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The role of the host defence response in the progression and outcome of ARDS: pathophysiological correlations and response to glucocorticoid treatment

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The role of the host defence response in the progression and outcome of ARDS: pathophysiological correlations and response to glucocorticoid treatment. G.U. Meduri. ©ERS Journals Ltd 1996.

ABSTRACT: The host defence response (HDR) to insults is similar regardless of the tissue involved and consists of an interactive network of simultaneously activated pathways that act in synergy to increase the host's chance of survival. Among this cascade of integrated pathways, three aspects of the HDR, inflammation, coagulation and tissue repair, are analysed separately to explain the histological and physiological changes occurring at the tissue level in unresolving acute respiratory distress syndrome (ARDS). Cellular responses in HDR are regulated by a complex interaction among cytokines, and cytokines have concentration-dependent biological effects. The degree of initial HDR may determine the progression of ARDS. On Day 1 of mechanical ventilation and over time, nonsurvivors of ARDS have significantly higher plasma and bronchoalveolar lavage inflammatory cytokine levels than survivors. In the absence of inhibitory signals, the continued production of HDR mediators prevents effective restoration of lung anatomy and function by sustaining inflammation with tissue injury, intra- and extravascular coagulation and proliferation of mesenchymal cells (fibroproliferation) with deposition of extracellular matrix resulting in fibrosis.

Glucocorticoids inhibit the HDR cascade at virtually all levels; their gradual and generalized suppressive influence protects the host from overshooting. In patients with exaggerated HDR, however, cytokine elevation may cause a concentration-dependent resistance to glucocorticoids by reducing glucocorticoid receptor binding affinity. Recent clinical and experimental studies have shown that effective containment of the HDR in unresolving ARDS may be achieved only if glucocorticoid administration is prolonged. A double-blind randomized study is in progress to evaluate the role of prolonged glucocorticoid treatment in unresolving ARDS.
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Acute respiratory distress syndrome (ARDS) describes the clinical syndrome associated with the morphological lesion termed "diffuse alveolar damage" (DAD). At presentation, (early) ARDS manifests with acute and diffuse injury to the endothelial and epithelial lining of the terminal respiratory units and causes increased vascular permeability with protein-rich exudative oedema. Despite improvements in supportive care and multiple therapeutic efforts directed at modifying the course of this condition, mortality still remains above 50%. In nonsurvivors of ARDS (table 1), DAD rapidly advances through three histological phases (exudative, fibroproliferative and fibrotic) with different clinicophysiological features. The objective of this review is to describe systematically the pathophysiology of unresolving ARDS relating to mechanisms of its development, structural alterations induced in the lung and functional consequences of these morphological changes. Intentionally, I will place the host defence response (HDR) at centre stage as a critical reference point for explaining how ARDS progresses in nonsurvivors and to provide a rationale to evaluate glucocorticoid (GC) treatment and other therapeutic strategies.

Most ARDS patients (85–95%) survive the initial direct or indirect insult that precipitated acute respiratory failure and progress into the reparative stage, where outcome varies (table 2) from complete recovery (groups 2 and 3) to rapid death from accelerated pulmonary fibrosis (group 5) [11]. In patients who recover, the permeability defect and gas exchange abnormalities do improve (adaptive response) [12]. Conversely, ineffective repair is manifested by progression of fibroproliferation, inability to improve lung function (groups 4 and 5) and unfavourable outcome (maladaptive response) [10]. Most nonsurvivors invariably develop fever, systemic inflammatory response syndrome (SIRS) [13], clinical manifestations of sepsis [14–16] and multiple organ dysfunction syndrome (MODS) [5] and die after a prolonged period of ventilatory support (group 4). At autopsy, ARDS nonsurvivors are found to have extensive pulmonary fibrosis in 55% and pneumonia in 69% of cases [10]. Patients whose disease results in ineffective lung repair require

Table 1. – Histological changes in unresolving ARDS

	Exudative	Proliferative	Fibrotic
Timing	Oedema	Organization (repair)	Fibrosis
	Early (<1 week)	Intermediate	Late (3 weeks)
Macroscopic			
Consistency	Rigid, heavy	Firm, consolidated	Spongy, cystic
Appearance	Haemorrhagic	Pale grey	Pale
Microscopic			
Vasculature	Endothelial injury (mild)	Endothelial injury	Endothelial injury
	Congestion	Intimal fibroproliferation	Distortion
	Neutrophil aggregates	Medial hypertrophy	Compressed
	Minimal thrombi	Thrombi	Proliferation
Alveoli	Type 1 pneumocyte necrosis	Type 2 pneumocyte proliferation‡	Fibrosis
	Inflammatory exudate	Myofibroblast invasion	Microcysts
	Hyaline membranes#	Increased fibronectin	
	Partial collapse	Collagen deposition	
Basement membrane	Denuded	Gaps with myofibroblast invasion	Disruption
Alveolar wall	Oedema	Myofibroblast proliferation	Thick collagen
Alveolar duct	Dilated	Myofibroblast proliferation	Fibrosis
Interstitium	Volume↑	Volume↑↑	Volume↑↑↑
	Oedema	Myofibroblast proliferation	Fibrosis
Pleura	Subpleural ischaemic changes	Subpleural necrosis	Subpleural necrosis

