

Nitric Oxide in Septic Shock: Directions for Future Therapy?

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Abstract

Septic shock is a major cause of death among patients in intensive care units. It has a mortality rate of 20% to 80%. The clinical syndrome of septic shock is characterised by hypotension, hyporesponsiveness to vasoconstrictors and volume depletion which will then lead to multiorgan dysfunction and death. Except for surgical and supportive care, no specific therapy is known. Recently interest has been focused on the role of nitric oxide (NO) in septic shock. Large amounts of NO released by the endothelium and vascular smooth muscle cells lead to profound vasodilation and hyporesponsiveness to vasoconstrictors. The cytotoxic effect of NO could also cause tissue injury and organ failure. Inhibition of NO synthase, the enzyme responsible for NO production, has been proposed as a new therapy for septic shock. However, experimental reports have provided conflicting results, demonstrating both beneficial and detrimental effects. A brief review of the role of NO in septic shock and the possible use of NO synthase inhibitors as potential therapeutic agents is presented here.

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Introduction

In 1980, Furchgott and Zawadzki¹ demonstrated that the relaxation of isolated arteries to acetylcholine required the presence of endothelial cells. This response was mediated by a labile humoral substance termed endothelium-derived relaxing factor (EDRF). Based on their very similar pharmacological profile, Furchgott² and Ignarro et al³ proposed that nitric oxide (NO) was responsible for the biological activity of EDRF. This triggered a massive interest in this small, freely diffusible, and potentially highly reactive molecule. The exponential increase in data relating to NO and its implications for physiology and pathology led to the naming of NO as the molecule of the year in 1992.⁴

The widespread involvement of NO in the physiological regulation of cellular function suggests that disturbance in its generation, release or action can lead to the pathogenesis of a wide number of disease states.⁵ Among these, overproduction of NO as implicated in pathophysiological states such as septic shock has been a major subject of interest. A large number of experimental studies and preliminary clinical trials have been carried out to study whether the therapy of septic shock is possible by the inhibition of NO production.

Biology of Nitric Oxide

NO, a simple and unique molecule, more than any other biological second messenger discovered to date, manifests a new paradigm for the way a molecule can regulate the biological environment in which it is produced. NO regulation, signal transduction, and cytotoxicity are strictly dependent upon its chemical reaction with oxygen and metals rather than specific structural interactions with physiological targets.

NO is now considered to be an important molecule formed not only in the endothelial cells but also in many other cells having different functions.⁶ It is an intracellular second messenger as well as an intercellular messenger (neurotransmitter, paracrine substance, autocoid or hormone) that regulates neighbouring cells. Signals from circulating NO complexes may also be transmitted to distant cellular targets.⁷ The synthesis of NO by the vascular endothelium serves to control vascular tone and to maintain a nonthrombogenic surface through vasodilation and inhibition of platelet aggregation. In the central nervous system NO is a neurotransmitter that underpins several functions, including the formation of memory. NO, derived from leukocytes and macrophages is considered to play a key role

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in cytotoxicity against microorganisms and tumour cells.⁸

The formation of NO is catalysed by a group of isozymes termed nitric oxide synthase (NOS).⁹ NO is produced from L-arginine by NOS with the concomitant production of L-citrulline. There are three distinct mammalian NOS. All three NOS isozymes have been purified, characterised, and cloned. There are many ways of naming the isoforms, the most straightforward way is to name them according to the order of their purification and the isolation of their cDNAs. The NOS isozymes are NOS type I (neuronal NOS or nNOS), NOS type II (inducible NOS or iNOS) and NOS type III (endothelial NOS or eNOS). Both eNOS and nNOS are constitutively expressed whereas the expression of iNOS requires induction by cytokines or other stimuli such as endotoxin. The homology among the three isoforms is around 50% to 55% at the amino acid level. The constitutive isoforms, eNOS and nNOS, are expressed by certain types of cells. However, when stimulated, most cells seem to be capable of expressing iNOS.

