Albumin and Lipid Enriched Albumin for the Critically Ill

UN Das

Abstract
Albumin, the principal transporter of plasma fatty acids, binds to majority of the drugs ingested, traps oxygen radicals and has potent anti-oxidant actions. Albumin binds to its specific binding sites on vascular endothelial cells and thus, prevents endothelial apoptosis. Albumin regulates the enzyme pyruvate dehydrogenase, the flux of glucose and lactate in astrocytes, and enhances the formation of anti-inflammatory lipoxins, resolvins and protectins from docosahexaenoic acid (DHA) and other polyunsaturated fatty acids that, in turn, could limit ischemia-induced neuronal damage. This may explain the beneficial action of DHA-enriched albumin in stroke and other critical diseases.

INTRODUCTION
Ischemic cerebrovascular disease causes significant morbidity and mortality. There has been little progress in the development of newer therapeutic strategies in the management of stroke except for the discovery that administration of recombinant human tissue-type plasminogen activator (rh-tPA), a thrombolytic agent, is of benefit when given within 3 hours of its onset. This suggests that more effective measures are needed for the treatment of stroke.

Sepsis is another major disease that causes more than 200,000 deaths per year in U.S.A. alone. Mortality in sepsis is due to multiple organ dysfunctions (MODS = multiorgan dysfunction syndrome) and so the prognosis of patients with sepsis is related to the severity of organ dysfunction. Clinical trials that studied the effect of agents that block the inflammatory cascade—corticosteroids, anti-endotoxin antibodies, tumor necrosis factor antagonists, and interleukin-1-receptor antagonists, have failed to reduce deaths due to sepsis. Recent studies suggest that albumin could be of benefit in sepsis. This beneficial action could be due to the ability of albumin to trap oxygen radicals and its antioxidant actions.

ALBUMIN
Albumin, the major protein produced by hepatocytes in the liver, is not stored in the liver but is excreted into the hepatic lymph system or the sinusoids and accounts for approximately 50% of the serum proteins. The circulation half-life of albumin is approximately 16 hours, whereas the degradation half-life of albumin is 17 to 20 days. Albumin maintains oncotic pressure and hence, its loss would result in dependent edema, hypotension, and, at times, circulatory collapse.

The indications for albumin therapy include: hypovolemia or shock, burns, hypoalbuminemia, surgery or trauma, cardiopulmonary bypass, acute respiratory distress syndrome (ARDS), hemodialysis, and sequestration of protein-rich fluids. The beneficial effects of albumin in cirrhosis of the liver are very modest and limited only to patients with slightly impaired renal function. In contrast, albumin infusions are effective in preventing the deterioration in renal function associated with large volume paracentesis or spontaneous bacterial peritonitis. This suggests that albumin prevents renal impairment by maintaining effective arterial blood volume in situations characterized by acute deterioration in circulatory function. The recent observation that concomitant administration of albumin and vasoconstrictor drugs acting preferentially in the splanchnic circulation normalizes almost completely circulatory function and improves renal function in patients with cirrhosis and hepatorenal syndrome opens a new indication for albumin infusions in patients with liver disease.

Albumin traps oxygen radical and quenches free radicals; inhibits copper ion-dependent lipid peroxidation and retards the formation of hydroxyl radicals. Many other antioxidants present in the body are not neuroprotective, whereas albumin shows neuroprotective and cytoprotective action. This beneficial action of albumin is due to its ability to mobilize docosahexaenoic acid (DHA) and, possibly, other polyunsaturated fatty acids (PUFAs) from liver and other tissues which, in turn, are converted to anti-inflammatory molecules such as protectins, lipoxins and resolvins that are...
of benefit in stroke and sepsis.