*: the endothelial cell layer is usually continuous with cell junction; endothelial gaps are rarely observed [1]; ‡: stem cell function for the entire epithelium; #: hyaline membranes are composed of fibrin, fibronectin and cellular debris adhering over denud-

Table 2. – Clinical classification of ARDS

	Clinical group				
	1	2	3	4	5
Source of injury controlled	No	Yes	Yes	Yes	Yes
Percentage of patients %	5–15	10	20–40	40–60	5–10
Average duration of ARF days	≤3	≤7	7–28	7–28	≤7
Evolution	Rapidly fatal	Rapid recovery	Slow improvement	Slow deterioration	Rapidly fatal
Survival	No	Yes	Yes	No	No
Causes of death	MODS	-	-	10–40% pulmonary 60–90% MODS	Pulmonary
Histological findings	Exudative phase of ALI	-	Fibroproliferation*	55% extensive fibrosis 69% pneumonia**	Severe fibrosis

*: data obtained from open lung biopsy[2–4]; **: data obtained from autopsy series [5–9]. ARF: acute respiratory failure; MODS: multiple organ dysfunction syndrome; ALI: acute lung injury; ARDS: acute respiratory distress syndrome. Reproduced with permission from reference [10].

continuous ventilation (unresolving ARDS) and are predisposed to develop pneumonia, extrapulmonary infections and sepsis. Sepsis has been associated with (but not proven to be causative of) death in 36–90% of ARDS nonsurvivors [15–18]. The hypothesis has been formulated on clinical criteria that, in unresolving ARDS, serious nosocomial infections, such as ventilator-associated pneumonia (VAP) amplify SIRS and lead to MODS and death [5, 6, 15, 16]. MODS develops more frequently in patients dying with sepsis than in those dying with refractory hypoxaemia [19–20]. Progression of fibroproliferation to extensive pulmonary fibrosis directly causes respiratory death in 15–40% of ARDS patients (table 2) [7, 15, 17].

Because there is no animal model to study the progression of ARDS, clinical investigation has become of primary importance for advancing understanding in this field. Our group has recently completed a series of clinical studies designed to clarify key pathogenetic aspects of ARDS progression and outcome. We have obtained serial quantitative measurements of HDR mediators in the lung and in the circulation and have correlated these measurements with pathophysiological variables over time. ARDS was studied during its natural progression and also in response to GC rescue treatment. During these investigations, a careful effort was made to accurately diagnose VAP (bronchoscopic methods) and pulmonary fibroproliferation (open-lung biopsy) following recently developed guidelines [21, 22]. In this review, I will highlight selected conceptual themes to provide a unifying pathogenetic model of ARDS, showing how an exaggerated and protracted HDR plays a key role in ARDS outcome and is accountable for the histological, laboratory, clinical and physiological findings observed during the course of unresolving ARDS.

Tissue host defence response

Tissue consists of organized groups of cells attached to an extracellular matrix (ECM) and surrounded by a network of blood vessels. The ECM occupies a significant proportion of the volume of any tissue and is indispensable for its structural integrity [23]. Tissue steady state, or homeostasis, is maintained by co-ordinating cell growth and proliferation with the production and turnover of ECM. Cells achieve a remarkable co-ordination by constantly signalling to themselves (autocrine activity) and each other (paracrine activity) by means of polypeptides called "cytokines" (also known as growth factors). Cell-cell and cell-matrix interactions, through cytokine networking, are essential not only for maintaining homeostasis, but also for providing a rapid defence (stress) response against intrinsic or extrinsic disturbing (infectious and noninfectious) forces.