Although the three isoforms (eNOS, nNOS and iNOS) have different molecular weights and variable cofactor requirements, all of their activities require nicotinamide adenine dinucleotide phosphate (reduced form) [NADPH], thus showing similarity with cytochrome P₄₅₀ reductase. All forms of NOS contain prosthetic groups such as flavin-adenine dinucleotide (FAD), flavin mononucleotide (FMN), tetrahydrobiopterin (BH₄), and a heme complex, iron protoporphyrin IX. All NOS isoforms

activity are dependent on calmodulin, except that in the inducible isoform, calmodulin is already present in a tightly bound form. All of the isoforms can be inhibited to a variable degree with N^G substituted L-arginine analogs such as nitro-arginine and methyl-arginine.¹⁰ The similarities and differences in the isoforms of NOS are listed in Table I.

Differing from other known secondary messenger molecules, NO does not act on a receptor in the cell membrane but instead has a series of intracellular targets. The best known target of NO is the heme-containing soluble guanylate cyclase.¹¹ NO activates guanylate cyclase by binding to iron in the heme which is at the active site of the enzyme, altering the enzyme's conformation and thus activating the enzyme, resulting in increased intracellular cGMP levels. An increase in cGMP in target cells will lead to several physiological responses such as vasodilation, inhibition of platelet aggregation, inhibition of the proliferation of vascular smooth muscle cells, and leukocyte modulation. Many of these responses are mediated by the activation of a protein serine/threonine kinase, which is the cGMP dependent protein kinase.¹² Subsequent cGMP-dependent phosphorylation of proteins results in intracellular Ca⁺⁺ sequestration or removal which will then lead to various physiological responses.¹³

Another heme-containing enzyme whose activity is influenced by NO is cyclooxygenase, which is the rate-limiting enzyme in the biosynthesis of prostaglandins, thromboxane A, and prostacyclins. The effects of NO on

TABLE I: SIMILARITIES AND DIFFERENCES IN THE ISOFORMS OF NITRIC OXIDE SYNTHASE

	Type I (nNOS)	Type II (iNOS)	Type III (eNOS)
Cell type	Brain, non-adrenergic non-cholinergic (NANC) nerves, etc	Macrophage, leukocytes, vascular smooth muscle cells, etc	Endothelium only
Molecular weight	160 kDa	130 kDa	135 kDa
Calcium	Dependent	Independent	Dependent
Inducibility	Constitutive	Inducible	Constitutive
Solubility	Soluble	Soluble	Particulate
Activators	Excitatory amino acid such as glutamate and NMDA	Endotoxin, cytokines	Acetylcholine, ATP, ADP, bradykinin, thrombin, shear stress
Release	Transient release of minute quantities (picomoles) of NO	Sustained release of large amount (nanomoles) of NO	Transient release of minute quantities (picomoles) of NO
Main physiological function	Neurotransmission and memory formation	Cytotoxicity effects towards tumour cells and invading microorganisms after immunological stimulation	Maintenance of a basal vasodilator tone, regulation of blood flow, and inhibition of adhesion and activation of circulating blood cells
Location of human chromosome	12	17	7

NOS: nitric oxide synthase; nNOS: neuronal NOS; iNOS: inducible NOS; eNOS: endothelial NOS; NMDA: N-methyl-D-aspartate; ATP: adenosine triphosphate; ADP: adenosine diphosphate

the cyclooxygenase are not uniform. Activation of cyclooxygenase by NO has been shown to increase prostaglandin production,¹⁴ while other studies demonstrated that NO inhibits the activity of cyclooxygenase.¹⁵ Such an effect would affect the production of prostaglandins within the vascular smooth muscle.

NO can also bind to non-heme iron, particularly to the iron in the iron-sulfur centres in numerous enzymes, thus altering their biological activity.¹⁶ These enzymes include those in the respiratory cycle, the tricarboxylic acid (TCA) cycle and the DNA synthesis pathway. Interaction of NO with these enzymes provides the cytotoxic and cytostatic effects of NO.¹⁷ NO has also been reported to enhance ADP-ribosylation of proteins and poly-(adenosine 5'-diphosphoribose) synthetase (PARS) is one of the targets for NO-mediated ADP-ribosylation.¹⁸ Pronounced activation of PARS by NO in the vasculature during pathological condition such as septic shock can rapidly deplete the intracellular concentration of its substrate, NAD⁺, slowing the rate of glycolysis, electron transport and ATP formation in smooth muscle cells, and may therefore lead to a loss of both contractile and relaxant ability of various blood vessels.