**USE OF ALBUMIN IN THE CRITICALLY ILL**

Hypoalbuminemia, common in the critically ill, is associated with high degree of morbidity and mortality. Paradoxically, administration of 25% albumin to hypalbuminemic and critically ill medical and surgical patients did not show any benefit despite having achieved and maintained serum albumin levels of 25 g/L (2.5 g/dL) or greater,11-13 In contrast, in a prospective, controlled, randomized study of a mixed population of critically ill patients, administration of albumin to correct hypoalbuminemia showed improved respiratory, cardiovascular, and central nervous system functions.14 These controversial results could be attributed to: heterogeneity of patients studied; presence of other illnesses such as infections, associated renal, liver or lung diseases; abnormalities in their immune responses to the underlying disease or infection(s); variations in their cytokine profiles, endogenous concentrations of polyunsaturated fatty acids and their products; nutritional status; associated reactive hyperglycemia and insulin resistance; and possibly, differences in the dose of albumin employed.15-23 In this context, the observation that albumin shows cytoprotective actions by enhancing the formation of anti-inflammatory molecules: lipoxins, resolvins and neuroprotectin D1 (NPd1) is interesting.

**NEUROPROTECTIVE ACTION OF ALBUMIN**

Animal Studies

Albumin therapy (2.0 to 2.5 g/kg) administered within 2 hours after the onset of stroke, is effective in improving neurological status in animals both by a reduction in infarction volume and cerebral edema.24-26 Sprague-Dawley rats given different doses of human albumin (0.63 or 1.25 g/kg) at different time intervals (2, 3, 4, or 5 hours after onset of middle cerebral artery occlusion), showed that 1.25 g/kg albumin dose significantly improved the neurological score compared with control at 24, 48, and 72 hours.27 Although, several different doses of albumin is effective in improving neurological function, for reducing the volume of cerebral infarction, cerebral edema, diminished brain infarction and improved local perfusion to zones of critical blood flow reduction the optimum dose appears to be 1.25 g/kg.28 Ischemia-induced blood-brain barrier dysfunction permits albumin to penetrate into the brain parenchyma, where it was taken up by cortical neurons and thus, are protected from ischemic injury.28 Albumin prevented necrosis of neurons in tissue zones of residual ischemic injury by preserving glial and endothelial elements and normalized diffusion coefficient of water even in zones of residual histological injury.28 Albumin regulates the enzyme pyruvate dehydrogenase in astrocytes that helps in the flux of glucose and lactate which aids in the limitation of neuronal damage due to cerebral ischemia. These evidences suggest that albumin has a direct protective effect on both parenchymal and vascular tissues of the brain, and protects vulnerable neurons of the hippocampus from injury and diminished contusion volume.29,30

**ALBUMIN IN SEPSIS**

Studies done in an infant rat model it was noted that phosphate buffered saline (PBS) containing 0.5% bovine serum albumin (PBS-BSA) protected against challenge by Haemophilus influenzae type b compared to the control.31 Albumin traps oxygen radicals, inhibits copper ion-dependent lipid peroxidation and retards the formation of hydroxyl radicals. Nitric oxide (NO) is an endogenous vasodilator and modulator of inflammation. During endotoxemia, the beneficial effects of NO are overwhelmed by the inflammatory cascade, resulting in a functional depletion of NO. S-nitroso-albumin (S-NO-alb) is a stable NO thiol complex that slowly releases NO into the vascular micro-environment. In a porcine model of LPS-induced cardiopulmonary dysfunction, pretreatment with intravenous S-NO-alb improved cardiopulmonary dysfunction, blunted LPS-induced hypoxic response and reduced neutrophil activation. These results suggest that S-NO-alb modulates endotoxin-induced pulmonary dysfunction, attenuates neutrophil priming and blocks early mortality.32

