

Role of lipids in sepsis

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Abstract

Activation of phospholipase A₂, excess formation of proinflammatory eicosanoids, decreased generation of lysophosphatidylcholine (LPC) and enhanced lev-

els of ceramide play a significant role in sepsis. This suggests that lipids participate in the pathogenesis of sepsis.

Keywords: Sepsis, ω -3 and ω -6 fatty acids, Lysophosphatidylcholine, prostaglandins, tumor necrosis factor, interleukins, free radicals, nitric oxide, High mobility group box-1, macrophage migration inhibitory factor, polyunsaturated fatty acids

Introduction

It is estimated that more than 750,000 cases of sepsis are diagnosed, and accounts for about 200,000 deaths per year in U.S.A. alone. The incidence of sepsis is increasing by 1.5% annually due to the ageing of the population in the U.S.A. in whom the incidence is increasing.

Sepsis is defined as systemic inflammatory response to infection and when hypotension and multiorgan dysfunction occurs, it results in septic shock. Noninfectious disorders such as trauma, pancreatitis, and major abdominal and cardiovascular surgery also result in sepsis and septic shock. Multiple organ dysfunction syndrome (MODS) is the most common cause of death among patients in non-coronary critical care units.

Bacterial toxins, inflammatory mediators secreted by neutrophils, macrophages, and T cells, endothelial injury, disturbed homeostasis, and microcirculatory failure contribute to the development of MODS. Sepsis impairs immune function by inducing defects in innate immunity and excessive lymphocyte apoptosis.

Immune System in Sepsis

Originally, it was thought that over stimulation of the immune system causes death from sepsis. Studies performed in animal models of sepsis used large doses of endotoxin

or bacteria that induced the release of high levels of tumor necrosis factor- α (TNF- α). This "cytokine storm" explains why anti-TNF- α antibodies improved survival. Similar cytokine storm is seen in patients with meningococemia [1]. But, several subsequent studies showed that elevated TNF- α levels are not common in meningococemia [2]. It is interesting to note that anti-TNF- α antibody enhanced mortality in experimental animals after cecal ligation and puncture or endotoxemia [3]. In a neutropenic model of sepsis, anti-TNF- α and anti-interleukin-1 (IL-1) antibodies enhanced fatality [4]. Furthermore, sepsis and tuberculosis infection are common in patients with rheumatoid arthritis who received anti-TNF antibodies or TNF antagonists [5]. This suggests that total blockade of pro-inflammatory cytokine actions is harmful.

Patients with sepsis can not clear infection(s), show reduced delayed hypersensitivity, and are susceptible to nosocomial infections [6]. In the initial stages of sepsis, there is an increase in inflammatory mediators and later as sepsis continues immunosuppression occurs. This explains why some respond to anti-TNF- α therapy, corticosteroids, and anti-inflammatory drugs whereas others fail to do so. Identification of the inflammatory and immunosuppressive phases of sepsis and instituting appropriate therapy may help to reduce mortality.

Inflammatory and immunosuppressive phases of sepsis

On exposure to bacteria, or release of endotoxin and/or exotoxin, innate immune system, endothelial cells and

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other cells are stimulated. This leads to the release of interleukin-1 (IL-1), IL-2, IL-6, IL-8, TNF- α , platelet activating factor (PAF), endorphins, eicosanoids, nitric oxide (NO), reactive oxygen species (ROS), macrophage migration inhibitory factor (MIF), chemokines, and increase in the expression of adhesion molecules. These molecules have actions on the cardiovascular system, kidneys, lungs, liver, central nervous system, and coagulation cascade that may result in renal failure, myocardial dysfunction, acute respiratory distress syndrome (ARDS), hepatic failure, and disseminated intravascular coagulation. These events ultimately lead to death in sepsis [7,8].

Abnormal innate immune response and excessive apoptosis of lymphocytes occur in sepsis [9,10] that impairs the response to pathogens. Lymphocyte apoptosis could be due to stress-induced release of glucocorticoids that causes immunosuppression and contributes to mortality in sepsis. Interferon- γ (IFN- γ) that activates macrophages is beneficial in sepsis [11] and inhibition of lymphocyte apoptosis improved survival in sepsis [12].