One of the more recently described mechanism of NO signalling, at least in terms of its pathophysiological effects on cells, is the formation of peroxynitrite.¹⁹ NO reacts with superoxide (O₂⁻) generated during cellular oxidative injuries to form the free radical peroxynitrite. Peroxynitrite is capable of inducing a number of oxidation and cytotoxic processes, including inhibition of the activity of membrane sodium-potassium antiport²⁰ and inhibition of calcium pumps in the sarcoplasmic membrane.²¹ It is conceivable that such effects would also impair the contractility of the smooth muscle cells.

NO itself has very short half-life, therefore generation of NO is normally measured by indirect method. Measurement of the end products of NO oxidation, nitrites and nitrates, by colorimetric Greiss reaction is one of the most common way.²² Formation of radioactive-labelled citrulline from labelled L-arginine is perhaps the most widely used method for the assay of NOS activity.²³ The stimulation of the soluble guanylate cyclase has also been used as the basis of assays for NOS.²⁴ To study the mRNA expression of various NOS isoforms, techniques such as Northern-blot²⁵ and quantitative reverse transcription-polymerase chain reaction (RT-PCR)²⁶ have been used.

Nitric Oxide and Septic Shock

During episodes of septic shock, profound physiological changes are often observed, including the development of systemic vasodilation, the production of fever, and the activation of the immune system. These changes are induced by a wide range of cellular mediators, such as endotoxin and various cytokines. There is increasing

evidence to indicate that NO plays an important role as one of the mediators of septic shock, in addition to its described physiological functions. However, the precise role of NO in septic shock remains unclear.²⁷

Three lines of evidence indicate that NO plays a critical role in septic shock:

- 1) Ca⁺⁺ independent NOS expression is widespread in endotoxaemia. Macrophages, hepatocytes, endothelial cells, smooth muscle cells, cardiac myocytes, and neuroglial cells all express Ca⁺⁺ independent NOS in experimental models of endotoxaemia.²⁸ By using RT-PCR, we have also shown that endotoxin-treated mice demonstrated a time-dependent increase in the expression of liver iNOS mRNA expression.²⁶
- 2) NO concentrations are raised in patients with sepsis and its presence correlates with the fall in systemic vascular resistance characteristic of septic shock.²⁹
- 3) Inhibiting NO action in several experimental animal models of shock prevents the development of both shock and tissue injury and confers a significant survival advantage.³⁰

In general, NO acts as an important autocrine homeostatic modulator for the vascular system. Therefore, a natural consequence of the impairment of the endothelial vascular NO production is disease or predisposition to disease.⁵ Smooth muscle cells and other non-endothelial cells in the vascular wall are not normally capable of synthesising NO. However, in sepsis or septic shock, lipopolysaccharide (LPS) and cytokines lead to an excessive induction of Ca⁺⁺ independent NOS in cells of the vascular wall (probably mostly smooth muscle cells). This enzyme then produces a large amount of NO which activates soluble guanylate cyclase in an autocrine fashion and leads to various symptoms of septic shock (Fig. 1). Excessive production of NO by the endothelium and vascular smooth muscle cells contributes to the vasodilation and hyporesponsiveness to vasoconstrictors observed in animal models and patients of septic shock.³¹ In skin and other tissues NO can cause local vasodilation and alter microvascular permeability leading to oedema and erythema. Alterations in vascular perfusion rates and tissue oedema or the well-documented cytotoxic effect of NO will therefore lead to tissue damage and organ failure. NO may also modulate protein synthesis in organs such as the liver and therefore result in the end-stage organ failure seen in prolonged sepsis.³²

Although widespread Ca⁺⁺ independent NOS expression can result in vascular collapse and haemodynamic shock,³³ it appears that the vasodilation induced by NO in response to sepsis may be part of the adaptive response aimed at counteracting circulating vasoconstrictors and maximising tissue perfusion.²⁷ Mediators released during sepsis, such as thromboxane and endothelin-1, cause vasoconstriction, and NO production provides an important coun-