The host defence response to insults is similar regardless of the tissue involved and consists of an interactive network of simultaneously activated pathways that act in synergy to increase the host's chance of survival. Among this complex cascade of integrated pathways, five aspects of the HDR are important for understanding the clinical development and evolution of ARDS (table 3): inflammation, coagulation (intravascular clotting and extravascular fibrin deposition), modulation of the immune response, tissue repair and activation of the

Table 3. – Components of the host defence response

Inflammation
  Vasodilation and stasis
  Increased expression of adhesion molecules
  Increased permeability of the microvasculature with exudative oedema
  Leucocyte extravasation
  Release of leucocyte products potentially causing tissue damage
Coagulation
  Activation of coagulation
  Inhibition of fibrinolysis
  Intravascular clotting
  Extravascular fibrin deposition
Modulation of the immune response
  Fever
  Induction of heat-shock proteins
  Release of neutrophils from the bone marrow
  Priming of phagocytic cells
  T-cell proliferation
  Antibody production
Tissue repair
  Angiogenesis
  Epithelial growth
  Fibroblast migration and proliferation
  Deposition of extracellular matrix and remodelling
Activation of the hypothalamic-pituitary-adrenal axis
  Release of ACTH with cortisol production
  ACTH and cortisol modulation of the sympathetic nervous system
  Cortisol modulation of acute-phase protein production by the liver[†]

*: initially polymorphonuclear cells and later monocytes; [†]: elevated levels of circulating glucocorticoids synergize with IL-6 in inducing hepatic synthesis and secretion of "acute-phase" reactants, such as fibrinogen, protease inhibitors, complement C3, ceruloplasmin, haptoglobin, and C-reactive protein. ACTH: adrenocorticotrophic hormone; IL-6: interleukin-6.

hypothalamic-pituitary-adrenal (HPA) axis with production of glucocorticoids. Activation of the sympathetic system to release catecholamines and increased hepatic production of acute-phase reactants are an integral part of the host defence response and are under the influence of glucocorticoids. During stress, epinephrine response parallels HPA axis stimulation [24]. Glucocorticoids and ACTH affect several key regulatory enzymes in catecholamine biosynthesis and influence the number and functional state of adrenergic receptors [24]. Catecholamine-glucocorticoid interactions play an important role in maintaining vascular tone [24]. Acute phase reactants are essential in many aspects of the host defence (fibrinogen (coagulation and tissue repair), complement C3 (opsonization), C-reactive protein (inhibitor of neutrophil chemotaxis) and protease inhibitors (limit tissue damage during inflammation)) and their production is enhanced by glucocorticoids [25, 26]. The HDR is essentially a protective response of tissues, which serves to destroy, dilute, or contain injurious agents and to repair any consequent tissue damage. Repair consists of replacing injured tissue by regenerating native parenchymal cells and filling defects with fibroblastic tissue. Three aspects of the HDR, (inflammation, coagulation and tissue repair) can be analysed separately to explain the histological and physiological changes occurring at the tissue level in unresolving ARDS.

Cytokines, glucocorticoids and catecholamines are capable of modulating gene expression, primarily by altering the rate at which a given gene is transcribed into messenger ribonucleic acid (mRNA). For this process to occur, the deoxyribonucleic acid (DNA) sequences that regulate transcription of that particular gene must be identified by the transacting moiety. This recognition process appears to be performed almost entirely by specific proteins called "transcription factors." These regulatory proteins are able, with great selectivity, to bind with high affinity to the correct DNA-binding site [24]. Cytokines and glucocorticoids enhance the expression and activity of transcription factors [24]. Important transcription factors associated with the HDR and discussed in this review include the glucocorticoid receptor complex, nuclear factor interleukin-6 (NF-IL6), NF-κB, activator protein-1 (AP-1 [c-Jun and c-Fos]), and the heat-shock transcription factor (HSF) [24].

Cellular responses in HDR are regulated by a complex interaction among cytokines with final effects on the surrounding microenvironment not directly induced by the initiating insult. In this regard, cytokines have concentration-dependent biological effects [27]. At low concentration they regulate homeostasis; at progressively higher concentrations they mediate proportionally stronger local and then systemic responses. Among a broad spectrum of proximal mediators, cytokines of the interleukin-1 (IL-1) and tumour necrosis factor (TNF) family appear uniquely important in initiating all key aspects of the host defence response. TNF-α and IL-1β stimulate their own and each other's secretion, and both promote the release of IL-6 (NF-IL6 is the transcription factor responsible for IL-6 gene activation after IL-1 stimulation). The cell most commonly associated with initiating the host defence response cascade is the tissue macrophage or the blood monocyte [25]. Once released, TNF-α and IL-1β act on epithelial cells, stromal cells

(fibroblasts and endothelial), the ECM and recruited circulating cells (neutrophils, platelets, lymphocytes) to cause secondary waves of cytokine release with amplification of the HDR [25].