Enhanced release of free radicals and proteases by neutrophils induces tissue injury. Intrapulmonary sequestration of neutrophils and their inappropriate activation causes acute respiratory distress syndrome (ARDS) in sepsis. Granulocyte colony stimulating factor (G-CSF) that increases the number of neutrophils and augments their function, improved survival of patients with sepsis [13,14], although some other studies did not support this conclusion. These studies suggest that leukocytosis *per se* is not harmful and at times may be beneficial in sepsis. Furthermore, IFN- γ improved survival in a subgroup of patients with sepsis by restoring macrophage HLA-DR expression (which is suppressed in sepsis) and TNF- α production [9]. Thus strategies adopted to enhance the function of the innate and/or adaptive immune system may benefit patients with sepsis.

Cytokines, macrophage migration inhibitory factor (MIF), free radicals, and high-mobility group B1 (HMGB1) protein play a pivotal role in sepsis. Both MIF and HMGB1 participate in sepsis, and inhibition of their synthesis or action improved survival in experimental animals [15]. But their exact role in humans remains to be firmly established. Recent studies suggested that lipids play an important role in sepsis.

Essential fatty acids and inflammation

Essential fatty acids (EFAs), linoleic acid (LA, 18:2 ω -6) and alpha-linolenic acid (ALA 18:3 ω -3), form precursors to long-chain polyunsaturated fatty acids

(LCPUFAs) and eicosanoids. LCPUFAs such as gamma-linolenic acid (GLA, 18:3 ω -6), dihomo-GLA (DGLA, 20:3 ω -6), arachidonic acid (AA, 20:4 ω -6) are formed from LA and eicosapentaenoic acid (EPA, 20:5 ω -3) and docosahexaenoic acid (DHA, 22:6 ω -3) from ALA, (see **Figure 1** for metabolism of EFAs). AA and EPA form precursors to 2 and 3 series prostaglandins (PGs), thromboxanes (TXs), and 4 and 5 series leukotrienes (LTs). Non-steroidal anti-inflammatory drugs (NSAIDs) inhibit the activity of cyclooxygenase (COX) to prevent the formation of pro-inflammatory PGs and TXs. On the other hand, steroids block the activity of both COX and lipoxygenase (LO) enzymes to inhibit the formation of PGs, TXs, and LTs [16,17]. EPA and DHA have anti-inflammatory actions. This is due to their ability to displace AA from the cell membrane and suppress the formation of inflammatory eicosanoids from AA (such as 2 series PGs, TXs, and LTB₄). Furthermore, eicosanoids from EPA show less pro-inflammatory actions compared to those formed from AA. Exudates obtained in the resolution phase of inflammation treated with aspirin, DHA, and EPA produce bioactive 17R-hydroxy-containing di- and tri-hydroxydocosanoic acids termed resolvins. Resolvins dampen neutrophil infiltration and suppress IL-1 and TNF- α synthesis [18,19]. This is one mechanism by which ω -6 and ω -3 fatty acids AA, EPA, and DHA modulate inflammation. This suggests that formation of adequate amounts of resolvins at the site of inflammation is necessary for tissue repair, and restoration of normal structure and function of the inflamed tissues. It is possible that dysregulation of inflammation that occurs in sepsis is due to an alteration in the metabolism of EFAs.

Essential fatty acids in sepsis

LPS (lipopolysaccharide) endotoxin, a component of the bacterial cell wall, is known to regulate the synthesis of eicosanoids from their precursors. LPS increases the synthesis of macrophage PGs by transcriptional upregulation of COX-2 enzyme [20,21]. Eicosanoids have both pro-inflammatory and anti-inflammatory and immunosuppressive actions depending on the type of eicosanoid formed [16,17]. In the initial stages of sepsis, there is increased release of AA from the cell membrane that leads to the formation of pro-inflammatory eicosanoids. But, it is known that anti-inflammatory agents are not useful in sepsis.