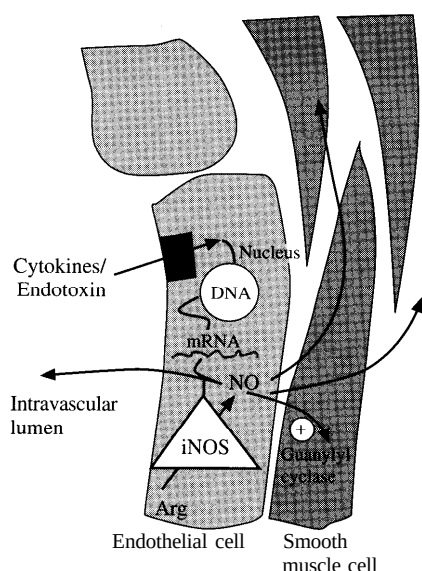


Fig. 1. Overproduction of NO by vascular cells in septic shock. During septic shock, cytokines or endotoxin in the circulation might induce iNOS in vascular endothelium to synthesise pathological level of NO. NO diffuses to neighbouring cells and lead to various symptoms of septic shock.

ter-regulatory mechanism. Furthermore, NO is able to block platelet aggregation and leukocyte adhesion which will help in the prevention of microvascular stasis and thrombosis.³⁴

Clinical Characteristics of Septic Shock

Septic shock is one of the most frequent and serious problems confronting clinicians.³⁵ It is the most frequent cause of mortality in modern clinics, especially in intensive care units. The mortality rate in patients with septic shock ranges from 20% to 80%. The clinical syndrome of septic shock is characterised by peripheral arteriolar vasodilation which leads to a hyperdynamic state with low systemic vascular resistance, high cardiac output, hypotension, hyporeactivity to vasoconstrictor agents, inadequate tissue perfusion, vascular damage, and disseminated intravascular coagulation leading to multiple organ dysfunction and death.³⁶

Septic shock is usually initiated by the infection of gram-negative bacteria as well as gram-positive organisms and fungi.³⁷ LPS, which is an integral part of the outer layer of the gram-negative bacteria cell wall, is considered the most important exogenous mediator of septic shock. Upon lysis of the bacteria, LPS is released into the surrounding medium as a complex with proteinaceous material, forming micelle-like solute known as endotoxin. Therefore septic shock is also termed as endotoxin shock or endotoxaemia. LPS activates various cell types as well as enzyme systems in the extracellular medium. This interaction results in the release of various categories of endogenous mediators of septic

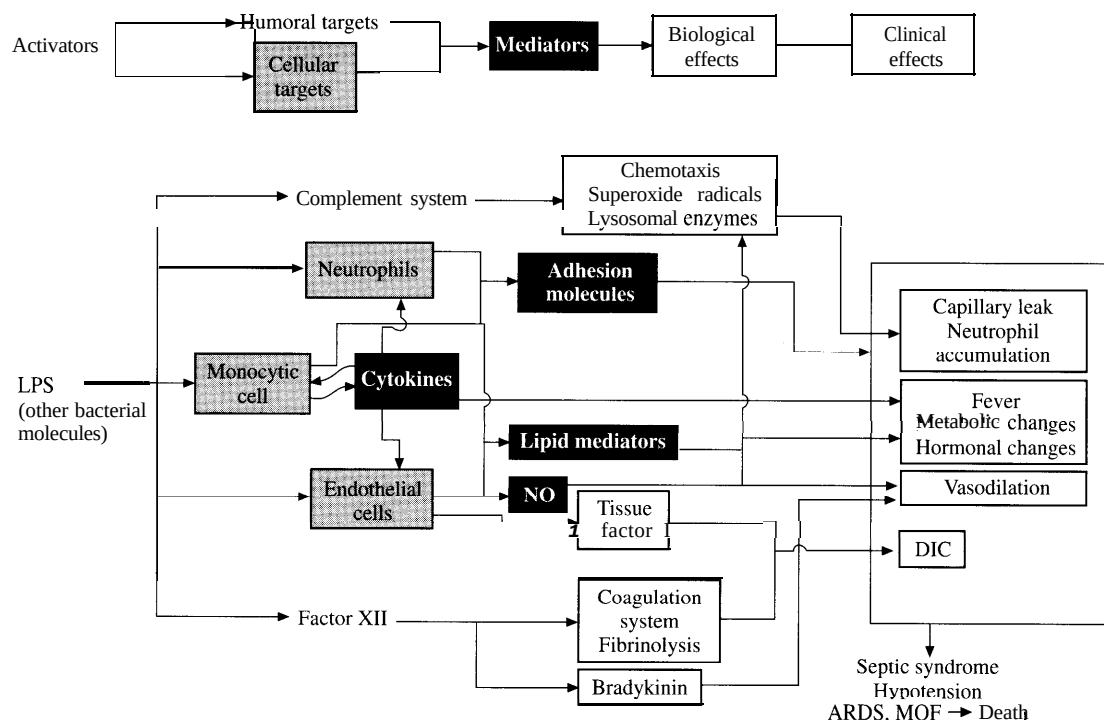
shock. LPS can also activate the complement system, the blood clotting cascade and the kallikrein-kinin system. Each of these systems will then contribute to the pathogenesis of sepsis.³⁸