In the absence of inhibitory signals, the continued production of HDR mediators sustains inflammation with tissue injury, intra- and extravascular coagulation and proliferation of mesenchymal cells (fibroproliferation) with deposition of ECM, resulting in fibrosis [28]. Generation of inflammatory cytokines in HDR is normally strongly controlled by a number of homeostatic regulatory mechanisms, including shedding of specific cytokine receptors on host cells, synthesis of endogenously generated cytokine antagonists, synthesis of anti-inflammatory cytokines, downregulation, and activation of the HPA axis with production of GCs. Naturally occurring cytokine antagonists, such as IL-1 receptor antagonist (IL-1RA) and soluble TNF receptor (sTNFR), are extremely potent regulators of the HDR. Anti-inflammatory cytokines (IL-4 and IL-10) downregulate TNF-α, IL-1β, and IL-8 and upregulate the expression of IL-1RA [25]. IL-4 mimics the action on human monocytes of the synthetic glucocorticoid dexamethasone [29]. IL-4 also enhances apoptosis of monocytes, leading to reduced accumulation of these cells in chronic inflammation [25]. It appears that IL-4 and IL-10 released in the vicinity of the reactive tissue site and the activity of glucocorticoids produced through the stimulation of the HPA axis are essential to regulating the termination of the HDR [25].

Production of glucocorticoids and the host defence response

Glucocorticoids inhibit the HDR cascade at virtually all levels; their gradual and generalized suppressive influence protects the host from overshooting [30]. This influence is assumed to be sufficiently delayed in relation to the initial stress stimulus to allow the appropriate defence mechanisms to become activated [30, 31]. However, this protective mechanism is not always effective, as demonstrated by the exaggerated and autodestructive host reaction seen in nonsurvivors of sepsis or ARDS. The encouraging results of recent studies with prolonged GC administration in patients with ARDS [2, 3, 32–34] and sepsis [35–37] indicate that the HPA axis plays an important role in resolving the HDR, which could be enhanced by exogenous GC administration.

Peripherally generated TNF-α, IL-1β, and IL-6 activate the HPA axis independently at some or all of its levels, and in combination their effects are synergistic [38, 39]. The HPA axis responds in a graded manner to greater intensities of stress with increased production of ACTH and GCs. ACTH is the predominant, but not the only, regulator of GC secretion. Other factors, including angiotensin and vasopressin, also influence adrenal GC secretion [40]. Glucocorticoids are secreted directly into the circulation immediately after their synthesis. Cortisol, the major human GC, circulates bound (95%) to a corticosteroid-binding globulin (CBG) synthesized primarily by the liver, thus providing a large reservoir that is released at sites of inflammation or tissue remodelling [41]. Glucocorticoids exert most of their effects through specific, ubiquitously distributed (3,000–100,000 per cell) intracellular GC receptors (GCR), which are found abundantly in diverse genes encoding cytokines as well as other mediators or cellular regulatory elements of the HDR. The affinity of a given glucocorticoid for its receptor is the major determinant of its biological activity.

GCR activation and translocation have been studied extensively, and the molecular mechanisms involved in these processes were recently reviewed [24, 42]. Inactivated GCRs are anchored to a heat-shock protein (HSP) heterocomplex, which is composed of HSP90, HSP70 and HSP56, that seems to facilitate the response of the GCR to GCs [43]. After steroid-binding, the HSP heterocomplex facilitates the transport of the GCR to the nucleus and dissociates from the receptor, thereby "exposing" the DNA-binding site. The hormone-activated receptor acts as a transcription factor by binding to specific DNA sequences, termed "GC response elements". Binding of the receptor to the GC response elements modulates the rate of transcription of specific mRNAs, which in turn affects the levels of encoded protein products [24]. The GC-GCR complex is subsequently dissociated from the host cell DNA and is "inactivated" by reassociation with the HSP heterocomplex.

Several cytokines produce their cellular effects by activating transcription factors (AP-1 and NF-κB), which activate or repress target genes that are regulated in an opposing manner by GCR [42]. Activated GCRs inhibit the transcription of several cytokines (IL-1, IL-2, IL-6, TNF, interferon-γ (IFN-γ) and others) by binding to transcription factors (AP-l and NF-κB) and blocking their cellular action [42, 44]. Glucocorticoids also suppress the synthesis of phospholipase A_2 (PLA$_2$), cyclo-oxygenase 2 and nitric oxide synthase 1 genes, decreasing the production of prostanoids (PLA$_2$ is the rate-limiting enzyme in eicosanoid metabolism), platelet-activating factor and nitric oxide, three key molecules in the inflammatory pathway [42, 45]. Glucocorticoids induce lipocortin production [45] and have an inhibitory effect on the expression of adhesion molecules [42]. Glucocorticoids also act in synergy with IL-1RA [47] and IL-4 [29] to control the HDR.