It was observed that EPA and DHA are of benefit in sepsis [22,23]. In sepsis, inappropriate vascular

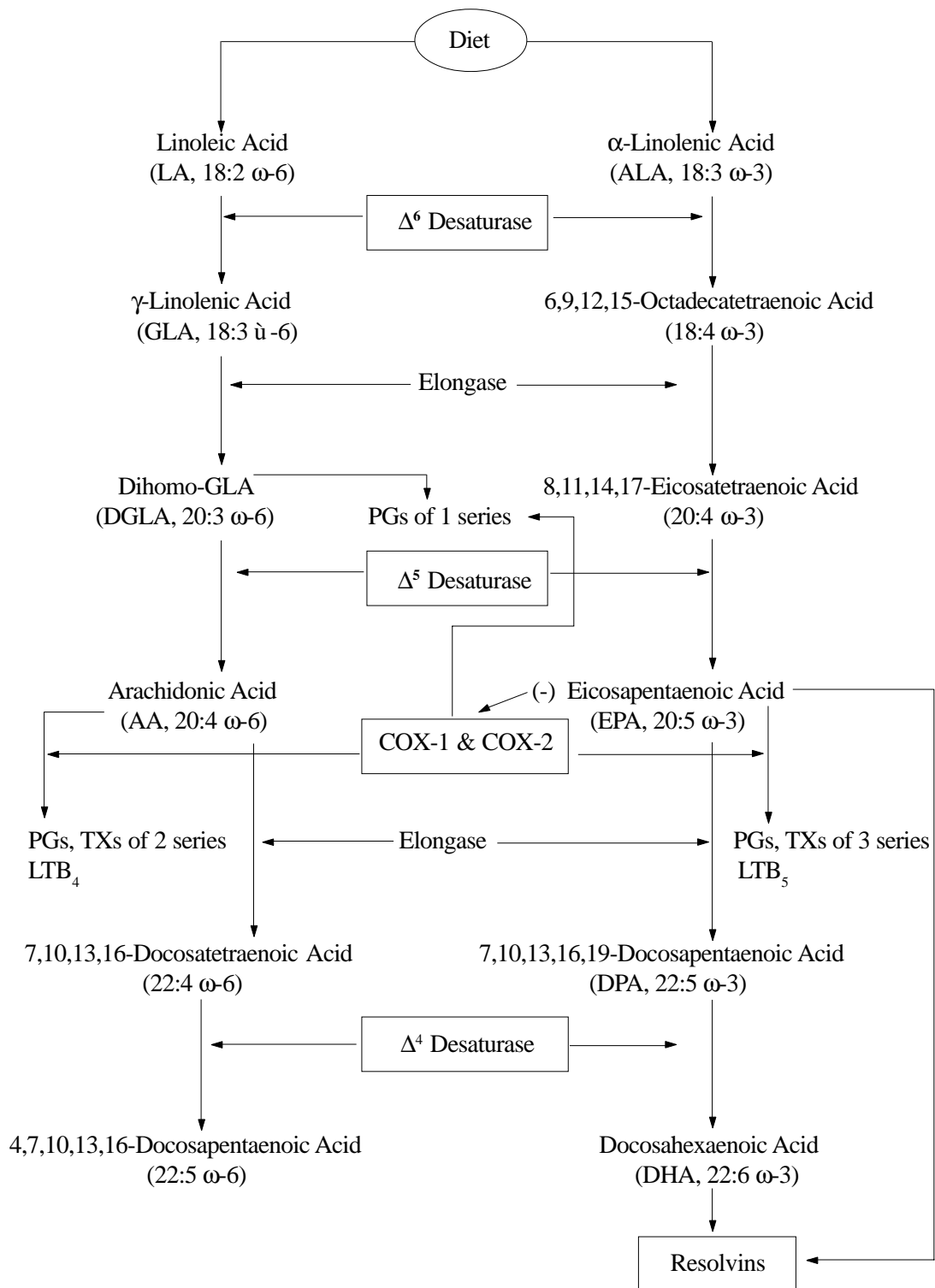


FIGURE 1. METABOLISM OF ESSENTIAL FATTY ACIDS.

thrombosis and hypotension could be due to increased production of TXA_2 and PGI_2 . ω -3 fatty acids may restore this imbalance to normal by inhibiting AA me-

tabolism. In a prospective, multicentered, double-blind, randomized controlled trial EPA + GLA produced significant improvements in oxygenation ($\text{PaO}_2/\text{FIO}_2$),

decreased necessity for ventilatory support and reduced length of stay in the intensive care units in those with ARDS compared with controls [24]. Earlier, I observed that patients with sepsis have low plasma concentrations of GLA, AA, and EPA [25]. Furthermore, TNF- α treatment caused EFA deficiency and decreased phospholipid EPA and DHA in human umbilical vascular endothelial cells (HUVEC) [26]. On the other hand, EPA and DHA suppress TNF- α production and action. This suggests that there is a close interaction between EFAs and their metabolites and TNF- α that implies their role in sepsis.

