of a physiological measure before treatment was initiated, such as treatment with an adrenergic agent or a vasopressin-receptor agonist such as the study drug.

[0061] In the context of the present invention, the terms "treatment," "treating", "therapeutic use," or "treatment regimen" as used herein may be used interchangeably are meant to encompass prophylactic, palliative, and therapeutic modalities of the methods, uses and kits of the present invention. This includes any and all applications of the presently claimed methods, uses or to a subject having, or suspected of having septic shock, SIRS or less severe septic shock. Thus, any prevention, amelioration, alleviation, reversal, or complete elimination of septic shock, SIRS or less severe septic shock, or criteria of septic shock, SIRS or less severe septic shock, is encompassed by the present invention.

[0062] 'Septic shock' is herein defined by the presence of two or more of the systemic inflammatory response syndrome (SIRS) criteria¹³ (including fever or hypothermia, tachypnea or need for mechanical ventilation, tachycardia and abnormal white blood cell

count), proven or suspected infection, at least one new organ dysfunction, and hypotension despite adequate fluid resuscitation requiring vasopressor support of at least 5 µg/min of norepinephrine (or equivalent).

[0063] 'Less severe septic shock' is herein defined as septic shock and with at least one of the following characteristics:

- a) requiring low dose baseline vasopressor support (such as less than or equal to 14µg norepinephrine /min).
- b) requiring one or less vasopressors at baseline
- c) low baseline lactate levels (such as ≤ 1.4 mmol/L)
- d) a low APACHE II score at baseline (such as ≤ 25)
- e) void of signs of renal dysfunction at baseline, including patients currently void of signs yet at increased risk of developing renal dysfunction or renal failure.

[0064] 'Known or suspected infection' is herein defined by clinical suspicion of infection (microbiology cultures were positive or pending and the patient was being treated using antibiotics).

[0065] 'Hypotension' is herein defined by: systolic blood pressure (SBP) less than 90 mmHg or decrease in SBP by at least 40 mmHg for more than one hour while central venous pressures (CVP) remained adequate (≥ 12 mmHg) or at least 500 mL of saline was infused or if vasopressors were infused to maintain blood pressure.

[0066] A "vasopressor" is an agent, factor or composition, frequently a pharmaceutical composition, that causes vasoconstriction.

[0067] 'Vasopressor requirement' or a 'patient requiring vasopressors' is herein defined as patients in need of ≥ 5 µg/min of norepinephrine or equivalent in the preceeding 24 hours, OR patients requiring high vasopressor doses (norepinephrine equivalent ≥ 15 µg/hr) to maintain a mean arterial pressure of 65-75 mmHg.

[0068] 'Norepinephrine equivalent dose' is herein calculated as [norepinephrine (µg/min)] + [dopamine (µg/kg/min) ÷ 2] + [epinephrine (µg/min)] + [phenylephrine (µg/min) ÷ 10] after the study of Patel *et al.*⁸

[0069] Acutely ill subjects may experience dysfunction or failure of one or more organs (e.g. heart, liver, lung, kidney, pancreas, and the like), necessitating mechanical intervention to maintain life. Monitoring of physiological indicators (e.g. urine output, heart rate, blood pressure, body temperature, respiratory rate, and blood or plasma enzymes, metabolites and the like) may provide an indication of risk, injury, failure or complete loss of organ function. The subject's degree of organ dysfunction may be described using a scoring system, such as the Sepsis-Related Organ Failure Assessment (SOFA) (Moreno et al., 1999) or Brussels score¹⁶, while individual organ function/dysfunction/failure may be assessed using other defining features, e.g. acute renal failure and the RIFLE criteria.

[0070] 'Acute renal failure' is herein defined according to the consensus RIFLE serum creatinine criteria:

R-Risk; Increased serum creatinine x1.5 patient baseline

I-Injury; Increased serum creatinine x2 patient baseline

F-Failure; Increased serum creatinine x3 patient baseline or if patient baseline creatinine is $\geq 350 \mu\text{mol/l}$ (4mg/dl) and acute rise is $\geq 44 \mu\text{mol/l}$ (0.5 mg/dl).

L-Loss; Complete loss of renal function (dialysis) for at least 4 weeks.

E-End stage; Renal failure (dialysis) for at least 3 months.

[0071] 'New organ dysfunction' is herein defined as respiratory (ventilated and $\text{PaO}_2/\text{FiO}_2 < 300 \text{ mmHg}$), renal (urine output $< 30 \text{ mL/hour}$ or less than 0.5 mL/kg body weight, for at least one hour), coagulation (platelet count $< 80,000/\text{mm}^3$), and neurologic (Glasgow Coma Score < 12 , prior to receiving sedation).

[0072] 'Serious adverse events' that could occur and may warrant discontinuing treatment with catecholamine vasopressors and vasopressin-receptor agonist includes: (1) acute S-T segment elevation confirmed by 12-lead electrocardiogram, (2) serious or life-threatening (hemodynamically unstable) cardiac arrhythmias, (3) acute mesenteric ischemia, (4) digital ischemia, or (5) hyponatremia (serum sodium $< 130 \text{ mmol/L}$).

[0073] 'Open-label catecholamine vasopressors' include epinephrine, norepinephrine, dopamine and dobutamine that were initiated at baseline or as an adjunct to the main treatment, such as the study drug. 'Corticosteroids' refers to any analogues of cortisol, corticosterone or aldosterone as well as any compounds that resemble or mimic their

function in a subject, including but not limited to cortisone, hydrocortisone, prednisone and methylprednisolone.

[0074] A “sufficient time to treat less severe septic shock” or “sufficient time to prevent organ failure” in reference to the duration of time that a subject was administered vasopressin, arginine vasopressin (AVP), lysine vasopressin (LVP), triglycil-lysine vasopressin, octopressin, ornipressin, and desmopressin (DDAVP), or a vasopressin-receptor agonist is the time period from when the first administration commenced to when administration discontinued,, including the initiation of weaning of the subject from vasopressin, arginine vasopressin (AVP), lysine vasopressin (LVP), triglycil-lysine vasopressin, octopressin, ornipressin, and desmopressin (DDAVP), or a vasopressin-receptor agonist.

[0075] General aspects of the invention and its use

[0076] Previous use of vasopressin for septic shock has been limited to severe or refractory septic shock. Surprisingly, as presented in example 1 vasopressin treatment does not improve 28-day or 90-day mortality rates for patients with septic shock in general. Neither does vasopressin treatment improve 28-day or 90-day mortality rates for patients with more severe septic shock. In sharp contrast with the current use and knowledge of vasopressin treatment for septic shock, low-dose vasopressin treatment significantly reduces 28-day and 90-day mortality rates for patients with less severe septic shock and for patients treated with corticosteroids.

[0077] In particular, patients with less severe septic shock as represented by low-dose vasopressor support at baseline ($\leq 14\mu\text{g}$ norepinephrine /min), requirement for only one vasopressor at baseline, low baseline lactate levels ($\leq 1.4\text{mmol/L}$) and no renal dysfunction (renal data is presented in example 2) exhibit reduced mortality rates in response to vasopressin treatment.

[0078] Furthermore as presented in example 2, vasopressin treatment reduces the rate of progression to renal failure or loss in patients with no baseline renal dysfunction, including patients at risk of developing such.

[0079] Patients with less severe septic shock differ from patients with more severe septic shock in at least two significant ways:

a) They either lack damage to internal organs, or if they have sustained damage it is reversible (not permanent) and,

5 b) Their immune system has not entered into a state of particularly aggressive, albeit generally futile, cycle of increasing cytokine levels and tachyphylaxis.

[0080] It is well known that endotoxins induce the release of cytokines. It has also been shown that high levels of catecholamines can induce or potentiate cytokine release. Elevated levels of cytokines cause tachyphylaxis (that is, hypo-responsiveness) in
10 response to vasopressors by downregulating the number of receptors that mediate vasopressor-response. Extremely elevated cytokine levels may explain the reduced vasopressor response in more severe septic shock. This may be particularly true for vasopressin-receptors and its agonists, making treatment with it ideal for patients with less severe septic shock.

15 [0081] Furthermore, corticosteroids have been shown to reverse tachyphylaxis against exogenous vasopressors, providing an explanation for why vasopressin becomes more efficient in septic shock when used in conjunction with corticosteroids.

[0082] Patients with less severe septic shock have more reversible organ dysfunction and a more generous physiological reserve. Treatment with high doses of catecholamines may
20 lock patients in the more severe category by causing irreversible adverse damage that may limit the physiological reserve and vasopressor responsiveness. For example, vasopressin does not seem to improve the outcome for patients who have already sustained significant renal injury, compared to patients who do not have any signs of renal dysfunction at baseline.

25 [0083] As described herein, a vasopressin-receptor agonist may be used for the treatment of septic shock in patients who have less severe septic shock. The method may include one of the following:

[0084] A vasopressin-receptor agonist may be used for the treatment of septic shock in patients with septic shock requiring low dose vasopressor support at baseline (such as $\leq 14 \mu\text{g}$ norepinephrine /min).

5 [0085] A vasopressin-receptor agonist may be used for the treatment of septic shock in patients with septic shock and with low lactate levels (such as $\leq 1.4 \text{ mmol/L}$).

[0086] A vasopressin-receptor agonist may be used for the treatment of septic shock in patients with septic shock that require support from only one or less vasopressors at baseline.

10 [0087] A vasopressin-receptor agonist may be used for the treatment of septic shock that show no signs of renal dysfunction at baseline.

[0088] A vasopressin-receptor agonist may be used for the treatment of patients with septic shock at increased risk of developing renal failure or renal dysfunction.

15 [0089] A vasopressin-receptor agonist may be used for the treatment of patients with septic shock in conjunction with treatment using corticosteroids, including but not limited to the administration of one or more doses of cortisone, hydrocortisone, prednisone and methylprednisolone.

[0090] A vasopressin-receptor agonist may be used in combination with other vasopressors and/or any agents that may improve the outcome for the patient.

20 [0091] A vasopressin-receptor agonist may be infused into the subject, for example in a vein.

25 [0092] The dose of vasopressin, or a vasopressin receptor agonist may be expressed as an infusion rate e.g “per minute”. The quantity of vasopressin, or vasopressin receptor agonist may be described as “units” or abbreviated as “U”. A unit of vasopressin or a vasopressin receptor agonist is known in the art, and may be described as equivalent to the pressor activity of 0.5 mg of the USP Posterior-pituitary Reference Standard. As an example, 1 mg of synthetic vasopressin corresponds to 600 units. A subject may receive a dose from about 0.001 to about 0.1 units or any amount therebetween, or from about 0.001 to about 0.05 units or any amount therebetween, or from about 0.005 to about 0.004

units or any amount therebetween, or from about 0.01 to about 0.03 units or any amount therebetween. A suitable dose may be determined based on standard protocols used in the particular institution or medical facility, or may be provided by the manufacturer or supplier of the vasopressin or vasopressin receptor agonist, or other drug used for the treatment of the subject. Alternately, a suitable dose may be determined empirically and adjusted by a practitioner as needed. The specific units by which a dose is described is not critical – one of skill in the relevant art will readily be able to interconvert the units as necessary, given the conditions of treatment, the manner in which the vasopressin or vasopressin receptor agonist is provided (e.g. concentration), the mode of administration, or the like.

[0093] The dose of a vasopressin-receptor agonist that is used may be low, such as corresponding to 0.01-0.03U of vasopressin /min.

[0094] A vasopressin-receptor agonist may be infused in the form of a liquid, for example corresponding to 0.12U vasopressin/mL.

[0095] The drugs, agonists and other pharmaceuticals that may be used for treatment of a subject with septic shock or less severe septic shock may be administered alone or in combination, in any of several ways - including, for example, continuous intravenous infusion, continuous intraarterial infusion, intermittent intravenous infusion, intermittent intraarterial infusion, subcutaneous injection, intraperitoneal injection, intramuscular injection, intravenous injection, epidermal or transdermal administration, mucosal membrane administration, orally, nasally, rectally, or vaginally. See, for example, Remington– The Science and Practice of Pharmacy, 21st edition. Gennaro et al editors. Lippincott Williams & Wilkins Philadelphia. Carrier formulations may be selected or modified according to the route of administration.