The host defence response in ARDS

ARDS is characterized by acute onset of diffuse and severe HDR of the lung parenchyma to a direct or indirect insult that disrupts the alveolocapillary membrane with loss of compartmentalization [48]. The magnitude of the initial HDR appears to be a major determinant of the progression and outcome in ARDS. Our group recently reported that on Day 1 of mechanical ventilation (MV), nonsurvivors of ARDS had significantly (p< 0.0001) higher plasma (fig. 1) and bronchoalveolar lavage (BAL) TNF-α, IL-1β, IL-2, IL-4, IL-6 and IL-8 levels than survivors [49, 50]. Although cytokine levels may not reflect activity, these findings are similar to those reported by others (table 4) and agree with studies indicating that the evolution of ARDS is determined by the extent of initial pulmonary HDR in the form of alveolar denudation, basement membrane destruction, vascular permeability and quantity of intra-alveolar exudate [74–77].
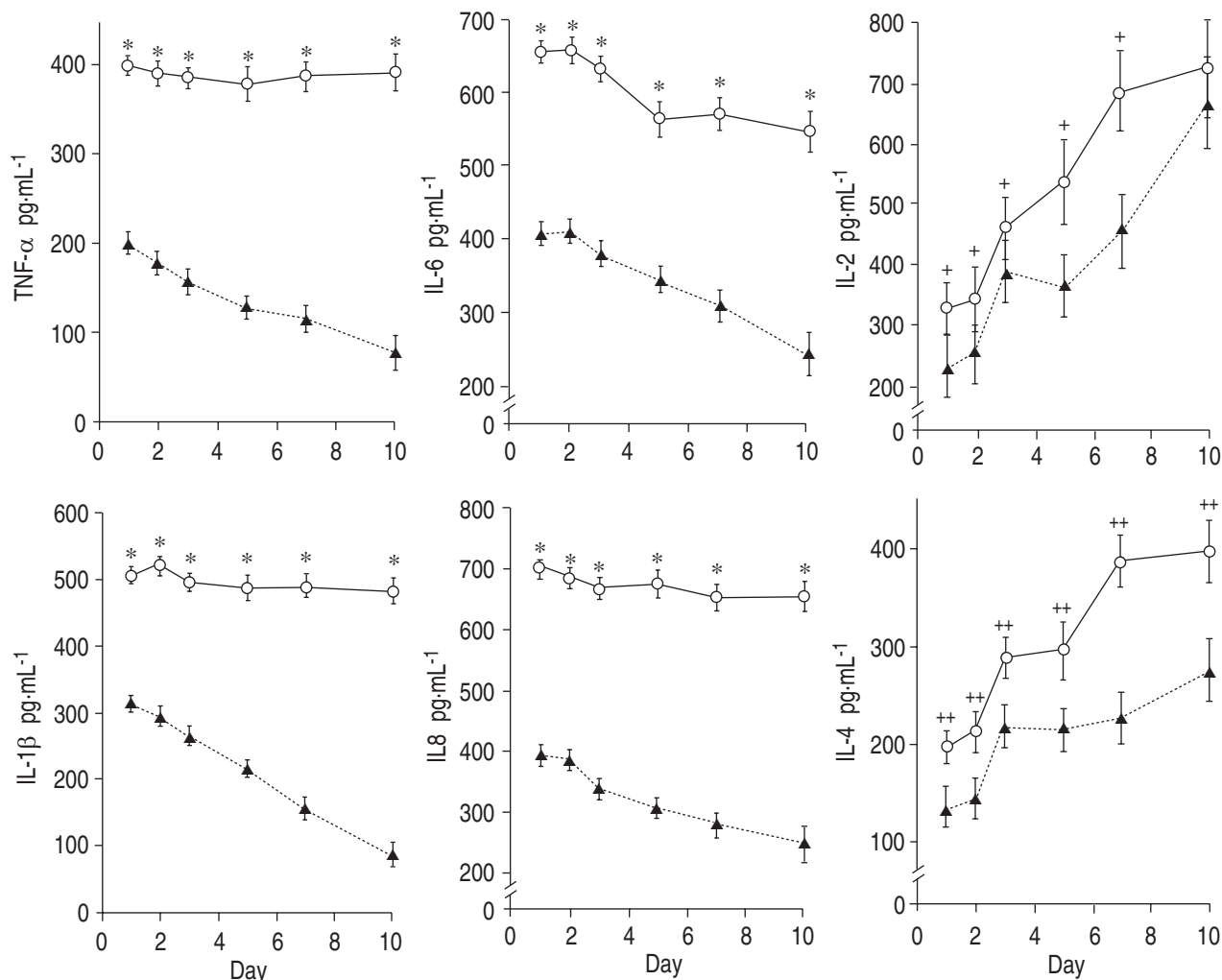
Fig. 1. – Plasma TNF-α, IL-1β, IL-6, IL-8, IL-2 and IL-4 levels over time in survivors and nonsurvivors of ARDS. ○: nonsurvivors. ▲: survivors; *, +, ++: p<0.05, p<0.003, p<0.009 vs survivors. Reproduced with permission from reference [49].