Phospholipase A₂ in sepsis

Increased amounts of eicosanoids formed during sepsis are due to the activation of phospholipase A₂ (PLA₂). Inhibition of cytosolic PLA₂ significantly attenuated lung injury, polymorphonuclear neutrophil sequestration, and deterioration of gas exchange and decreased lung injury in a murine model of ARDS [27]. PLA₂ activation induced specific changes in β adrenergic receptors seen during sepsis [28]. Furthermore, β adrenergic agonist restored PL (phospholipids) to normal levels and reduced PLA₂ activity to normal in sepsis [29]. Decreased plasma concentrations of lysophosphatidylcholine (LPC) strongly predict sepsis-related mortality [30]. These evidences suggest that lipids play a critical role in sepsis.

Acute septic shock and severe sepsis

Patients with acute septic shock syndrome die within 24-48 hours, whereas some with septic shock develop severe sepsis. Severe sepsis runs a long course over several weeks and patients succumb to the disease slowly. Autopsy in these (severe sepsis) patients shows only minimal signs of inflammation or necrosis. Some with severe sepsis develop septic shock. This suggests that severe sepsis and acute septic shock are two phases of the same syndrome and can occur in the same patient but at different time periods indicating that the causative mediators of them are different.

Overproduction of TNF- α triggers circulatory collapse, renal and hepatic failure, and widespread inflammation and injury. TNF- α -mediated septic shock occurs suddenly giving little time to generate sufficient amounts of neutralizing TNF- α antibodies in the body. On the other hand, TNF- α is not significantly increased in severe sepsis. High-mobility group box-1 (HMGB1)

may cause severe sepsis. HMGB1, released by activated macrophages and monocytes, induces the release of other proinflammatory cytokines; causes epithelial cell barrier dysfunction, lung injury, fever and lethality but not shock. Antibodies to HMGB1 protected experimental animals against severe sepsis [31,32]. Despite these advances, it is still not clear what triggers the development of severe sepsis in patients with acute septic shock and vice versa.

Lysophosphatidylcholine in sepsis

Lysophosphatidylcholine (LPC), a major component of oxidized low-density lipoprotein, stimulates monocytes, macrophages, T lymphocytes and neutrophils. LPC stimulated neutrophils destroy ingested bacteria by increasing the production of hydrogen peroxide (H₂O₂) (33). LPC facilitates elimination of bacteria. In patients with sepsis, plasma levels of LPC are low. LPC decreased TNF- α and IL-1 β production and enhanced T_H1 (T helper type 1) cytokines interferon- γ (IFN- γ), IL-2 and IL-12. Deactivation of neutrophils is common in sepsis and hence is unable to produce adequate H₂O₂ to kill the bacteria. LPC by activating neutrophils and enhancing the neutrophil H₂O₂ production helps in the elimination of the invading bacteria. In addition, LPC prevented LPS-induced TNF- α and IL-1 β release from neutrophils, and protected experimental animals from sepsis-induced lethality. These results suggest that LPC is effective against sepsis and septic shock. Since LPC concentrations are significantly lower in patients who die of sepsis compared to those who survive, further studies are needed to explore the use of LPC in the clinic.

Conclusions

Acute septic shock and severe sepsis are common in intensive care units. ω -3 and ω -6 fatty acids, eicosanoids, LPC, and ceramide seem to play a significant role in sepsis. Cytokines and lipids interact with each other that may have relevance to their role in sepsis and its outcome. Ceramide has a significant role in ARDS (34). Hence, it is important to study the exact interaction between cytokines, free radicals, ceramide, LPC and other lipids and their specific roles in sepsis (**Figure 2**). Such knowledge helps to devise better methods of management of patients with acute septic shock and severe sepsis to improve their recovery.