[0096] Dosage schedules may be dependent on, for example, the subject's condition, age, gender, weight, route of administration, formulation, or general health. Dosage schedules may be calculated from measurements of adsorption, distribution, metabolism, excretion and toxicity in a subject, or may be extrapolated from measurements on an experimental animal, such as a rat or mouse, for use in a human subject. Optimization of dosage and treatment regimens are discussed in, for example, Goodman & Gilman's The

Pharmacological Basis of Therapeutics 11th edition. 2006. LL Brunton, editor. McGraw-Hill, New York, or Remington– The Science and Practice of Pharmacy, 21st edition. Gennaro et al editors. Lippincott Williams & Wilkins Philadelphia.

[0097] One way of administering vasopressin or a vasopressin-receptor agonist is described in the following infusion protocol: Infusion may be started at 5 mL/hr, and increased over the first hour by 2.5 mL/hr every 10 minutes, to a constant target rate of 15 mL/hr while the bedside nurse can titrate open-label catecholamine vasopressors to maintain a constant target mean arterial pressure (MAP) (such as 65 – 75 mmHg). If this protocol is used, the vasopressin infusion would be starting at 0.01 U/min and titrated to a maximum of 0.03 U/min. Furthermore, if the target MAP is reached upon maximum infusion, open-label catecholamine vasopressors may not be needed. An initial target MAP of 65 - 75 mmHg is recommended; however, the target MAP may be set by the individual ICU physician. Infusion of study drug may be continued at 15 mL/hr until the patient improves such that open-label catecholamine vasopressors are no longer required, or until the patient develops serious adverse events or dies. Weaning of the study medication may for example be commenced when the target MAP has been maintained while off all vasopressors for a time-period such as eight hours. The infusion of vasopressin-receptor agonist may for example be decreased in 2.5 mL decrements every hour while the target MAP is maintained. In the event of hypertension despite the above weaning protocols (MAP > 85 mmHg), open-label catecholamine vasopressors and the vasopressin-receptor agonist may be weaned more rapidly.

[0098] The invention may further comprise a kit for use in treating a subject with septic shock, or less severe septic shock. The kit may comprise vasopressin, or a vasopressin-receptor agonist, and instructions for its use. The instructions may comprise steps involving assessment of the severity of the subject's sepsis or suspected sepsis, or SIRS, or other clinical events associated with sepsis. The instructions may provide information with respect to characteristics of a recommended subject, e.g. a subject with less severe septic shock as described herein, or a contraindicated subject. The kit may further provide a vasopressor for use in combination with the vasopressin or vasopressin-receptor agonist, along with information of reduced or alternate doses suitable for treatment of a subject with septic shock, or less severe septic shock.

EXAMPLE 1

[0099] Description of Study Procedures

[00100] This trial was conducted between July 2001 and April 2006 in 27 centers
5 in Canada, Australia, and the USA and was approved by the research ethics boards of all
participating institutions. Written informed consent was obtained from all patients, their
next of kin or another surrogate decision maker as appropriate.

[00101] *Patient selection criteria*

[00102] Patients greater than 16 years of age who had septic shock resistant to
10 fluids and low dose norepinephrine were considered for enrollment.

[00103] *SIRS criteria were:*

(1) fever ($>38^{\circ}\text{C}$) or hypothermia ($<36^{\circ}\text{C}$),

(2) tachycardia (heart rate >90 beats per minute),

(3) tachypnea (respiratory rate >20 breaths per minute or $\text{PaCO}_2 < 32$ mmHg) or need for
15 mechanical ventilation,

(4) abnormal leukocyte count ($>12,000$ cells/ mm^3 , <4000 cells/ mm^3 , or $>10\%$
immature [band] forms).

[00104] *Exclusion criteria*

(1) unstable coronary syndrome (acute myocardial infarction during this episode
20 of shock based on the combination of history, electrocardiogram, and enzyme changes
(as defined by investigator),

(2) greater than 24 hours had elapsed since the patient met entry criteria,

(3) use of open-label vasopressin for blood pressure support during the current
hospital admission,

(4) malignancy or other irreversible disease or condition for which six-month
25 mortality was estimated to be $\geq 50\%$,

(5) acute mesenteric ischemia either proven or suspected. A patient could be excluded by the investigator if, in their judgment, the condition was strongly suspected but not proven by conventional criteria or the attending physician had initiated presumptive therapy,

5 (6) death anticipated within 12 hours,

(7) underlying chronic heart disease (NYHA class III or IV) and shock,

(8) physician and team were not committed to aggressive care,

(9) severe hyponatremia (serum sodium < 130 mmol/L),

(10) traumatic brain injury (GCS < 8 prior to onset of sepsis),

10 (11) Raynaud's phenomenon, systemic sclerosis or vasospastic diathesis,

(12) pregnancy (positive serum β -HCG).

[00105] Other reasons for not enrolling included improving condition, next of kin unavailable, refusal of subject consent, refusal of physician consent, enrolled in another study, other.

[00106] *Drug Infusion*

[00107] The study drug infusion was started at 5 mL/hr, and increased over the first hour by 2.5 mL/hr every 10 minutes, to a constant target rate of 15 mL/hr while the bedside nurse titrated open-label catecholamine vasopressors to maintain a constant target MAP (65 – 75 mmHg). Thus, the blinded vasopressin infusion was started at 0.01 U/min and titrated to a maximum of 0.03 U/min while the blinded norepinephrine infusion was started at 5 μ g/min and titrated to a maximum of 15 μ g/min. An initial target MAP of 65 - 75 mmHg was recommended; however, the attending ICU physician could modify the target blood pressure in each patient. Infusion of study drug was continued at 15 mL/hr until the patient died, developed a serious adverse event, or improved such that open-label catecholamine vasopressors were no longer required. Neither crossover to the alternate arm nor open-label vasopressin was permitted.

[00108] Open-label catecholamine vasopressors were only increased if the target MAP was not reached on maximal study drug infusion. Weaning of open-label catecholamine vasopressors was only permitted when the target MAP had been reached

during the study drug infusion. Weaning of study drug was only commenced when the target MAP was maintained for eight hours without any open-label catecholamine vasopressors.

[00109] Study drug infusion was discontinued or interrupted if any of the following
5 predetermined serious adverse events occurred: (1) acute S-T segment elevation confirmed by 12-lead electrocardiogram, (2) serious or life-threatening (hemodynamically unstable) cardiac arrhythmias, (3) acute mesenteric ischemia, (4) digital ischemia, or (5) hyponatremia (serum sodium < 130 mmol/L). If the clinical team noted an adverse event that they considered to be related to study drug, then the study drug was discontinued for
10 at least eight hours and a serious adverse event was reported. Study drug could be restarted if, in the judgment of the investigator or attending physician, the adverse event had been treated, the condition had reversed and the event was not thought to be a result of the study drug or study protocol.

[00110] If vasopressor support was required during the same ICU admission after a
15 patient had been weaned from study drug, the study drug was preferentially re-infused, providing no exclusion criteria were met.

[00111] *Randomization and Blinding*

[00112] Treatment allocation, to either vasopressin or norepinephrine, was blinded
20 by a central phone-in randomization system accessed by the participating institutions' study pharmacists. A computer-generated randomization list of variable permuted blocks of 2, 4, and 6 was used for treatment allocation, which was stratified by (1) center, and (2) severity of shock in the hour prior to randomization (less severe septic shock stratum was defined as < 15 µg/min norepinephrine equivalent and more severe septic shock stratum was defined as ≥ 15 µg/min norepinephrine equivalent). Both study drug infusions were
25 prepared locally by unblinded study pharmacists. All clinical staff, investigators, research personnel, patients and families were blinded to the treatment allocation for the duration of the trial. If the patient developed serious adverse reactions, neither crossover to the alternate arm nor open-label vasopressin was permitted.

[00113] *Study drug details*

[00114] After receiving the allocation code from the central randomization system, study pharmacists at each center were responsible for preparation of the study drug. Both vasopressin (30 U) and norepinephrine (15 mg) were mixed in and infused from identical 250 mL D5W intravenous bags to give final concentrations of 0.12 U/mL of vasopressin and 60 µg/mL of norepinephrine.

[00115] *Co-interventions*

[00116] The effect of co-interventions was minimized because the medical teams were not aware of treatment allocation. Other aspects of managing septic shock and critical illness were at the discretion of the ICU team. However, important co-interventions potentially influencing the primary outcome were recorded.

[00117] *Weaning of vasopressors*

[00118] The weaning of open-label catecholamine vasopressors was allowed only when target mean arterial pressure (MAP) had been reached during the study drug infusion. Open-label catecholamine vasopressors were increased only if target MAP was not reached on maximal study drug infusion. When the study drug infusion was at 15 mL/hr, then open-label norepinephrine was weaned by 1-2 µg/min every 5 - 10 minutes while maintaining target MAP.

[00119] Weaning of the study medication was commenced when the target MAP was maintained while off all open-label catecholamine vasopressors for eight hours. The infusion of study medication was decreased in 2.5 mL decrements every hour while the target MAP was maintained. Time of discontinuation of study drug for at least 12 hours was recorded. In the event of hypertension despite the above weaning protocols (MAP > 85 mmHg), open-label catecholamine vasopressors and study drug could be weaned more rapidly.

[00120] If re-infusion of vasopressors was required once a patient had been weaned from vasopressor support, study drug was recommenced first. In the case of emergency situations that required re-infusion of vasopressors and the study drug was not immediately available, any open-label vasopressor (except open-label vasopressin) could be infused to maintain blood pressure until the study drug was available. An inability to

re-infuse study drug within 12 hours (e.g. study pharmacist unavailable) was reported as a protocol violation.

[00121] *Plasma vasopressin levels*

[00122] Blood for measurement of plasma vasopressin levels was collected at six
5 sites (St Paul's, Mount. Sinai, Ottawa General, Toronto General, The Alfred and The
Mayo Clinic hospitals) at five timepoints; baseline, 6, 24, 72 hours and 7 days. Blood was
collected in chilled EDTA bottles, cold centrifuged and the plasma stored at -70°C until
analysis. Vasopressin was analyzed after extraction on reversed phase column by double
antibody immunoassay (Buhlman Laboratories, Basel, Switzerland) at London
10 Laboratory Services Group, London, Ontario. The sensitivity limit of this assay is 0.39
pmol/L. The precision between runs was 11% at a mean of 1.9 pmol/L, 9.1% at a mean of
10.2 pmol/L, and 8.6% at a mean of 16.5 pmol/L. The within run variations were lower at
all levels.

[00123] Within the five collecting centers, 107 of 359 (29.8%) patients infused
15 with study drug had vasopressin assays done.

[00124] *Outcomes*

[00125] Study personnel visited patients in hospital and contacted patients or
healthcare professionals for participants who left the hospital to determine 28-day and 90-
day survival. Length of stay in the ICU and hospital was defined as the time spent in ICU
20 or hospital respectively during the index hospitalization. A patient was considered
discharged on the day of transfer home, to alternate care (off active care while in hospital)
or to a chronic care facility.

[00126] *Protocol / Data Monitoring*

[00127] To ensure compliance with the study protocol across the multiple sites and
25 verify accurate data collection a process of monitoring case report forms (CRF) was
undertaken. Eight trained monitors visited a representative sample of sites to verify
important data on the CRF against source documents. This included appropriate
ascertainment of consent, accurate application of inclusion and exclusion criteria,
compliance with study drug infusion (initiation, maintenance and weaning), 28-day and

90-day mortality, time to hemodynamic stability, protocol violations and any serious adverse events. Geographical and budgetary restrictions prevented 100% monitoring.

[00128] In total 454 (58.3%) charts in 16 sites in Canada and Australia were monitored. In general the quality of data collected on the CRF was very good, there was consistent interpretation of the protocol and CRF, and the determination of outcome was 100% correct. In summary two additional serious adverse events were captured, there were 21 minor variations in the consent documents including incorrect version of consent form used, no witness or investigator signatures, 10 patients were enrolled outside of the 24 hour inclusion window and one patient was found not to have a new additional organ failure on inclusion.

[00129] *Statistical analysis*

[00130] An independent Data and Safety Monitoring Committee (DSMC) evaluated two pre-planned interim analyses after 194 and 388 patients were enrolled. Sequential stopping rules for safety and efficacy were used according to the Lan-DeMets method¹⁴ using an O'Brien-Fleming approach.