Table 4. – Studies showing a relationship between magnitude and duration of host defence response as measured by circulatory or pulmonary inflammmatory cytokine levels and outcome

Higher initial elevation in nonsurvivors		
Sepsis	TNF-α	[51, 56]
	sTNFR	[57]
	IL-1β	[56, 58]
	IL-6	[51, 56, 58–64]
	IL-8	[65]
ARDS	TNF-α	[49, 50*, 66*, 67]
	IL-1β	[49, 50*, 68]
	IL-2	[49]
	IL-4	[49]
	IL-6	[49, 50*, 68]
	IL-8	[49, 50*, 69*, 70*, 71*]
Persistent elevation in nonsurvivors		
Sepsis	TNF-α	[51, 56, 58, 59, 72, 73]
	IL-6	[56, 73]
ARDS	TNF-α	[49, 50*, 67, 68]
	IL-1β	[49, 50*]
	ILK-6	[49, 50*, 68]
	IL-8	[49, 50*]

*: bronchoalveolar lavage. TNF-α: tumour necrosis factor-α; sTNFR: soluble tumour necrosis factor receptor; IL: interleukin; ARDS: acute respiratory distress syndrome.

In agreement with others [66, 78–81], we have found an increased BAL:plasma ratio for all measured cytokines [49, 50]. Increased BAL levels indicate increased intrapulmonary production and not leakage into the lung from increased vascular permeability [79]. In early ARDS, alveolar macrophages have been shown to be an important source of TNF-α [82, 83], IL-1β [84] and IL-8 production [85]. Experimental work indicates that alveolar macrophages and neutrophils represent different temporal and cellular waves of cytokine expression in early lung injury [86]. We have found patients, stratified by the presence or absence of sepsis, with direct and indirect ARDS to have similar plasma and BAL inflammatory cytokines levels [87].

Of significant importance, during the progression of ARDS, we have found nonsurvivors to have persistent and marked elevation of plasma and BAL inflammatory cytokine levels over time, while survivors had a rapid reduction in inflammatory cytokine levels (fig. 1) [49, 50]. Furthermore, in nonsurvivors the BAL:plasma ratio for TNF-α, IL-1B, IL-6 and IL-8 remained unchanged throughout the course of the disease, while it rapidly decreased in survivors [50]. Other groups have also reported nonsurvivors of ARDS or sepsis to have persistent elevation of inflammatory cytokines (table 4) or

other components of the HDR (*i.e.*, PLA_2, leukotrines and complement) over time [51, 67, 88–90]. This finding is important to understanding why treatment modalities of limited duration may be ineffective in ARDS. It is well-accepted that during ARDS the HDR is not limited to the lung [91]. In patients with unresolving ARDS, disruption of alveolocapillary membrane [92] causes release of cytokines into the systemic circulation and contributes to the development and/or maintenance of SIRS and MODS [48, 68]. Strong correlation among TNF-α, IL-1β, IL-6 and IL-8 at the onset of ARDS and over time is consistent with a broad and integrated host defence response [49].

In patients with early ARDS, one study found soluble TNF receptor inhibitor levels to be significantly increased compared to pre-ARDS levels and to correlate (r=0.71; p<0.005) with BAL TNF-α levels [66]. Administering IL-1RA in experimental ARDS decreases vascular permeability and neutrophil influx [93]. At the onset of ARDS, we have found plasma IL-4 to be significantly (p<0.001) higher in nonsurvivors. Plasma and BAL IL-4 levels increased over time, irrespective of outcome. We have found plasma IL-10 levels to be significantly higher in the early phase of ARDS in patients who improve lung function by Day 7 of MV (personal unpublished data). Others have found increased BAL IL-10 levels to correlate with survival [94].