Although the pathogenesis of septic shock has been a matter of intensive study for many years, it is not yet completely understood. The vascular and tissue alternations in shock have been recognized to be mainly due to the complex interaction of cells, cytokines, and humoral pathways. A large number of mediators and inflammatory cells interact with endothelial and vascular smooth-muscle cells to interfere with blood flow, ultimately leading to microvascular failure. Figure 2 shows a diagram of the possible pathogenesis of septic shock.

Activated complement is considered an important element in the pathogenesis of septic shock.³⁹ It augments permeability of endothelia, causes immune adherence of platelets, helps endotoxin to activate platelets, induces production of tissue factor by leukocytes and endothelial cells, induces cytokine production in monocytes and potentiates neutrophils in inflicting injury on endothelia. Another potential mediator of septic shock is the platelet activating factor (PAF). Its effects include induction of platelet aggregation as well as increased vascular permeability and hypotension.⁴⁰ Factor XII (Hageman factor) in the coagulation cascade is also known to play a central role in the pathogenesis of septic shock. Activation of these mediators will lead to disseminated intravascular coagulation.⁴¹ Cyclooxygenase products such as thromboxane A₂ (a potent vasoconstrictor)⁴² and prostacyclin (potent vasodilator)⁴³ are also increased in endotoxin-treated animals. Interaction between these vasoconstrictors and vasodilator influences has been presumed to be the reason for the complex cardiovascular profile in endotoxin shock.

Monocytes/macrophages play a pivotal role in the mediation of pathogenesis of septic shock. Under the stimulation of LPS, monocytes release a large array of cytokines. Activation of the cytokine network occupies a key position in the chain of events that lead to shock, in that their production and action follow early activation of the plasma system (complement, intrinsic blood clotting pathway) and precede release of the other tissue-derived mediators (eicosanoids, PAFs). Cytokines such as tumour necrosis factor- α (TNF- α),⁴⁴ interleukin-1 (IL-1),⁴⁵ γ -interferon (γ -INF),⁴⁶ interleukin-6 (IL-6)⁴⁷ and interleukin-10 (IL-10)⁴⁸ have been considered central mediators of septic shock. Neutrophils that are activated either directly by LPS or indirectly through the actions of cytokines, may damage tissue by releasing oxygen metabolites and lysosomal enzyme, or may cause microemboli after aggregation.⁴⁹

More recently, a new concept indicates that endotoxin, alone or in combination with cytokines, induces the



LPS: lipopolysaccharide; LBP: LPS-binding protein; DIC: disseminated systemic coagulation; ARDS: adult respiratory distress syndrome; MOF: multiple organ failure. Fig. 2. Interaction of humoral factors, cytokines and the immune cells in the pathogenesis of septic shock (modified from Glauser et al⁷⁰).

expression of a large amount of iNOS in the vascular wall. Besides, hypotension during septic shock also reflect the increased synthesis of the potent vasodilator NO. Therefore, NO is also considered as an important mediator for the pathogenesis of septic shock.

Current Therapy for Septic Shock

The current treatment of sepsis such as with fluid replacement, antibiotics and artificial ventilation⁵⁰ is merely supportive and is not directed at the underlying mechanisms of shock. Despite full intensive therapy and conventional pharmacological treatments, the mortality rate from the sepsis syndrome is still high.