[00131] We calculated that 776 patients were required to be enrolled, randomized and infused with study drug to detect an absolute 10% difference in mortality, assuming a mortality rate of 60% in the norepinephrine group and a two-sided alpha error of 5% and a power of 80%. Midway through the trial while blinded to all data, the Executive Committee, in conference with the DSMC, determined that patients who were randomized but never infused would not be included in the primary analysis, as their omission would be equally distributed between groups, would be unrelated to treatment allocation and would not bias outcome ascertainment.¹⁵ We increased the total number of patients enrolled to achieve the target sample size. Our data exclude with 95% confidence a harm of vasopressin of greater than 2.9% or a benefit of greater than 10.7%.

[00132] The primary outcome was 28-day all cause mortality.

[00133] Secondary outcomes were:

- (1) 90-day mortality
- (2) Days alive and free of organ dysfunction over the first 28 days according to

the Brussels criteria¹⁶ (Table 1). To further evaluate cardiovascular, respiratory and renal function, we also analyzed days alive and free of vasopressor use, mechanical ventilation, and renal replacement therapy.

(3) Days alive and free of SIRS, defined as free of two or more of the four SIRS inclusion criteria.

(4) Days alive and free of steroid use.

(5) ICU and hospital length of stay.

Patients were also analyzed according to the *a priori* strata of more severe and less severe septic shock.

Table 1. BRUSSELS ORGAN DYSFUNCTION DEFINITIONS

Organs	Normal	Mild	Clinically Significant Organ Dysfunction		
			Moderate	Severe	Extreme
Cardiovascular (systolic blood pressure, mmHg)	> 90	≤ 90 fluid responsive	≤ 90 not fluid responsive	≤ 90 pH 7.3	≤ 90 pH 7.2
Pulmonary (PaO ₂ /FiO ₂ , mmHg)	> 400	400 - 301	300 – 201	200 – 101	100
Neurologic (GCS)	15	14 - 13	12 – 10	9 – 6	≤ 5
Coagulation (platelet count, x10 ³ /mm ³)	> 120	120 - 81	80 – 51	50 – 21	≤ 20
Renal (creatinine, μmol/L [mg/dL])	<133 [<1.5]	133 – 175 [1.5-1.9]	176 – 300 [2.0-3.4]	301 – 442 [3.5-4.9]	≥443 [≥5.0]
Hepatic (bilirubin, μmol/L [mg/dL])	< 20 [<1.2]	20 – 32 [1.2-1.9]	33 – 99 [2.0-5.9]	100 – 199 [6.0-11.9]	≥200 [≥12.0]

[00134] Days alive and free (DAF) calculations: DAF was scored as 1 if the patient was alive and free of organ dysfunction (normal or mild dysfunction). DAF was scored as 0 if the patient had organ dysfunction (moderate, severe, or extreme) or was not alive. A low DAF score indicates more organ dysfunction because a low score indicates fewer days alive and free of organ dysfunction. Each of the 28 days after meeting the inclusion criteria was scored. For any 24-hour period in which there is no measurement of a variable, we carried forward the value from the previous 24-hour period. If any variable was never measured, it was assumed to be normal. Once a patient was discharged home they were considered free of organ or failure.

[00135] The data analyst and investigators remained blinded to treatment allocation while undertaking the final analyses. The primary analysis comparing 28-day mortality between the two treatment groups was performed using an unadjusted Chi-square test and all patients were analyzed according to the treatment group allocated at randomization.

5 Results are presented as absolute and relative risks and 95% confidence intervals. Kaplan-Meier curves describing the estimated probability of survival in the two treatment arms as a function of time from admission into the study were compared using the log-rank test statistic. Because of the complex nature of septic shock patients and to adjust for any imbalances between the two treatment groups at baseline, a logistic regression procedure
10 was used to adjust raw values for 28 day mortality using significant covariates predicting outcomes. Age, illness severity (APACHE II score at baseline), serious co-morbid illness and other baseline covariates predicting outcome (at a threshold p value of 0.2) were entered into the model. Results are presented as odds ratios (OR) and 95% confidence intervals.

15 [00136] The treatment effect within each subgroup was assessed by the within stratum analysis, chi-squared test. Thus, the *a priori* test we defined was analysis by subgroup (not an interaction statistic). We also tested for the interaction of stratum and treatment to determine if there was a differential effect on mortality using logistic regression analysis. Secondary outcomes including organ dysfunction analysis and
20 adverse event rates were compared using parametric procedures (independent t-test), non parametric procedures (Wilcoxon Rank Sum test) or the Fisher exact test. Analysis was conducted using SAS software (version 9.1.3) and all p values were 2 sided.

[00137] Results and interpretation

[00138] The DSMC recommended that the study be continued without protocol
25 modification after both interim analyses. Of 6229 screened patients, 802 were randomized after obtaining informed consent (**Figure 1**). Of these 802 patients, two withdrew consent following study drug infusion and 21 patients were not infused for various reasons (**Figure 1**). Reasons for not infusing patients allocated to norepinephrine include Acute MI/elevated troponin (6), norepinephrine requirements dropped <5 ug/min (3),
30 transferred to OR before study drug could be given, >24hrs elapsed (2), Open-label vasopressin given (1), death (1). Reasons for not infusing patients allocated to vasopressin

include Vasopressin requirements dropped $<5 \mu\text{g}/\text{min}$)5), Acute MI / Elevated (2),
Withdrawal of care (1).

[00139] One patient was lost to follow-up before day 28; thus 779 patients were
randomised and infused with study drug; 778 were included in the final primary analysis:
5 396 in the vasopressin and 382 in the norepinephrine group, respectively (**Figure 1**).

Baseline characteristics were well matched between the two groups (**Table 2**). Enrolled
patients were severely ill as indicated by the APACHE II scores³, proportion with new
organ dysfunction, serum lactate levels and norepinephrine infusion rates at study entry.
There was no difference in baseline vasopressor combinations, tidal volumes used, use of
10 corticosteroids or the use of activated protein C between vasopressin and norepinephrine
groups.

[00140] Blood pressure and heart rate in both treatment groups throughout study
are shown in **Figure 2**. The mean infusion rates of study drug were within 2 mL/hr
between treatment groups over the first five days. The rates of norepinephrine infusion in
15 the vasopressin compared to the norepinephrine group are shown in **Figure 4**.

[00141] There was no significant difference in the primary outcome, 28-day
mortality, between the vasopressin-treated group (35.4%) and the norepinephrine-treated
group (39.3%, $P = 0.26$) (**Table 3, Figure 3**). Similarly there was no significant difference
20 in mortality at 90 days (43.9% vasopressin group vs. 49.6% norepinephrine group, $P =$
0.11) or organ dysfunction rates (**Table 3**). The results remained non-significant after
multivariate logistic regression analysis (vasopressin 28-day mortality OR 0.88, 95% CI
0.62 - 1.26, 90-day mortality OR 0.81, 95% CI 0.57 - 1.16).

TABLE 2. DEMOGRAPHICS AND BASELINE CHARACTERISTICS OF PATIENTS

Characteristic	Norepinephrine group N (%) (N = 382)	VASOPRESSIN GROUP N (%) (N = 397)
Age (years)	61.8 ± 16	59.3 ± 16.4
Male Gender	229 (59.9)	246 (62.0)
Recent surgical history	132 (34.6)	151 (38.0)
Elective	8 (2.1)	6 (1.5)
Emergency	124 (32.5)	145 (36.5)
APACHE II score	27.1 ± 6.9	27.0 ± 7.7
Ethnicity*		
Caucasian	320 (83.8)	336 (84.6)
Pre-existing conditions		
Ischemic Heart Disease	65 (17.0)	68 (17.1)
Congestive Heart Failure	30 (7.9)	28 (7.1)
COPD	72 (18.8)	55 (13.9)
Chronic Renal Failure	48 (12.6)	40 (10.1)
Diabetes	88 (23.0)	77 (19.4)
Liver Disease	36 (9.4)	52 (13.1)
Alcoholism	53 (13.9)	55 (13.9)
Injection Drug Abuse	14 (3.7)	20 (5.0)
Cancer	104 (27.2)	85 (21.4)
Immunocompromised	72 (18.8)	67 (16.9)
Solid Organ Transplant	17 (4.5)	14 (3.5)
Steroid use	86 (22.5)	82 (20.7)
Recent Trauma	16 (4.2)	23 (5.8)
New Organ Failure		
Cardiovascular	382 (100)	397 (100)
Respiratory	341 (89.3)	342 (86.1)
Renal	258 (67.5)	264 (66.5)
Hematology/Coagulation	84 (22.0)	118 (29.7)
Neurologic	89 (23.3)	101 (25.4)
Number of organ dysfunctions	2.5 ± 1.1	2.6 ± 1.1
Source of Infection		
Lung	165 (43.2)	162 (40.8)
Abdomen	100 (26.2)	111 (28.0)
Other**	117 (30.6)	124 (31.2)
Pathogen Type in Cultures		
Gram positive alone	59 (15.4)	80 (20.2)
Gram negative alone	43 (11.3)	40 (10.1)
Mixed Organisms	139 (36.4)	143 (36.0)
Other	51 (13.4)	62 (15.7)

No pathogen	90 (23.6)	72 (18.1)
Hemodynamic variables		
Systolic Blood Pressure (mm Hg)	110 ± 17	108 ± 17
Mean Arterial Pressure (mm Hg)	73 ± 10	72 ± 9
Arterial pH	7.31 ± 0.1	7.32 ± 0.1
Serum lactate (mmol/L)	3.5 ± 3.0	3.5 ± 3.2
Vasoactive Drug Dosage at Randomization		
Norepinephrine (µg/min)	20.7 ± 18.1	20.7 ± 22.1
[µg/kg/min]	[0.28 ± 0.26] (n=329)	[0.26 ± 0.27] (n=344)
Epinephrine (µg/min)	8.6 ± 9.4	14.6 ± 16.9
[µg/kg/min]	[0.12 ± 0.15] (n=23)	[0.20 ± 0.29] (n=27)
Dopamine (µg/kg/min)	7.3 ± 5.3 (n=28)	7.6 ± 6.4 (n=26)
Dobutamine (µg/kg/min)	5.1 ± 3.7 (n=41)	6.4 ± 5.2 (n=48)
Milrinone (µg/kg/min)	0.4 ± 0.3 (n=10)	0.3 ± 0.1 (n=13)
Phenylephrine (µg/min)	151 ± 74	157 ± 90
[µg/kg/min]	[2.03 ± 1.17] (n=71)	[2.04 ± 1.31] (n=85)
Vasoactive Drug infusions		
Norepinephrine alone	222 (58.1)	224 (56.4)
No Norepinephrine	53 (13.9)	53 (13.4)
Two or more vasopressors	111 (29.1)	124 (31.2)
Time from meeting inclusion criteria to study drug infusion (hours)	11.5 ± 9.4	11.9 ± 8.9

Plus-minus values are means ± SD. COPD denotes chronic obstructive pulmonary disease, and APACHE II Acute Physiology and Chronic Health Evaluation II³

*Ethnicity was determined by the local study coordinators

**Other sites of infection included the blood, skin, central nervous system, bones and joints, cardiac system and reproductive organs.

Table 3. Analysis of the rates and risks of death from any cause

and secondary outcomes*

	NOR- EPINEPHRINE GROUP	VASOPRESSIN GROUP	P VALUE**	ARR *** % (95% CI)	RR **** (95% CI)
	N / total N (%)				
All randomized patients					
28-day mortality	154/395 (39.0)	144/404 (35.6)	0.33	3.3 (-3.4 to 10.1)	0.91 (0.76 to 1.09)

90-day mortality	194/392 (49.5)	177/400 (44.3)	0.14	5.2 (-1.7 to 12.2)	0.89 (0.77 to 1.04)
All randomized and infused patients					
28-day mortality	150/382 (39.3)	140/396 (35.4)	0.26	3.9 (-2.9 to 10.7)	0.90 (0.75 to 1.08)
90-day mortality	188/379 (49.6)	172/392 (43.9)	0.11	5.7 (-1.3 to 12.8)	0.88 (0.76 to 1.03)

Variable	Norepinephrine group (N=382)	VASOPRESSIN GROUP (N=396)	P Value
<u>Days alive and free of organ dysfunction*****</u>			
Cardiovascular	17 (0, 24)	19 (0, 24)	0.58
Vasopressor use	17 (0, 24)	19 (0, 24)	0.61
Respiratory	2 (0, 14)	3.5 (0, 16)	0.15
Ventilation	6 (0, 20)	8.5 (0, 20)	0.24
Renal	18.5 (3, 28)	21.5 (4, 28)	0.54
Renal replacement therapy	23 (5, 28)	25 (6, 28)	0.64
Hepatic	24.5 (3, 28)	25 (5, 28)	0.80
Hematologic	23 (3, 28)	24 (5, 28)	0.48
Neurologic	15 (0, 24)	15 (0, 24)	0.57
Days alive and free of any organ failure	0 (0, 6)	0 (0, 9)	0.14
Days alive and free of SIRS (2/4 criteria)	6 (0, 15)	6 (0, 18)	0.21
Days alive and free of steroid use	13.5 (1, 24)	16 (1, 25)	0.33
ICU length of stay	16 (8, 32)	15 (7, 29)	0.14
Hospital length of stay	26 (15, 53)	27 (13, 52)	0.23

* Patients were analyzed in the treatment group to which they were assigned at randomization

** Two-sided P values are based on Pearson's chi-squared tests.

***ARR denotes absolute risk reduction (mortality rate in norepinephrine group minus mortality rate in vasopressin group). CI denotes confidence interval.