Overall a strong line of evidence supports the view that an overaggressive and protracted HDR, rather than the aetiological condition precipitating respiratory failure, is the major factor influencing outcome in ARDS. In agreement with this statement are the findings that ARDS patients may not improve despite appropriate treatment of the precipitating disease, while mortality may decrease when the HDR is adequately suppressed by prolonged GC administration [95]. In ARDS, continued production of HDR mediators prevents effective restoration of lung anatomy and function by sustaining ongoing injury, coagulation and fibroproliferation (the three act in synergy). New injury to previously spared endothelial and epithelial surfaces occurs in conjunction with an amplified reparative (coagulation and fibroproliferation with deposition of ECM) process over previously damaged areas. At histology, these two processes can be seen adjacent to each other [1] and have been described in detail [92]. Persistent endothelial and epithelial injury protracts vascular permeability in the lung and systemically. Intravascular coagulation decreases available pulmonary vascular bed, while intra-alveolar fibrin deposition promotes cell-matrix organization by fibroproliferation [96]. Most clinical and physiological derangements observed in unresolving ARDS are attributable to unrestrained coagulation and fibroproliferation and are discussed below.

Coagulation

During ARDS, normal intra-alveolar fibrinolytic activity (urokinase-like plasminogen activators produced by alveolar macrophages) and endothelium anticlotting activity (heparin-like molecules, thrombomodulin) are severely compromised and lead to accelerated vascular and extravascular fibrin deposition [97–99]. Megakaryocytes are markedly increased in the lungs of ARDS patients [100]. In patients with ARDS, BAL has increased procoagulant activity due to tissue factor associated with factor VII [101] and concomitant depression of fibrinolytic activity attributable to increased levels of both plasminogen activator inhibitor-1 (PAI-1) and antiplasmin [101–103]. In humans [101, 104], derangements of intra-alveolar fibrin turnover occur early (1–3 days) and persist for ≥14 days. Depressed BAL fibrinolytic activity at day 7 of ARDS correlates with poor outcome [101]. Despite a depression in fibrinolytic activity, BAL fibrinogen degradation products (FDP) are markedly increased and remain elevated over time and correlate significantly with BAL total protein concentration and number of neutrophils [104]. Patients with ARDS also have marked and prolonged systemic procoagulant activity and rapid exhaustion of the fibrinolytic system [105]. In sepsis, high levels of PAI-1 are associated with a poor prognosis [106] and correlate with TNF, IL-6 and complement 3a levels [51]. Virtually all ARDS patients have increased circulating FDP levels [107]. Disseminated intravascular coagulation (DIC) is a frequent finding in ARDS and carries a higher mortality rate. Even in patients without DIC, a reduction in circulating platelets of at least 50% of the initial values is frequently observed during the course of ARDS, and nonsurvivors have a greater degree of thrombocytopenia than survivors [108]. Thrombocytopenia does not result from decreased platelet production, but from decreased platelet survival (one third of normal), a result of increased pulmonary sequestration [109].

Patients with early ARDS, as well as patients with late unresolving ARDS, show diffuse pulmonary sequestration of intravenously administered radiolabelled fibrinogen [110]. Pathways for increased fibrinogen uptake include: 1) increased microvascular permeability with exudation of fibrinogen into pulmonary interstitial and intra-alveolar oedema; 2) intravascular and extravascular fibrin formation; and 3) fibrinogen binding to injured endothelial cells [110]. Recovery from lung injury is associated with normalization of radiolabelled fibrinogen uptake [110]. As a result of downregulation of thrombomodulin and the expression of tissue factor, cytokines TNF-α, IL-1β, and IL-6 alter the surface of the endothelium from an anticoagulant into a procoagulant moiety and are able to enhance the synthesis of PAI-1 [106, 111].

Pulmonary thromboemboli are a frequent histological finding in patients with unresolving ARDS subjected to open-lung biopsy [92] and are found on postmortem exam in 95% of ARDS nonsurvivors [112, 113]. Macrothrombi (in arteries greater than 1 mm diameter) are found by postmortem angiography in 86% of patients and are more prevalent in patients who died in the early phase of ARDS [113]. Microthrombi are as prevalent as macrothombi but tend to be distributed throughout all phases of ARDS. Filling defects at angiography correlate with the severity of ARDS, the degree of pulmonary hypertension and the presence of DIC [112]. Unfortunately, anticoagulant treatment does not improve outcome in ARDS [112] and this has applied to any treatment directed at one single facet of the complex host response. Ischaemic or avascular necrosis, particularly in the subpleural regions, is a common feature of unresolving ARDS [92, 112].

Fibrin deposition also influences the course of tissue injury and repair. Thrombin, fibrin and fibrin degradation products (FDPs) play an important role in amplifying inflammation by promoting neutrophil chemotaxis, adhesiveness and by directly causing increased endothelial permeability [99, 114, 115]. Experimental infusion of thrombin or FDP causes acute lung injury, mediated by neutrophil activation *via* complement and arachidonic acid metabolites [99]. Thrombin is also a potent inducer of platelet degranulation with additional release of host defence response mediators. Coagulation and fibrinolysis also interact with the kallikrein-kinin system with release of bradykinin. Bradykinin increases vascular permeability and stimulates collagen production by fibroblasts [116].