Acute lung injury occurs in severe sepsis requiring mechanical ventilation. In a rodent model of acute lung injury induced by intratracheal LPS, 25% albumin resuscitation diminished cytokine-induced neutrophil chemoattractant messenger RNA concentrations, nuclear factor-kappaB (NF-κB) translocation, decreased plasma lipid peroxidation, reduced plasma concentrations of tumor necrosis factor-α, interleukin-6, macrophage inflammatory protein-2, and hydrogen peroxide formation and enhanced interleukin-10 levels, and protected from lung injury.33 Resuscitation with albumin reduced ventilator-induced lung injury after hemorrhagic shock, but not after endotoxic shock, suggesting that the mechanisms leading to ventilator-induced lung injury after hemorrhage differ from those after endotoxemia.34 PEG-Alb (albumin covalently linked to polyethylene glycol), wherein PEG is covalently linked to human albumin at multiple sites on the protein, the effective volume of PEG-Alb is increased 13- to 16-fold compared with unmodified albumin. In an LPS (lipopolysaccharide) model of shock, rats treated with PEG-Alb showed better blood pressure, lower haematocrit consistent with haemodilution and less lung injury than rats treated with unmodified albumin or saline. In a CLP (caecal ligation and puncture) model of sepsis, PEG-Alb was more effective than albumin or saline in maintaining blood pressure and in decreasing haematocrit. Rats with LPS- or CLP-induced shock, PEG-Alb were retained within blood vessels unlike albumin that extravasates into the interstitial space,35 indicating that PEG-Alb may be more effective in the treatment of shock associated with capillary leak.

The beneficial action of albumin in critical illness is further
supported by the observation that albumin resuscitation improved decreased ventricular contractility and myocardial oxygenation by improving ventricular dysfunction by reducing myocardial hypoxia in endotoxemic rats, an effect that is attributed to an albumin-induced reduction in inducible nitric oxide synthase protein and messenger RNA expression following endotoxin injection. PEG-BSA (PEG coupled to bovine serum albumin) significantly improved microvascular flow and perivascular and tissue P(O2), normalized shear rate, and decreased perivascular nitric oxide concentration. These beneficial effects are due to improved fluid retention by PEG-BSA and its ability to modulate NO levels.

Human studies

Despite these beneficial results observed in animal models of sepsis with albumin studies in patients with sepsis have given controversial results. Rackow et al studied twenty consecutive patients with severe sepsis who were randomized to fluid challenge with 5% albumin or 10% low MW hydroxyethyl starch (pentastarch) solutions. Although all of the patients had similar impairments in oxygenation and chest roentgenograms, those patients who received albumin showed significantly reduced morbidity and mortality.

In a case-controlled study of patients who sustained burns of > or =20% total body surface area, those who received albumin showed decreased likelihood of mortality compared to those who did not receive albumin. In contrast, in a multicenter, randomized, double-blind trial in a heterogeneous population of patients in the ICU, no significant benefit was seen in those who received albumin infusions. Despite these controversial results, since albumin traps oxygen radicals, suppress production of pro-inflammatory cytokines, and enhances endothelial NO generation, desirable actions in the setting of sepsis and in the critically ill, more thorough studies are needed to explore the clinical values of albumin. In this context, the observation that albumin when conjugated with docosahexaenoic acid (DHA) shows beneficial actions in the treatment of stroke is particularly interesting.

**ALBUMIN FOR STROKE**

Ischemic cerebrovascular disease is a leading cause of death and long-term disability. Studies performed in the middle cerebral artery occlusion albino rat model, intravenous administration of moderate dose of albumin (1.25 g/kg) as late as 4 hours after onset of middle cerebral artery occlusion, improved the neurological score, reduced infarct volume in cortex (by 65%), subcortical regions (by 52%), and total infarct (by 61%) Although the exact mechanism by which albumin prevents brain damage is not clear, one potential explanation appears to be its ability to induce selective mobilization of n-3 polyunsaturated fatty acids (PUFAs) especially that of DHA (22:6) and docosapentaenoic acid (DPA, 22:5 n-3) from liver.