****RR denotes relative risk

*****Days alive and free calculations: Organ dysfunction for each organ system is defined as being present during each 24-hour period if there was evidence of moderate, severe, or extreme organ dysfunction according to the Brussels criteria.¹⁶ A low score

indicates more organ dysfunction because a low score indicates fewer days alive and free of organ dysfunction. Values are median (interquartile range), *P* values are based on Wilcoxon Rank Sum test.

[00142] There were no significant differences in the overall rates or specific categories of serious adverse events in the vasopressin and norepinephrine groups (overall rates 10.3% and 10.5%, respectively, *P* = 1.0) (Table 4). There was a trend to a higher rate of digital ischemia in the vasopressin compared to norepinephrine group (2% versus 0.5%, *p* = 0.11).

TABLE 4. SERIOUS ADVERSE EVENTS IN PATIENTS WHO HAD SEPTIC SHOCK

Variable	Norepinephrine group N (%)	VASOPRESSIN GROUP N (%)	P Value*
At least one serious adverse event	40 (10.5)	41 (10.3)	1.00
Acute myocardial infarction / ischemia	7 (1.8)	8 (2.0)	1.00
Cardiac arrest	8 (2.1)	3 (0.8)	0.14
Life-threatening arrhythmia	6 (1.6)	8 (2.0)	0.79
Acute mesenteric ischemia	13 (3.4)	9 (2.3)	0.39
Hyponatremia (serum Na < 130mmol/L)	1 (0.3)	1 (0.3)	1.00
Digital ischemia	2 (0.5)	8 (2.0)	0.11
Cerebro-vascular accident	1 (0.3)	1 (0.3)	1.00
Other	2 (0.5)	5 (1.3)	0.45

* TWO-SIDED *P* VALUES ARE BASED ON THE FISHER EXACT TEST

[00143] The overall rates of serious adverse events were low in the vasopressin and norepinephrine groups (approximately 10% in each). Previous studies raised the possibility that vasopressin infusion may increase the incidence of cardiac arrest.⁵ In contrast, we found that of 11 cardiac arrests reported in this study, eight occurred in the norepinephrine group while three occurred in the vasopressin group. Our selection of a low dose of vasopressin (0.03 U/min) and careful exclusion of patients who had acute coronary syndromes or severe heart failure could account for the lack of adverse cardiovascular effects of vasopressin infusion. If vasopressin becomes routine therapy and

is given to septic shock patients with co-morbid heart disease, the adverse reactions to vasopressin could be increased. Other reported adverse effects of both vasopressin and norepinephrine include decreased cardiac output,^{5, 10, 17} mesenteric ischemia,^{18, 19} hyponatremia (vasopressin), skin necrosis^{10, 20} and digital ischemia.²¹ More patients in the vasopressin group had digital ischemia compared to the norepinephrine group (2.0% vs 0.5%, p=0.11); one patient in the vasopressin group required surgical intervention.

[00144] Plasma vasopressin levels were extremely low at baseline (median 3.2 pmol/L, interquartile range 1.7-4.9 pmol/L) and did not change in the norepinephrine group. Low dose vasopressin infusion increased vasopressin levels to medians of 73.6 pmol/L (interquartile range 58.6 to 94.7 pmol/L) at six hours and 98.0 pmol/L (interquartile range 67.1 to 127.8 pmol/L) at 24 hours (**Figure 5**).

[00145] Baseline characteristics of the more severe and less severe strata are presented in **Table 5**. In patients who had less severe septic shock (5 – 14 µg/minute norepinephrine infusion at randomization), the vasopressin group compared to the norepinephrine group had lower 28-day mortality (26.5% and 35.7% respectively, P=0.05) and 90-day mortality (35.8% and 46.1% respectively, P=0.04, **Table 6**). In contrast, there were no differences in mortality between vasopressin and norepinephrine in the more severe septic shock stratum ($\geq 15\mu\text{g}/\text{min}$ norepinephrine) (28-day mortality 44.0% and 42.5% respectively, P=0.76, and 90-day mortality 51.8% and 52.8% respectively, P=0.84, **Table 6**). The test for the interaction of treatment group by severity of shock subgroup was not significant (P=0.10). To assess the robustness of the subgroup results we performed several post-hoc secondary analyses of the results according to different indicators of illness severity (**Table 7**). There was a significant interaction of treatment and subgroups by lactate quartile (P=0.04) and the number of vasopressors used at baseline (P=0.04). In patients in the lowest lactate quartile, ≤ 1.4 mmol/L, the vasopressin group had a lower 28-day mortality than the norepinephrine group (18.9% v 33.8% respectively, P=0.03, **Table 7**), whereas there was no difference in mortality between the vasopressin and norepinephrine groups in the higher lactate quartiles. Similarly there was a lower mortality in the vasopressin group compared to the norepinephrine group (31.3% v 39.9% respectively, P=0.04, **Table 7**) in patients receiving only one vasopressor at

baseline where as there was no difference in mortality between the vasopressin and norepinephrine groups if two or more vasopressors were used.

TABLE 5. DEMOGRAPHICS AND BASELINE CHARACTERISTICS OF PATIENTS ACCORDING TO SEVERITY OF SEPTIC SHOCK

Characteristic	Less severe septic shock	More severe septic shock	P VALUE
	N (%)	N (%)	
	N = 378	N = 401	
Age (years)	60.5 ± 16.2	60.6 ± 16.3	0.99
Male Gender	229 (60.6)	246 (61.3)	0.83
Recent surgical history	141 (37.3)	142 (35.4)	0.59
APACHE II score	25.3 ± 7.2	28.7 ± 7.1	<0.001
Ethnicity Caucasian	324 (85.7)	332 (82.8)	0.36
Time from meeting inclusion criteria to study drug infusion (hours)	12.3 ± 10.3	11.2 ± 7.9	0.08
<u>Pre-existing conditions</u>			
Ischemic Heart Disease	65 (17.2)	68 (17.0)	0.62
Congestive Heart Failure	36 (9.5)	22 (5.5)	0.30
COPD	74 (19.6)	53 (13.2)	0.04
Chronic Renal Failure	40 (10.6)	48 (12.0)	0.51
Diabetes	73 (19.3)	92 (22.9)	0.28
Liver Disease	43 (11.4)	45 (11.2)	0.62
Alcoholism	61 (16.1)	47 (11.7)	0.13
Injection Drug Abuse	22 (5.8)	12 (3.0)	0.10
Cancer	89 (23.5)	100 (24.9)	0.56
Immunocompromised	61 (16.1)	78 (19.5)	0.30
Solid Organ Transplant	15 (4.0)	16 (4.0)	0.62
Steroid use	86 (22.8)	82 (20.4)	0.47
Recent Trauma	18 (4.8)	21 (5.2)	0.76
<u>New Organ Failure</u>			
Cardiovascular	378 (100)	401 (100)	1.0
Respiratory	331 (87.6)	352 (87.8)	0.85
Renal	231 (61.1)	291 (72.6)	0.001
Hematology/Coagulation	85 (22.5)	117 (29.2)	0.03
Neurologic	82 (21.7)	108 (26.9)	0.15
Number of organ dysfunctions	2.4 ± 1.1	2.7 ± 1.1	<0.001
<u>Source of Infection</u>			
Lung	177 (46.8)	150 (37.4)	0.01
Abdomen	92 (24.3)	119 (29.7)	0.09
Other*	109 (28.8)	132 (32.9)	0.22
<u>Pathogen Type in positive cultures</u>			
Gram positive alone	64 (16.9)	75 (18.7)	0.54

Gram negative alone	34 (9.0)	49 (12.2)	0.54
Mixed organisms	144 (38.1)	138 (34.4)	0.54
Other & unknown	136 (36.0)	139 (34.7)	0.54
<u>Hemodynamic variables</u>			
Systolic Blood Pressure (mm Hg)	111.6 ± 16.2	107.2 ± 17.0	<0.001
Mean Arterial Pressure (mm Hg)	74.0 ± 9.2	71.6 ± 9.7	0.03
Arterial pH	7.35 ± 0.08	7.29 ± 0.11	0.11
Serum lactate (mmol/L)	2.4 ± 2.0	4.3 ± 3.6	<0.001
<u>Vasoactive Drug Dosage at Randomization</u>			
Norepinephrine (µg/min)	9.8 ± 5.5 (n=310)	30.0 ± 23.3 (n=363)	<0.001
Epinephrine (µg/min)	7.8 ± 6.1 (n=8)	12.6 ± 15.1 (n=42)	0.14
Dopamine (µg/kg/min)	7.3 ± 6.0 (n=19)	7.5 ± 5.8 (n=35)	0.92
Dobutamine (µg/kg/min)	4.7 ± 3.1 (n=24)	6.2 ± 5.0 (n=65)	0.09
Milrinone (µg/kg/min)	0.3 ± 0.1 (n=17)	0.4 ± 0.3 (n=16)	0.36
Phenylephrine (µg/min)	110 ± 71 (n=68)	188 ± 76 (n=88)	<0.001

Plus-minus values are means ± SD. COPD denotes chronic obstructive pulmonary disease, and APACHE II Acute Physiology and Chronic Health Evaluation II

**Other sites of infection included the blood, skin, central nervous system, bones and joints, cardiac system and reproductive organs*

Table 6. ANALYSIS OF THE RATES AND RISKS OF DEATH FROM ANY CAUSE BY SEVERITY OF SHOCK

Variable	Nor- epinephrine group	VASOPRESSIN GROUP	P- Value **	ARR *** % (95% CI)	RR **** (95% CI)
	N / total N (%)				
More severe septic shock stratum*					
28-day mortality	85/200 (42.5)	88/200 (44.0)	0.76	-1.5 (-11.2 to 8.2)	1.04 (0.83 to 1.3)
90-day mortality	105/199 (52.8)	103/199 (51.8)	0.84	1.0 (-8.8 to 10.8)	0.98 (0.81 to 1.18)
Less severe septic shock stratum*					
28-day mortality	65/182 (35.7)	52/196 (26.5)	0.05	9.2 (-0.1 to 18.5)	0.74 (0.55 to 1.01)
90-day mortality	83/180 (46.1)	69/193 (35.8)	0.04	10.4 (0.4 to 20.3)	0.78 (0.61 to 0.99)

*More and less severe septic shock were defined as requiring $\geq 15 \mu\text{g}/\text{min}$ or $5\text{-}14 \mu\text{g}/\text{min}$ norepinephrine equivalent at the time of randomization, respectively.

** Two-sided P values are based on Pearson's chi-squared tests.

***ARR denotes absolute risk reduction (mortality rate in norepinephrine group minus mortality rate in vasopressin group). CI denotes confidence interval.