Except for surgical and supportive care, limited success has been reported through the use of antibodies against endotoxin. Since sepsis begins with the proliferation of microorganisms at a nidus of infection, antimicrobial therapy is mandatory. In multicentre clinical trials of antibodies against endotoxin in patients, the mortality rate and incidence of organ failure were decreased only in a sub-group of the patients.⁵¹ The patients in this sub-group were those with blood cultures positive for gram-negative organisms and they were identified early in the course of infection. Thus, therapy with antibodies against endotoxin must be initiated at the early onset of the syndrome.

Future treatment of gram-negative sepsis may be aimed at inhibiting the host's inflammatory response to endo-

toxin. This would involve preventing neutrophil and macrophage activation, antagonizing cytokines and lipid mediators, or using anti-adhesion antibodies to prevent neutrophil-mediated injury. Other innovative therapies under development involve the use of interleukin antagonists.^{52,53} Despite these efforts, no improvement in survival rates has been consistently demonstrated.

In spite of the extensive research conducted, researchers have not been able to produce a magic bullet for antishock therapy. In septic shock, once an inflammatory stimulus is present, a cascade of events will take place. Therefore the likelihood that a single therapeutic agent directed at a mediator produced early in the inflammatory cascade will alter the outcome for the patients at all levels of the pathophysiologic response is actually very low. However, for patients with continued, persistent endotoxaemia or cytokinaemia these approaches may still have a beneficial effect. For most patients, a therapeutic agent that targets at mediators that act at distal sites in the inflammatory cascade might provide a theoretical advantage. One of such targets is the mediator of vasodilation-NO.

Therapeutic Potential of Inhibiting Nitric Oxide Production

NO donors have long been used, mainly as treatment for angina and other disease states. By utilising specific features of the metabolism of NO-carrying agents,

NO donors such as sodium nitroprusside, glyceryl trinitrate and molsidomine have been used as nitrovasodilators.⁵⁴

Experiments in dogs provided the first *in vivo* evidence for the role of NO in shock.⁵⁵ These initial observations were then followed by a series of reports demonstrating the effects of NOS inhibitors in various animal models of shock. Contradictory results have been observed in studies with septic shock animal models on the inhibition of NO. One example was a study with the prototypical NOS inhibitor, N^G-monomethyl-L-arginine (L-NMMA).¹⁰ While low doses of L-NMMA will prevent endotoxin shock, high doses will exacerbate the condition. The benefits of inhibited NOS are a sustained increase of systemic blood pressure while the adverse effects are decreased cardiac output and raised pulmonary vascular resistance.⁵⁶ Similarly, clinical trials for septic shock have been conducted with the same prototypical NOS inhibitor, L-NMMA. Early clinical results suggested L-NMMA did indeed reverse the hypotension; however, it is also decreased cardiac output and heart rate which adversely affected peripheral tissue perfusion.⁵⁷

Based on these experimental results, researchers hypothesised that the effects of agents that block NO synthesis from L-arginine during endotoxaemia depend critically on the differences in the degree of inhibition of the constitutive (cNOS) and inducible NOS (iNOS) enzymes. High concentrations of inhibitors that will inhibit both the inducible and constitutive NOS may remove all the NO-dependent vasodilator tone. As Nava and colleagues⁵⁶ showed, this effect, coupled with the presence of circulating vasoconstrictors, can lead to a dramatic fall in blood pressure, end-organ damage, and death. However, a lower dosage of the inhibitor or a higher dosage of the inhibitor together with a NO-donor may still provide a therapeutic avenue to reverse the vasodilation that is the main adverse feature of the septic syndrome. Other controversial data obtained from different research group might be due to other factors, such as variation in the sensitivity of different species to the initiators of shock, the choice and dose of NOS inhibitors used and the time of intervention with the therapeutic agent.