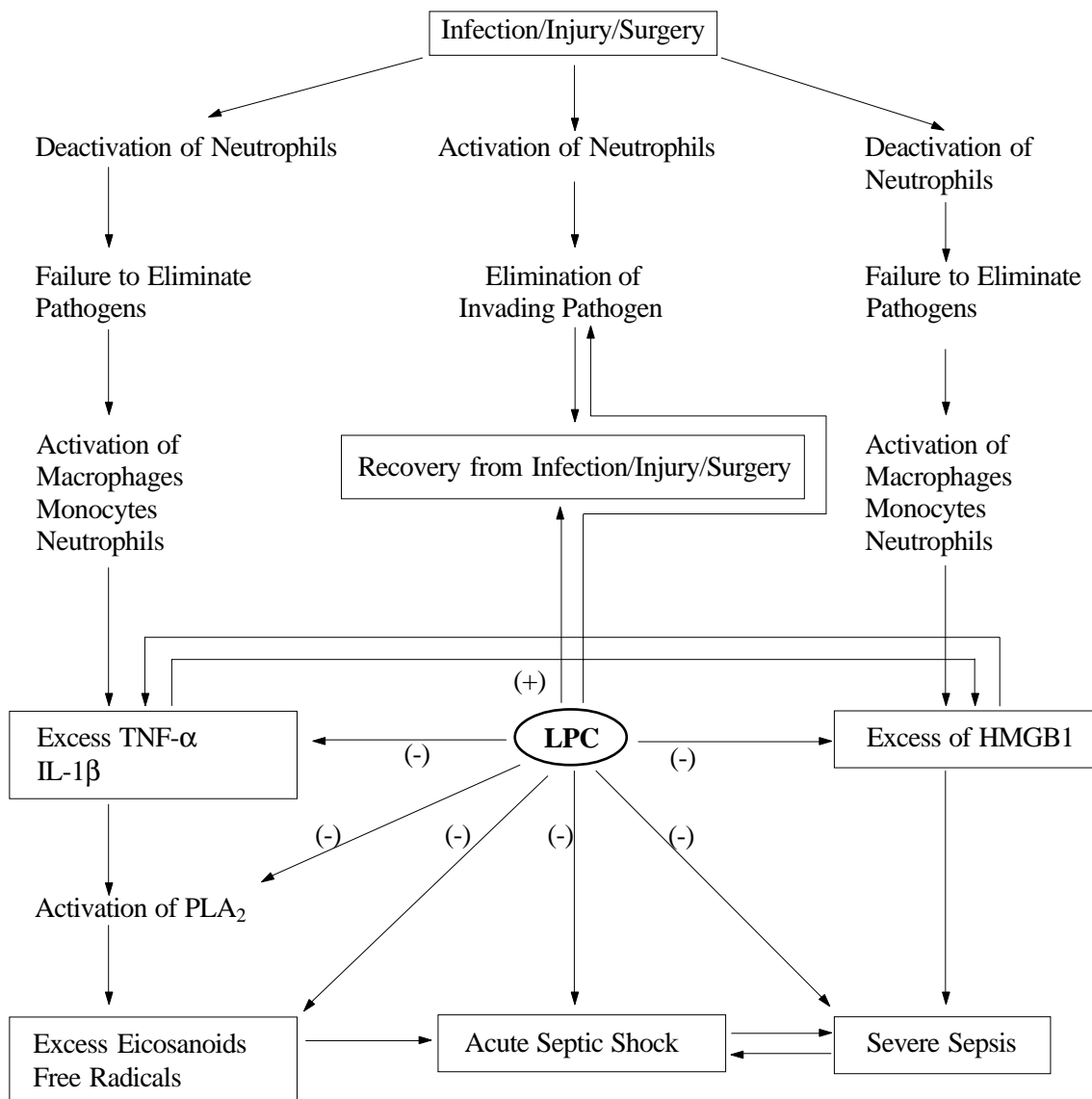


FIGURE 2. SCHEME SHOWING THE ROLE OF LYSOPHOSPHATIDYLCHOLINE (LPC) IN ACUTE SEPTIC SHOCK AND SEVERE SEPSIS. HMGB1 = HIGH MOBILITY GROUP BOX-1. (-) INDICATES NEGATIVE CONTROL/INHIBITION OF ACTIVITY/SYNTHESIS. (+) INDICATES POSITIVE INFLUENCE/RECOVERY FROM THE PROCESS.

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