****RR denotes relative risk

Table 7. Analysis of the Rates and Risks of Death from Any Cause in patients having Septic

Shock according to additional post-hoc subgroups

Post-hoc subgroups		NOREPINEPHRINE GROUP N / total N (%)	VASOPRESSIN GROUP N / total N (%)	P VALUE*	INTERACTION STATISTIC P VALUE**
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Treatment by lactate Quartile	First lactate quartile (≤ 1.4 mmol/L)	26/77 (33.8)	17/90 (18.9)	0.03	0.04
	Second lactate quartile (1.5 – 2.3 mmol/L)	36/87 (41.4)	35/85 (41.2)	0.98	0.65
	Third lactate quartile (2.4 – 4.4 mmol/L)	30/77 (39.0)	31/85 (36.5)	0.74	0.51
	Fourth lactate quartile (>4.4 mmol/L)	47/83 (56.6)	49/80 (61.3)	0.55	-
Treatment by number of vasopressors at baseline	One vasopressor	108/271 (39.9)	85/272 (31.3)	0.04	0.04
	Two or more vasopressors	42/111 (37.8)	55/124 (44.4)	0.31	-
Treatment by use of norepinephrine at baseline	Norepinephrine	123/329 (37.4)	120/343 (35.0)	0.52	0.31
	No norepinephrine	27/53 (50.9)	20/53 (37.7)	0.17	-
Treatment by pathogen identification	Pathogen identified	112/292 (38.4)	117/324 (36.1)	0.56	0.35
	No Pathogen identified	38/90 (42.2)	23/72 (31.9)	0.18	-
Treatment by APACHE II Quartiles	APACHE II <22	23/95 (24.2)	24/116 (20.7)	0.54	0.73
	APACHE II 23-27	39/119 (32.8)	32/92 (34.8)	0.76	0.59
	APACHE II 28-32	34/81 (42.0)	34/100 (34.0)	0.27	0.57
	APACHE II >32	54/87 (62.1)	50/88 (56.8)	0.48	-
Treatment by APACHE II ≤ 27 versus >27	APACHE II ≤ 27	62/214 (29.0)	56/208 (26.9)	0.64	0.50
	APACHE II >27	88/168 (52.4)	84/188 (44.7)	0.15	-
Treatment by Time to Study Drug Infusion	≤ 12 hrs	90/222 (40.5)	68/205 (33.2)	0.12	0.28
	>12 hrs	60/160 (37.5)	72/191 (37.7)	0.97	-

* Within subgroup analysis, two-sided P values are based on Pearson's chi-squared tests.

**Interaction statistic was calculated using logistic regression.

[00146] As presented in **Table 8**, vasopressin decreases the need for use of corticosteroids (as scored by more days alive and free of steroids) ($p = 0.06$) in less severe septic shock. This discovery is important because steroids have side effects such as increased risk of infection and increased risk of severe muscle weakness (also known as steroid neuromyopathy). Furthermore, as presented in **Table 9** vasopressin decreases the mortality of patients who have septic shock who are also treated with steroids, irrespective of the severity of septic shock. This is important because it may indicate a novel method to increase the efficacy of steroids in septic shock.

[00147] Other results presented in **Table 8** show that vasopressin decreases the need for mechanical ventilation (as scored by more days alive and free of ventilation) ($p = 0.09$) and decreases any organ failure (as scored by more days alive and free of any organ failure) ($p = 0.003$), systemic inflammatory response syndrome (SIRS) (as scored by more days alive and free of SIRS) ($p = 0.02$), respiratory dysfunction (as scored by more days alive and free of respiratory dysfunction) ($p = 0.01$) and hematologic dysfunction (as scored by more days alive and free of hematologic dysfunction) ($p = 0.10$) in less severe septic shock.

[00148] Several attributes of the stratified analysis should be considered. First, the criteria were prospectively defined and stratification into less or more severe shock occurred prior to randomization. Second, each stratum was large representing about one half of the study population. Third, the effect size was large – vasopressin was associated with an absolute mortality reduction of approximately 10% in the less severe shock stratum. Fourth, the result is biologically plausible. Finally, the results of the lactate quartile and numbers of vasopressors analyses support the idea that vasopressin may decrease mortality in patients who have less severe septic shock since vasopressin decreased mortality in the first (but not the second to fourth) lactate quartile and decreased mortality in patients receiving only one vasopressor (but not two or more) .

TABLE 8. SHOCK SEVERITY SUBGROUP ANALYSES

VARIABLE	LESS SEVERE STRATUM*		MORE SEVERE STRATUM*		P VALUE
	Norepinephrine group	VASOPRESSIN GROUP	Norepinephrine group	VASOPRESSIN GROUP	
Days alive and free of organ dysfunction**					
Cardiovascular	19 (1, 24)	21 (3.5, 24)	14 (0, 23)	12 (0, 23)	0.62
Vasopressor use	19 (1, 24)	21 (4,25)	14 (0, 24)	12.5 (0,24)	0.63
Respiratory	2 (0, 14)	6 (0,17)	2 (0, 14)	1 (0, 14)	0.65
Ventilation	10 (0, 21)	13 (0.5, 22)	3 (0, 19)	2 (0, 18)	0.87
Renal	24 (7, 28)	26 (11, 28)	13 (1, 28)	15.0 (1, 27)	0.49
Renal replacement therapy	28 (9, 28)	28 (13, 28)	19 (2.5, 28)	15.5 (1, 28)	0.44
Hepatic	27 (9, 28)	27.5 (13, 28)	22 (2, 28)	18 (1, 28)	0.23
Hematologic	25 (9, 28)	27 (14.5, 28)	22 (2, 28)	20 (1, 28)	0.38
Neurologic	15.5 (1, 24)	18 (4,24)	15 (0, 25)	7 (0, 23)	0.04
Days alive and free of any organ failure	0 (0, 4)	0 (0,12.5)	0 (0, 8)	0 (0, 4)	0.33
Days alive and free of SIRS (2/4 criteria)	7 (0, 16)	10 (1, 19)	5 (0, 15)	3 (0, 15)	0.62
Days alive and free of steroid use	15 (2, 28)	20 (7, 28)	12 (0, 22)	9 (0, 23)	0.52
ICU length of stay	17 (11, 33)	19 (10, 38)	15 (7, 32)	14 (6, 25.5)	0.33
Hospital length of stay	31 (16, 61)	31 (17, 67)	23 (10, 50)	24 (10, 43)	0.89

*More and less severe septic shock were defined as requiring ≥ 15 $\mu\text{g}/\text{min}$ or 5-14 $\mu\text{g}/\text{min}$ norepinephrine equivalent at the time of randomization, respectively.

** Days alive and free calculations: Organ dysfunction for each organ system is defined as being present during each 24-hour period if there was evidence of moderate, severe, or extreme organ dysfunction according to the Brussels criteria.¹⁶ A low score indicates more organ dysfunction because a low score indicates fewer days alive and free of organ dysfunction. Values are median (interquartile range), P values are based on Wilcoxon Rank Sum test.

Table 9. COMPARISON OF MORTALITY IN PATIENTS WITH OR WITHOUT STEROID TREATMENT

TREATMENT	MORTALITY (28 DAY) (%)	
	NOREPINEPHRINE	VASOPRESSIN
Steroids	44 %	35 %
No steroids	21 %	33 %

Interaction p value is < 0.05

EXAMPLE 2

Description of Study Procedures

[00149] All patients from the Vasopressin And Septic Shock Trial (VASST) were studied.

[00150] Acute renal failure was defined according to the consensus RIFLE serum creatinine criteria (urine output data was not available):

R-Risk; Increased serum creatinine x1.5 patient baseline

I-Injury; Increased serum creatinine x2 patient baseline

F-Failure; Increased serum creatinine x3 patient baseline or if patient baseline creatinine is $\geq 350 \mu\text{mol/l}$ (4mg/dl) and acute rise is $\geq 44 \mu\text{mol/l}$ (0.5 mg/dl).

L-Loss; Complete loss of renal function (dialysis) for at least 4 weeks.

E-End stage; Renal failure (dialysis) for at least 3 months.

[00151] If baseline serum creatinine levels were not available then they were calculated using the modification of diet renal disease (MDRD) formula⁴.

[00152] Outcome (28-day mortality) between vasopressin and norepinephrine treated groups was compared using a chi-square test. Multivariate logistic regression was used to adjust for potential confounders.

[00153] Results and interpretation

[00154] 779 patients were enrolled, randomized and infused with study drug. Renal function at enrolment according to rifle criteria is described in **Figure 7**. Whereas 40% of patients had no renal dysfunction at enrolment, 14% were at risk of developing renal dysfunction. At enrolment, 17% of patients had renal injury, 23% had renal failure and 6% had end-stage renal disease.

[00155] The overall 28-day mortality was 37.3% (290/778; 1 patient was lost to follow-up). 28-day mortality increases according to baseline RIFLE criteria (**Figure 8**). The presence of any renal dysfunction was associated with increased mortality compared to no renal dysfunction (44.3% vs 27.0%, respectively, $p < 0.001$). Furthermore, mortality was reduced in patients at risk of renal failure and treated with vasopressin, compared to norepinephrine at 28 days (30.8% and 54.7%, $p = 0.013$; **Table 11**) as well as at 90 days (37.3% and 62.3%, $p = 0.011$). In septic shock patients at risk of renal dysfunction, the risk of dying (adjusted for age, sex, APACHE II score and severity of septic shock as potential confounders) was significantly lower in patients treated with vasopressin, compared to norepinephrine (odds ratio 0.38, 95% confidence interval 0.17-0.88, $p = 0.024$). The rate of progression to renal failure or loss in patients at risk of renal dysfunction was significantly reduced in patients treated with vasopressin, compared to norepinephrine ($p = 0.028$; **Figure 9**).

[00156] Therefore, vasopressin treatment is associated with a reduction in mortality and progression to renal failure in patients at risk. Vasopressin does not seem to improve the outcome for patients who have already sustained significant renal injury.

Table 10. Baseline characteristics according to RIFLE criteria.

	No renal dysfunction N = 315	Risk N = 106	Injury N = 130	Failure N = 179	End-stage N = 49
Age	57.8 ± 16.6	61.2 ± 16.8	64.0 ± 14.8	61.9 ± 16.4	62.3 ± 13.3
Sex – male	203 (64.4)	69 (65.1)	75 (57.7)	99 (55.3)	29 (59.2)
APACHE II	23.7 ± 6.4	27.0 ± 6.2	28.0 ± 7.7	31.3 ± 6.5	31.5 ± 6.1
Severe shock (NE ≥ 15µg/min)	131 (41.6)	66 (62.3)	75 (57.7)	105 (58.7)	24 (49.0)
Serum creatinine at enrollment (µmol/l)	101 ± 66	154 ± 34	205 ± 39	321 ± 123	472 ± 207

Values in table are mean ± SD or N (%)

Table 11. 28-day mortality according to treatment and RIFLE criteria.

	Norepinephrine group mortality N (%)	Vasopressin group mortality N (%)	P-value
No renal dysfunction N = 315	45 (28.1)	40 (25.8)	0.643
Risk N = 105	29 (54.7)	16 (30.8)	0.013
Injury N = 130	22 (35.5)	29 (42.6)	0.473
Failure N = 179	39 (47.6)	43 (44.3)	0.666
End-stage N = 49	15 (60.0)	12 (50.0)	0.482

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[00157] All citations are herein incorporated by reference.

[00158] One or more currently preferred embodiments have been described by way of example. It will be apparent to persons skilled in the art that a number of variations and modifications can be made without departing from the scope of the invention as defined in the claims.

WHAT IS CLAIMED IS:

1. A method of treating a subject having less severe septic shock, the method comprising administering to the subject a drug selected from the group consisting of vasopressin, arginine vasopressin (AVP), lysine vasopressin (LVP), triglycil-
5 lysine vasopressin, octopressin, ornipressin, and desmopressin (DDAVP), for a sufficient time to treat less severe septic shock.
2. The method of claim 1 further comprising administering to the subject one or more corticosteroids.
3. A method of preventing organ failure in a subject having septic shock, the method
10 comprising administering to the subject a drug selected from the group consisting of vasopressin, arginine vasopressin (AVP), lysine vasopressin (LVP), triglycil-lysine vasopressin, octopressin, ornipressin, and desmopressin (DDAVP), for a sufficient time to prevent organ failure.
4. The method of claim 3 wherein the septic shock is less severe septic shock.
- 15 5. The method of claim 3 wherein the organ failure is renal failure.
6. The method of claim 1 or claim 3 wherein the drug is selected from the group consisting of vasopressin, (AVP), (LVP), triglycil-lysine vasopressin, octopressin, ornipressin, and (DDAVP), is administered in a dose corresponding to 0.01 to 0.03 U of vasopressin per minute.
- 20 7. A kit for use in treating a subject having less severe septic shock, comprising vasopressin or a vasopressin-receptor agonist, and instructions for use.
8. The kit of claim 7, wherein the instructions comprise dosing information, or instructions for selecting a subject having less severe septic shock.
9. A method of determining the eligibility of a subject for treatment of septic shock
25 with a drug selected from the group consisting of vasopressin, arginine vasopressin (AVP), lysine vasopressin (LVP), triglycil-lysine vasopressin, octopressin, ornipressin, and desmopressin (DDAVP), the method comprising:

- i. determining the subject's requirement for vasopressor support;
- ii. determining the subject's requirement for one or less vasopressors at baseline;
- iii. determining the subject's baseline lactate levels;
- iv. determining the subject's APACHE II score at baseline; and
- v. determining the subject's signs of renal dysfunction at baseline

wherein the subject is eligible for treatment of septic shock with a drug selected from the group consisting of vasopressin, (AVP), (LVP), triglycil-lysine vasopressin, octopressin, ornipressin, and (DDAVP) if the subject has one or more of:

- vi. a requirement for 14 microgram norepinephrine /min) or less;.
- vii. a requirement for one or less vasopressors at baseline;
- viii. a baseline lactate level ≤ 1.4 mmol/L;
- ix. an APACHE II score at baseline ≤ 25 ; or
- x. an absence of signs of renal dysfunction at baseline.