It is likely that induction of NO synthesis is the final common pathway for vasodilation in endotoxin shock. Unlike agents used to neutralise endotoxin and individual cytokines, various combinations of which may be needed for different groups of patients, NOS inhibitors offer a single therapeutic approach for the management of vasodilation and hypotension associated with the septic shock syndrome. Nevertheless, the effectiveness of NOS inhibition in the treatment depends critically on the degree of inhibition of the iNOS and cNOS isozymes. For example, L-arginine-based inhibitors of NOS such

as L-NMMA are usually not selective for iNOS. Therefore inhibition of NOS by these inhibitors *in vivo* may have potential disadvantages. Recently, some non-amino acid compounds such as guanidines,⁵⁸ S-alkylisothioureas,⁵⁹ amidines⁶⁰ and mercapto-alkyl-guanidines⁶¹ have been reported to potently inhibit NOS, with some of them having a certain degree of selectivity for iNOS. These compounds might become useful in septic shock treatment, but so far only limited data are available on their biologic activity *in vivo*.

Many other approaches have been suggested for the indirect inhibition of iNOS. Since NOS is a tetrahydrobiopterin-dependent enzyme, inhibition of tetrahydrobiopterin biosynthesis could limit the activity of NOS.⁶² This approach may be more specific for iNOS because cNOS has a tightly bound cofactor. Arginine depletion might be another approach to limit NO synthesis. Studies with arginase indicate that basal NO synthesis requires only small amounts of arginine, whereas pathologic overproduction of NO requires a large amount of substrate.⁶³ Therefore, depletion of arginine might limit the overproduction of NO in septic shock. Besides, administration of NO scavengers such as haemoglobin⁶⁴ and guanylate cyclase inhibitor such as methylene blue^{65,66} might be another possible approach in septic shock therapy.

Besides an obvious drawback in the lack of NOS isoform selectivity with the present NOS inhibitors, there are several other problems associated with clinical trials of NO inhibition in sepsis. Sepsis is a complex disorder with various stages of vascular responses and organ failure. Some evidence suggest that early cytokine mediators actually downregulate cNOS and impair vascular regulation quite early in sepsis.⁶⁷ At a later stage iNOS may become more important. Hence, NO-based therapy may be appropriate earlier on in sepsis to maintain vascular perfusion and prevent leukocyte adhesion⁶⁸ while selective iNOS inhibition may be desirable as sepsis progresses.

Before a specific iNOS inhibitor is discovered, the efficacy of using NOS inhibitors for the treatment of septic shock is still doubtful. However, in comparison with catecholamines, NOS inhibitors have theoretical and practical advantages. As a mechanism-based inhibitor of an overproduced endogenous vasodilator, vessel tone can be restored without causing non-specific constriction. In addition to that, some of the adverse cardiac effects of NOS inhibitors can be rectified by means of a combined administration of a NOS inhibitor with a positive inotropic agent such as dobutamine.⁶⁹ In this case, severe cardiac depression can be reversed and blood pressure will be restored. Therefore, it is possible that restoration of vascular tone by inhibition of pathologic NO production may provide the first step in multiagent therapy for septic shock.

Conclusion

NO is a low molecular weight, membrane permeable gas that is synthesised from L-arginine by the enzyme NOS. There are mainly two distinct types of NOS which are designated as cNOS and iNOS. cNOS is constitutively expressed and generate small amounts of NO which are responsible for neurotransmission, regulation of vascular tone and inhibition of platelet aggregations and leukocytes adhesion. In contrast, iNOS is expressed only after cell activation by immunological stimuli such as bacteria endotoxins, exotoxin or cytokines. However, once iNOS is expressed, it will cause prolonged synthesis of NO which is cytostatic or cytotoxic for tumour cells and certain pathogens. Considerable interest in the overproduction of NO in patients with septic shock has arisen since NO has been shown to contribute to the severe hypotension as well as direct tissue injury seen in these patients.

At present, the clinical management of septic shock is purely supportive and no proper therapy is available. Recent reports showing that NOS inhibitors might provide a possible treatment for septic shock have raised some hopes. However, the widespread biological actions of NO indicate that non-specific NOS inhibitors or non-tissue selective NO donors may have potential harmful side effects. The clinical use of these agents therefore will require extreme care and consideration. More experimental and clinical studies must be carried out to explore the role of NO in septic shock and delineate the potential for its inhibition in patients. Nevertheless, all the experimental studies and preliminary clinical data obtained so far strongly support the view that inhibition of NO synthesis will represent a novel and rational approach for the management of progressive vasodilation in septic shock patients.

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