**LIPOXINS, RESOLVINS, AND NEUROPROTECTIN D, PREVENT NEURONAL DAMAGE**

Neuronal membranes are rich in PUFAs: arachidonic acid (AA, 20:4 n-6), eicosapentaenoic acid (EPA, 20:5 n-3) and DHA. AA can be obtained from diet or by the conversion of dietary essential fatty acid (EFA) linoleic acid (LA, 18:2 n-6) by the action of the enzymes Δ6 and Δ5 desaturases; whereas EPA and DHA also can be obtained from diet or by the conversion of dietary EFA: α-linolenic acid (ALA, 18:3 n-3) by the same set of desaturases (Fig. 1 for metabolism of LA and ALA). AA and EPA give rise to the formation of various prostaglandins (PGs), thromboxanes (TXs) and leukotrienes (LTs) that are mainly pro-inflammatory in nature. AA, EPA, and DHA also form precursors to anti-inflammatory molecules: lipoxins (LXs), resolvins, and protectins that suppress inflammation. Thus, the balance between these mutually antagonistic compounds formed from PUFAs could determine the final outcome of the disease process. Nitration of unsaturated fatty acids can give rise to biologically active compounds called as nitrolipids that stimulate smooth muscle relaxation, block platelet activation, inhibit human neutrophil functions and suppress inflammation.

Aspirin converts AA, EPA and DHA to form aspirin-triggered 15-epimer LXs (ATLs) that are potent inhibitors of inflammation. Endothelial cells interact with PMNs

---

**Fig. 1**: Scheme showing the metabolism of essential fatty acids and formation of PGs, lipoxins, resolvins and NPD, from various PUFAs.
leading to the formation of 15R-HETE and its subsequent conversion to 15-epimeric LXs by aspirin-acetylated COX-2 enzyme. Deficiency or absence of LXs leads to adhesion of PMN to endothelial cells as a result of which endothelial damage occurs that could lead to initiation and progression of inflammation.

Compounds similar to 15R-HETE and 15-epimeric LXs are also formed from EPA and DHA that have potent anti-inflammatory actions and induce resolution of the inflammatory process and hence are called “resolvins” (Fig. 2). Resolvins inhibited cytokine generation, leukocyte recruitment, leukocyte diapedesis, and exude formation. Resolvins inhibit brain ischemia-reperfusion injury. Thus, lipoxins and resolvins formed from AA, EPA, and DHA have neuroprotective actions.

One of the 17-hydroxy-containing bioactive mediators derived from DHA is termed as neuroprotectin D1 (NPD1, also called as 10,17S-dihydroxydocosatetraene) since it reduced infiltration of PMNs, suppressed inflammation and showed neuroprotective properties. PND1 inhibited oxidative stress-induced apoptosis of human retinal pigment epithelial cells, and both LXs and NPD1 enhanced wound healing, and promoted brain cell survival via the induction of anti-apoptotic and neuroprotective gene-expression programs.

**ALBUMIN-DHA COMPLEX IS NEUROPROTECTIVE IN NATURE**

Administration of albumin-DHA complex containing 2.1 ± 0.1 micro (μ) mol DHA per milliliter of albumin to the 2-hour middle cerebral artery suture-occlusion animal model, a high degree neurobehavioral and histological neuroprotection was noted. Albumin-DHA complex facilitates DHA delivery to the brain though the astrocytic foot processes of the microvasculature so that significant amounts of NPD1 could be formed to prevent ischemia-reperfusion injury. DHA is necessary for ion channels, receptors, and transporters to maintain their integrity. DHA confers neuroprotection by opening background K+ channels and inhibiting apoptosis. Administration of albumin-DHA complex increased the formation of NPD1, and infusion of NPD1 reduced infarct size, diminished polymorphonuclear leukocyte infiltration, NF-κB activation, and pro-inflammatory cyclo-oxygenase-2 expression.

**ALBUMIN IS BENEFICIAL IN CEREBRAL MALARIA**

Children with severe falciparum malaria showed a survival benefit when infused with 4.5% albumin compared to other resuscitation fluids. The beneficial action of albumin is due to its colloidal properties that ameliorates brain swelling, improvement of blood flow by its direct effects on the endothelial cells, and its non-colloidal properties. It is likely that albumin infusion in severe malaria increased the formation of lipoxins and NPD1 due to the mobilization of DHA that, in turn, produced neuroprotection.

**CONCLUSIONS**

Albumin when complexed with DHA enhances the formation of NPD1, lipoxins, and resolvins that have anti-inflammatory actions and thus, is beneficial in stroke, sepsis, and severe falciparum malaria. The controversial and sometimes diametrically opposite results noted in various trials with regard to albumin therapy in sepsis and ARDS could be due to differences in the dose and duration of albumin therapy employed. It is likely that albumin used in different trials contained varying concentrations of DHA that could have skewed the results. Since albumin mobilizes DHA from the liver, the amount of DHA stored in the liver could be another variable that influenced the amounts of NPD1 formed. The activity of the enzymes concerned with the formation of NPD1 may also vary depending on the underlying clinical condition that could have contributed to variations in the response to albumin therapy reported.