10. Use of a drug selected from the group consisting of vasopressin, (AVP), (LVP), triglycil-lysine vasopressin, octopressin, ornipressin, and (DDAVP) as a catecholeamine-sparing agent for the treatment of less severe septic shock, wherein the catecholeamine is norepinephrine, and administered at a dose of less than 14 ug/minute.

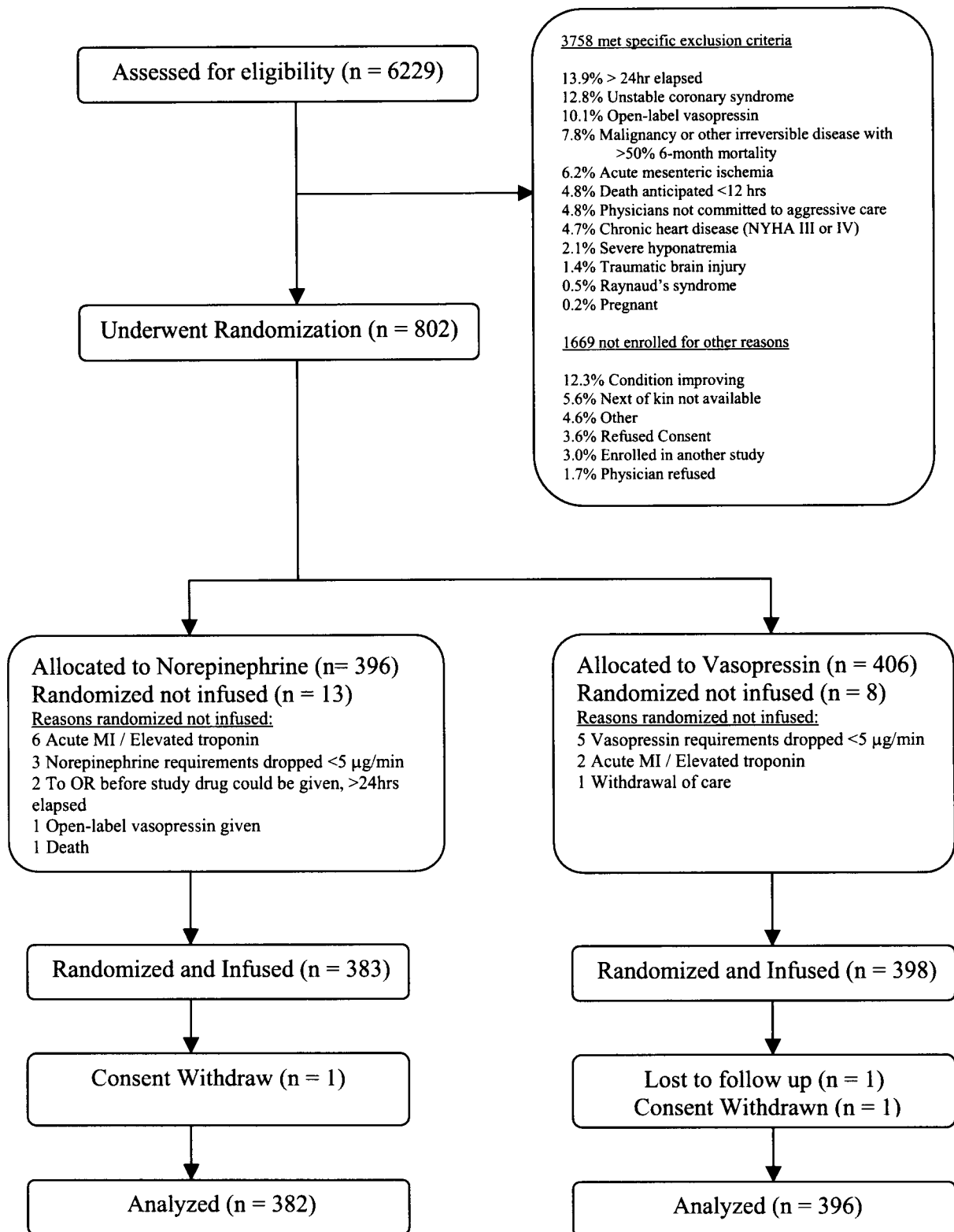


FIG. 1

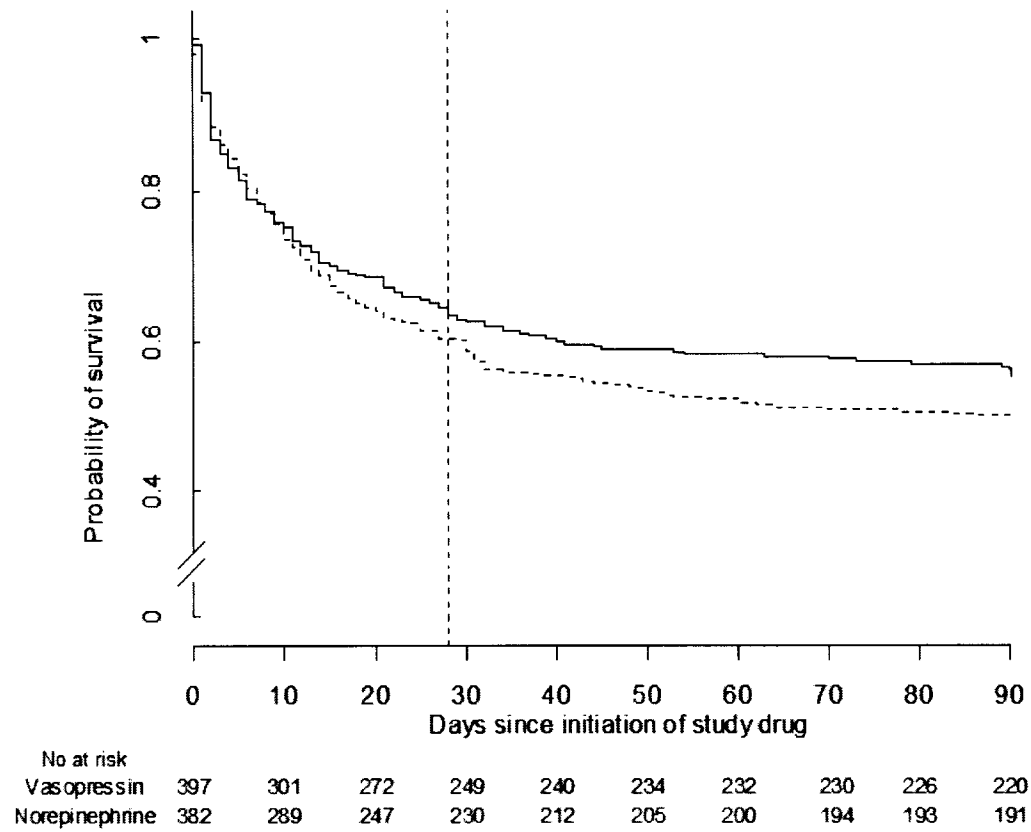


FIG. 2

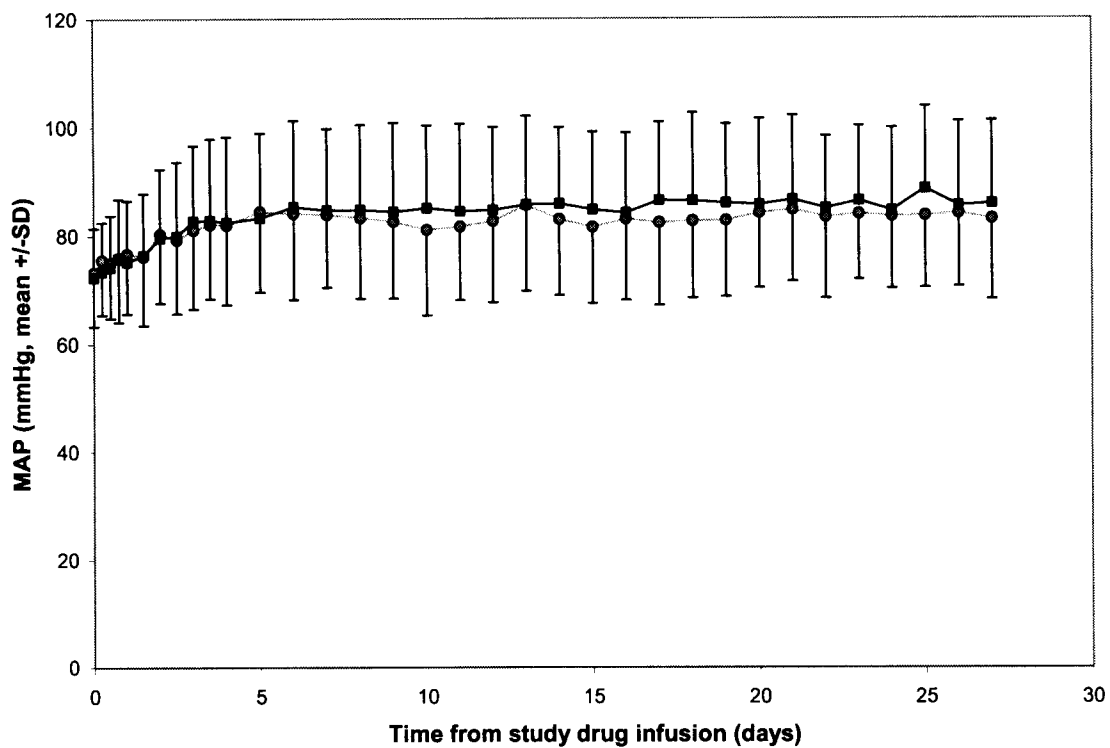


FIG. 3A

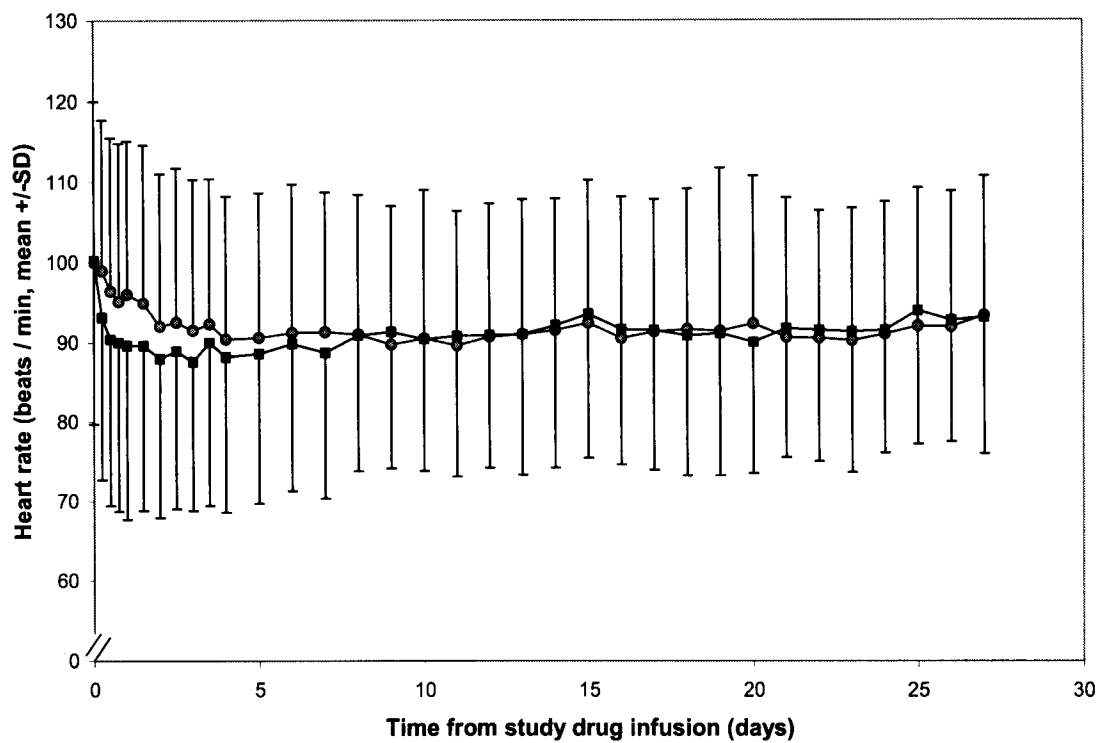


FIG. 3B

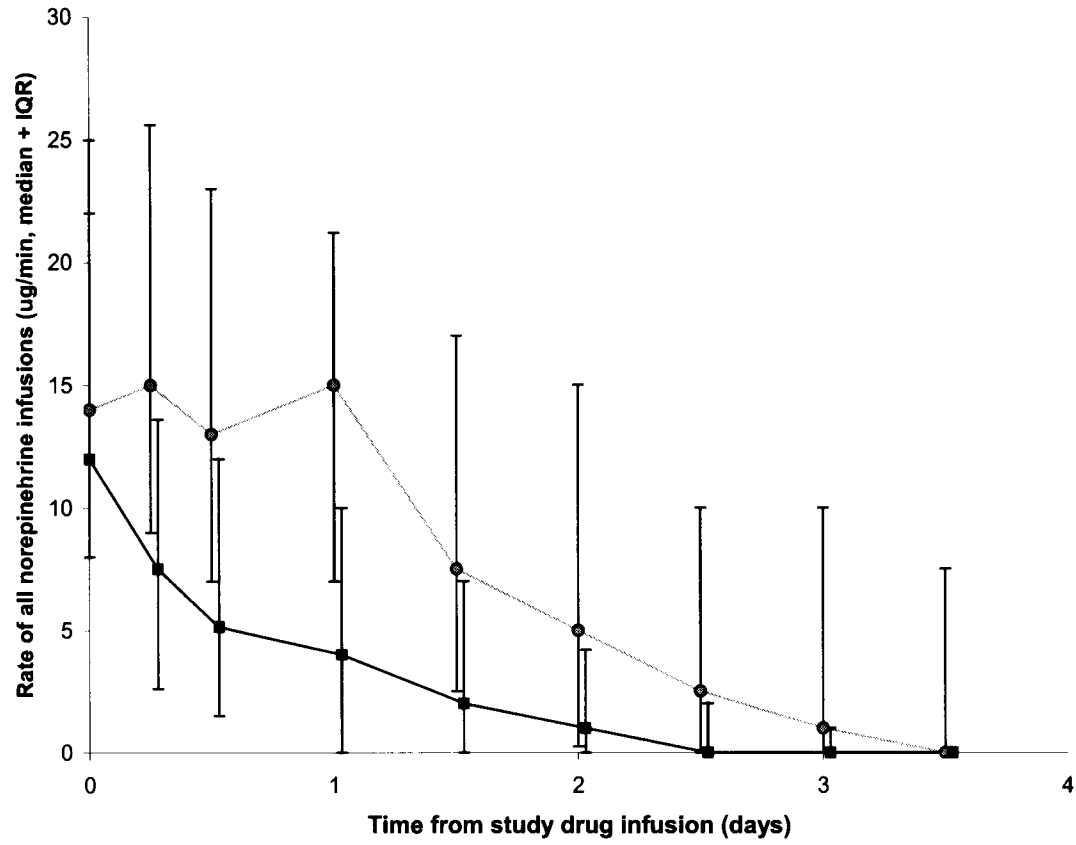


FIG. 4

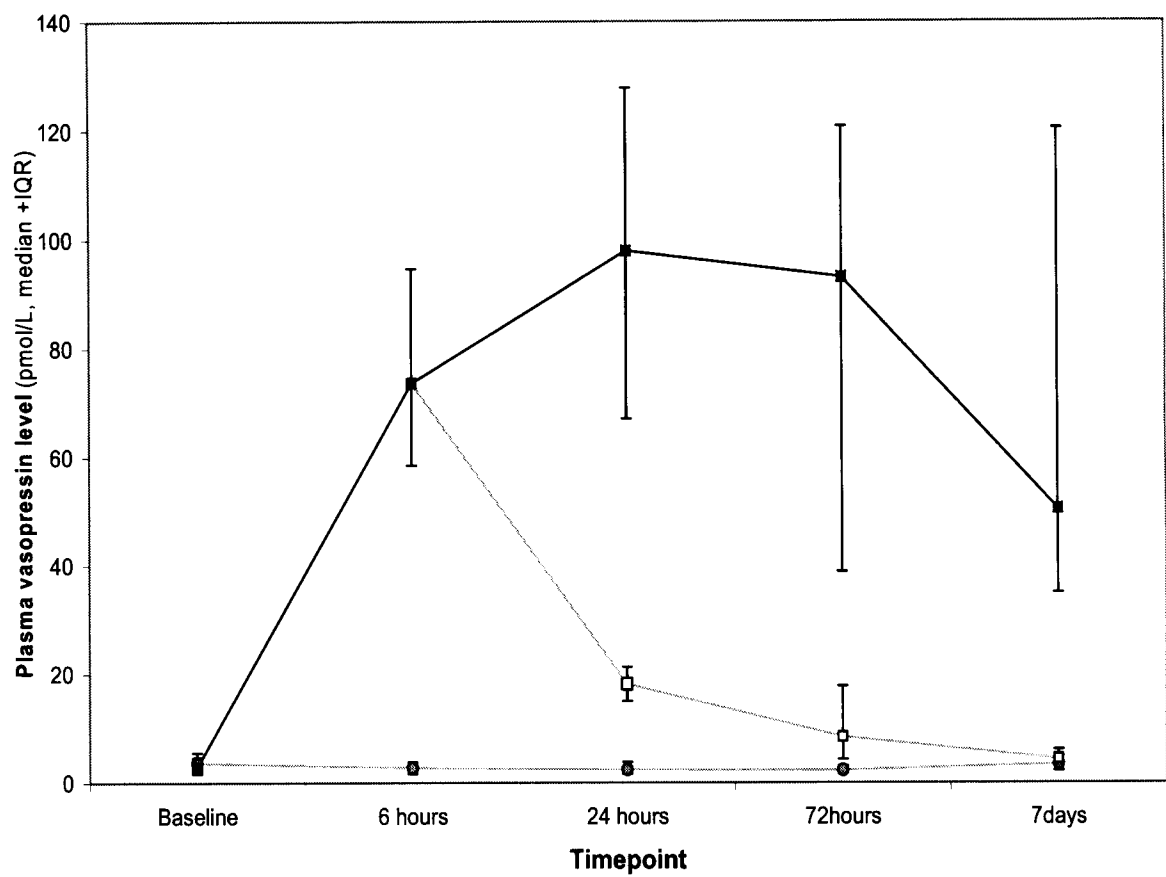


FIG. 5

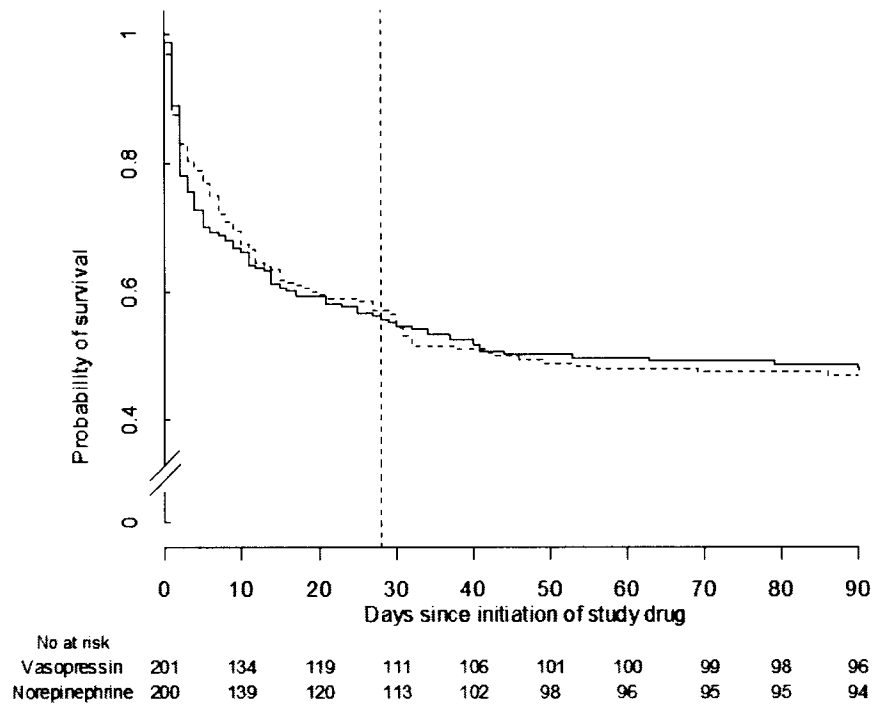


FIG. 6A

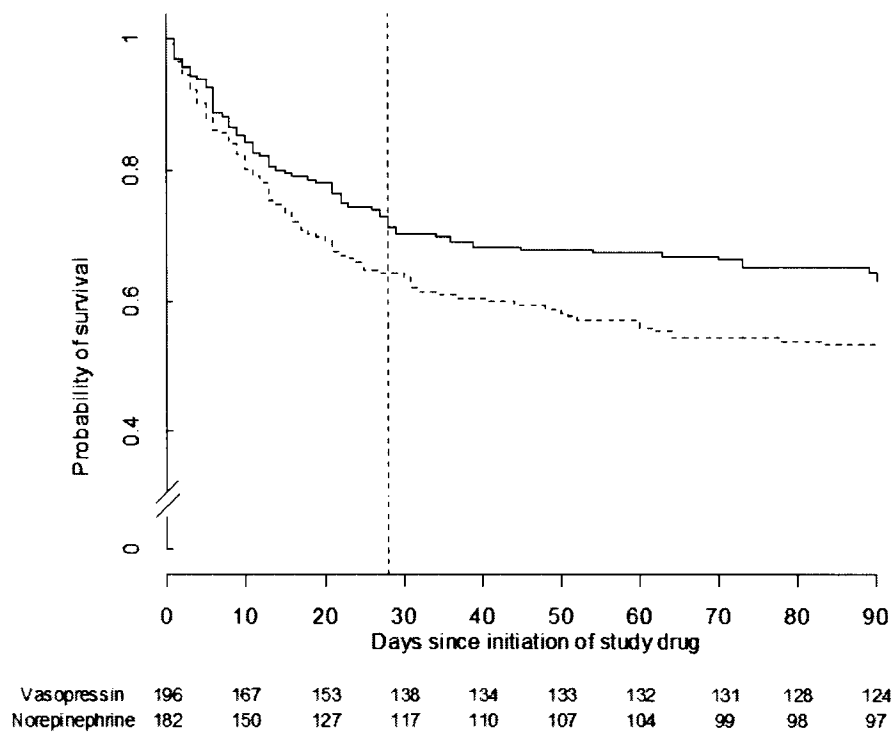


FIG. 6B

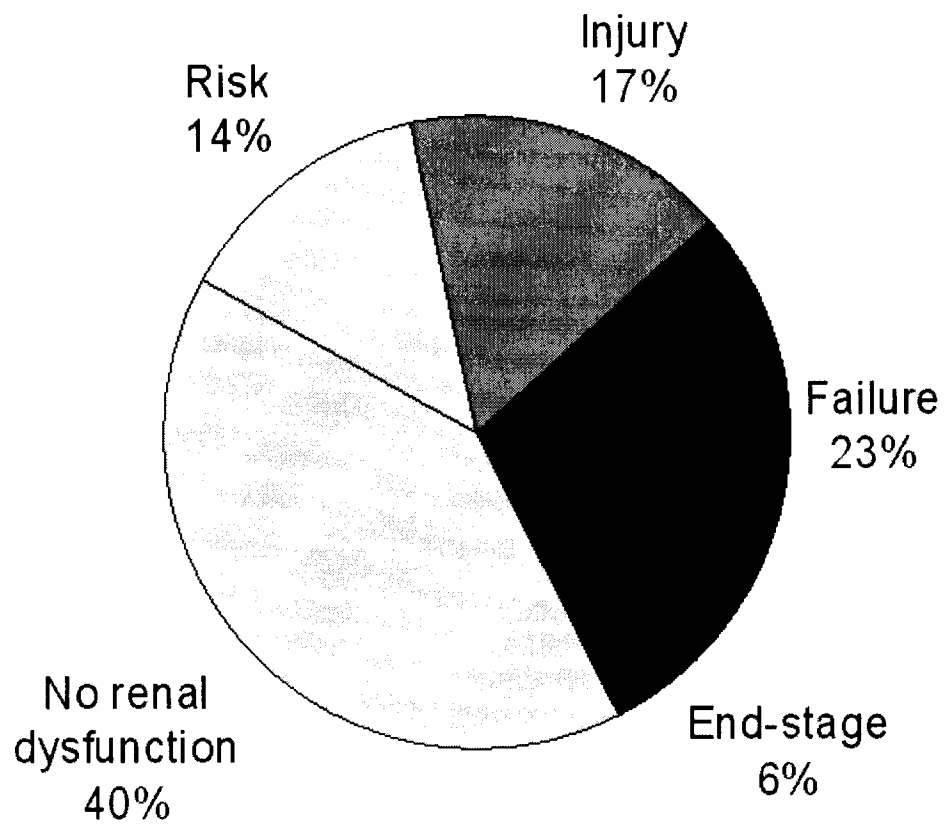


FIG. 7

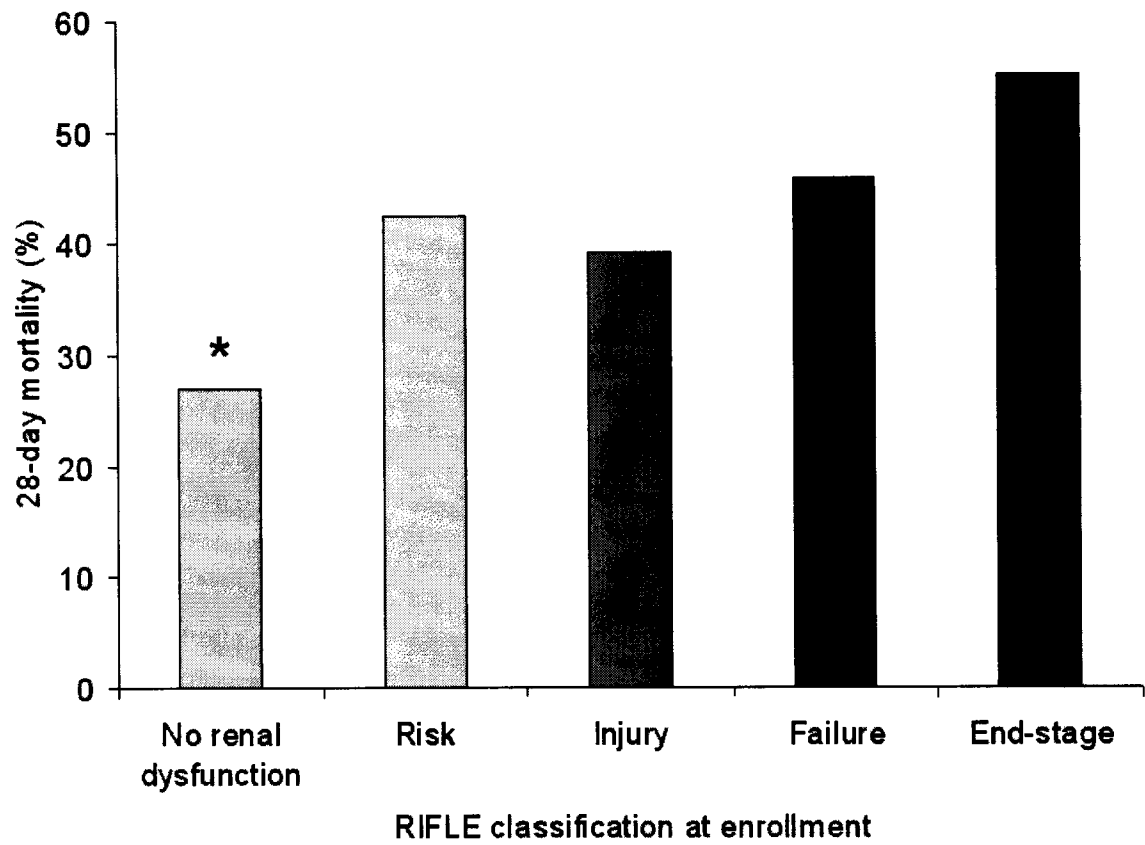


FIG. 8

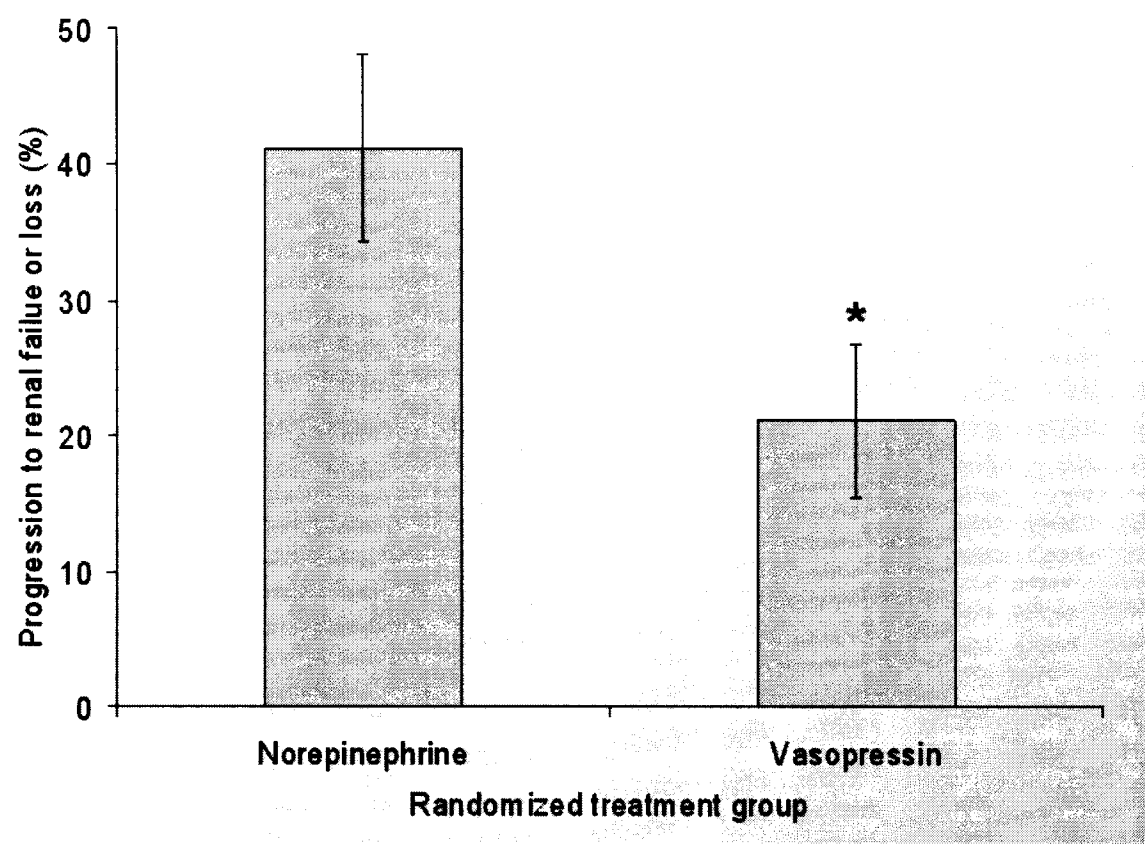


FIG. 9

INTERNATIONAL SEARCH REPORT

International application No.
PCT/CA2008/001346

A. CLASSIFICATION OF SUBJECT MATTER IPC: A61K 38/22 (2006.01) , A61K 31/57 (2006.01) , A61P 13/12 (2006.01) According to International Patent Classification (IPC) or to both national classification and IPC		
B. FIELDS SEARCHED Minimum documentation searched (classification system followed by classification symbols) IPC: A61K 38/22 (2006.01) , A61K 31/57 (2006.01) , A61P 13/12 (2006.01) Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched Electronic database(s) consulted during the international search (name of database(s) and, where practicable, search terms used) Canadian Patent Database, Pubmed, Delphion, Scopus: Keywords: vasopressin, septic shock, low-dose vasopressin, norepinephrine, Arginine vasopressin, AVP, coceptin, APACHE		
C. DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	Patel BM et al. "Beneficial effects of short-term vasopressin infusion during severe septic shock" Anesthesiology, 2002; 96(3): 576-582 whole document	2-3 and 5-8
X	Hall LG et al. "Fixed-dose vasopressin compared with titrated dopamine and norepinephrine as initial vasopressor therapy for septic shock", Pharmacotherapy 2004 Aug;24(8):1002-1012 whole document	1-5, 7-8 and 10
X	Sun Q et al. "Low-dose Vasopressin in the treatment of Septic shock in sheep", American Journal of Respiratory and Critical Care Medicine, 2003;168:pp481-486	1-6 and 10
☒ Further documents are listed in the continuation of Box ☐ See patent family annex.		
* Special categories of cited documents : "A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier application or patent but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed	"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art "&" document member of the same patent family	
Date of the actual completion of the international search 30 September 2008 (30-09-2008)		Date of mailing of the international search report 16 October 2008 (16-10-2008)
Name and mailing address of the ISA/CA Canadian Intellectual Property Office Place du Portage I, C114 - 1st Floor, Box PCT 50 Victoria Street Gatineau, Quebec K1A 0C9 Facsimile No.: 001-819-953-2476		Authorized officer Nicole Harris 819- 997-4541

INTERNATIONAL SEARCH REPORTInternational application No.
PCT/CA2008/001346**Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of the first sheet)**

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons :

1. ☒ Claim Nos. : 1-6

because they relate to subject matter not required to be searched by this Authority, namely :

Claims 1-6 are directed to a method for treatment of the human or animal body by surgery or therapy which the International Search Authority is not required to search under Rule 39.1(iv) of the PCT. However, this Authority has carried out a search based on the alleged effect or purpose/use of the product defined in claims 1-6.