Albumin fully restored granulocyte-macrophage (CFU-GM) and erythrocyte colony forming units to control values in experimental animals following trauma/hemorrhagic shock that could be attributed to the binding of circulating toxic factors to albumin. It is possible that albumin enhanced the formation of NPD1 lipoxins and resolvins that have prevented bone marrow suppression following trauma/hemorrhagic shock. Albumin kinetics are altered in those with sepsis and critically ill. The half-life time in sepsis is shorter compared to control group (8.2 ± 1.4 vs. 12.5 ± 1.7, P < 0.01), the transportation rate is also higher in sepsis than in the control group, suggesting that the distribution rate of albumin from vessel to tissue is increased and the decomposition rate of albumin is markedly changed in sepsis. In view of these facts, more thorough and well-designed studies are needed.

---

**Fig. 2:** Scheme showing the relationship between various mediators of tissue damage/resolution in various clinical conditions and the role of albumin, lipid-enriched albumin, PUFA's, lipoxins/resolvins, NPD1, and nitrolipids in these processes/diseases.
needed to determine the beneficial or adverse reactions to PEG-Alb, albumin+dHA, and S-NO-alb complex in severe malaria, stroke, sepsis and ARDS. Plasma levels of various pro- and anti-inflammatory molecules: IL-6, TNF-alpha, MIF (macrophage migration inhibitory factor), HMG1 (high-mobility group box 1), nitric oxide, lipid peroxides, anti-oxidants, and prostaglandins and lipoxins, resolvins, NPD, and nitrolipids need to be measured to know which of the harmful and/or beneficial molecules are altered in albumin therapy (Fig. 2). In these studies, kinetics of albumin, PEG-Alb, albumin+dHA complex, and S-NO-alb infused need to be studied to know the dynamics of the administered dose. Albumin has been reported to be safe and well tolerated when given in a dose range from 0.34 to 2.05 g/kg to patients with acute ischemic stroke42,63 implying that its beneficial action in this condition needs to be evaluated in humans.

Acknowledgements

Dr. Das was in receipt of Ramalingaswami fellowship of Department of Biotechnology, India during the period of this study.

REFERENCES


Announcement

State Chapter and City Branches
Attention Chairmen/Secretaries

A Meeting of Central API office bearers with State Chapter and City Branches of the Chairmen and Secretaries will be held during APICON at Greater Noida on 30th January 2009 at 4.00 p.m. at India Expo Centre, Greater Noida, NCR.
All Chairmen/Secretaries of the State Chapters/City Branches of API are requested to attend the meeting.

Dr. Sandhya Kamath
Hon. General Secretary

---

Announcement

Indian College of Physicians
(Sponsored Training Programme (STP))

The Indian college of Physicians under the auspices of APC is conducting sponsored training programme every year in various specialized subjects at specialized institutes recognized by IPC on the subjects of Cardiology, Gastroenterology, Nephrology, Hematology, Endocrinology, Oncology and Diabetology. The ICP gives certificate for the training to the candidates. Deserving candidates should apply to the Dean's Office on the address given below. The STP fellowship grant has been raised to Rs.7,500/- per month for 2 months and a total of 7 fellowships will be awarded during the training of ICP.

For further details please contact: Prof. Dr. A.K. Das, Director Prof. of Medicine and Medical SuperintendentJIPMER, Puducherry-6.
Tel. Office : 0413-2272735; Mobile : 9789457887; Email : ashokdas82@gmail.com
Prof. Dr. T.K. Dutta, HOD Medicine, JIPMER Puducherry-6.
Tel. Office : 0413-2272047; Mobile : 9443602330

The last date for application to reach to this office is 23/01/2009.

(Dr. A.K. Das)
Medical Superintendent