2. ☐ Claim Nos. :

because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically :

3. ☐ Claim Nos. :

because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows :

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claim Nos. :

4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claim Nos. :

Remark on Protest ☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.

☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.

☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/CA2008/001346

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant	Relevant to claim No.
A	Knaus WA et al. APACHE II: A Severity of Disease Classification System, Society of Critical Care Medicine, 1985 Oct;13(10):818-29.	1-10
P, X	Hiraoka E et al. " Successful Treatment of Refractory Septic Shock with Low Dose Vasopressin" Respiration and Circulation, 2007;55(10):1165-1168	1-8 and 10
P, X	Russell JA et al. "Vasopressor Versus Norepinephrine Infusion in Patients with Septic Shock", New England Journal of Medicine, 2008 Feb; 358(9):877-887	1-10
P, X	Leone M & Martin C. "Vasopressin Use in Septic Shock:an Update", Current Opinion in Anaesthesiology, 2008 Apr;21(2):141-147	1-10
P, X	Holmes CL and Walley KR "Arginine Vasopressin in the Treatment of Vasodilatory Septic Shock". Best Practice and Research: Clinical Anaesthesiology, 2008 Jun;22(2):275-286	1-8 and 10
P, X	Ertmer C et al. "Current Place of Vasopressin Analogues in the Treatment of Septic Shock". Current Infectious Disease Reports, 2008 Sept;10(5):362-367	1-8 and 10
A	Dellinger RD et al. "Surviving Sepsis Campaign Guidelines for Management of Severe Sepsis and Septic Shock", Critical Care Medicine, 2004; 32(3), 858-873 whole document	1-10