Animal studies have demonstrated that intra-alveolar fibrin deposition is typical of DAD, even when injury resolves without fibrosis, indicating that fibrin deposition of limited duration is essential for effective lung repair [117]. In the presence of a protracted host defence response, however, persistent intra-alveolar fibrin deposition contributes to airspace organization and fibrosis [97, 117, 118]. *In vitro* data demonstrate that fibrin forms a matrix on which fibroblasts may aggregate and secrete collagen [119]. In addition, thrombin binds to thrombin receptors on fibroblasts and promotes their proliferation [120].

Fibroproliferation

Fibroproliferation is a stereotypical reparative response to injury. In ARDS, pulmonary fibroproliferation manifests with the accumulation of myofibroblasts and their connective tissue products in the airspaces, interstitium, respiratory bronchioles and walls of the intra-acinar microvessels [113, 121]. Pulmonary fibroproliferation is a diffuse process, as indicated by the findings of chest computed tomography [92], gross inspection at surgery, microscopic analysis of biopsies from different lobes [122] and bilateral BAL findings [50]. At microscopy, however, regional heterogeneity exists, and focal areas of normal parenchyma are occasionally found [1, 123]. Unhalted fibroproliferation results in extensive fibrotic remodelling of the lung parenchyma. Macroscopically, the lung shows irregular zones of diffuse scarring with formation of numerous microcystic reorganized airspaces measuring 1–2 mm, most prominent in the subpleural zones [92].

Pulmonary fibroproliferation in ARDS shares a common pathogenetic mechanism with other fibroproliferative diseases, where degree and duration of the host defence response dictate the ultimate reparative outcome [28]. In this "linear" concept of tissue response to injury, mediators of the host defence response sustain the fibrotic process [124]. Fibrosis ensues when the host defence response is intense and prolonged, leading to released profibrotic moieties and trophic factors for mesenchymal cells. Experimental work indicates that severity of acute lung injury determines the intensity of chronic inflammation and fibrosis [125]. In agreement, we have found that patients with higher plasma IL-6 on days 1–3 of ARDS are more likely to develop accelerated fibroproliferation unresponsive to GC rescue treatment (fig. 2) [95].

Morphometric analysis of lung tissue in late ARDS has shown that intra-alveolar fibroproliferation predominates over interstitial fibroproliferation [121]. The histological sequence leading to intra-alveolar fibroproliferation has been clearly characterized [121]. Epithelial injury provides focal discontinuities (gaps) in the alveolar basement membrane (BM) resulting in direct communication of interstitial cells and matrix elements with the alveolar airspaces [92, 126]. Activated myofibroblasts from the interstitium migrate into the alveoli in response to chemotactic signals and attach to the luminal surface of the damaged BM [121, 126]. Myofibroblasts, once they have migrated into the alveoli, proliferate and actively produce collagen [126], transforming the initially fibrinous intra-alveolar exudate into myxoid connective tissue matrix and eventually into dense acellular fibrous tissue. TNF-α and IL-1β, among other HDR mediators, stimulate chemotaxis and are important modulators of fibroblast proliferation and collagen deposition [127].

Morphometric studies in nonsurvivors of ARDS have shown intra-alveolar (and, to a lesser degree, interstitial) fibroproliferation to occur within 7 days of the onset of ARDS and to have a rapid increase in the second and third week of respiratory failure [121]. The rate of progression varies [1, 3, 8]. Newly produced matrix stains intensely for cell-associated fibronectin and type 3 procollagen [126]. Type III collagen (newly formed, flexible, and more susceptible to digestion by collagenase) predominates in the intermediate proliferative phase, while type I collagen (composed of thick fibrils, more resistant to digestion) is the major collagen present in the late fibrotic phase. In nonsurvivors, the collagen content of the lung is increased two- to threefold after 2 weeks of ARDS and parallels the development of fibrosis [7]. Patients with ARDS dying after 7 days of respiratory failure have, in contrast to survivors, persistent elevation of extracellular matrix components (procollagen III) in the BAL and serum, indicating ongoing fibrogenesis [77, 128–130]. Patients with a procollagen III level greater than 1.75 $U·mL^{-1}$ in BAL on day 7 had a 72% mortality rate compared with a 20% mortality rate in patients with a procollagen III level less than 1.75 $U·mL^{-1}$ [131]. BAL procollagen III levels correlate with histological evidence of intra-alveolar fibrosis [132]. Finding fibrosis at open-lung biopsy or transbronchial biopsy is a poor prognostic factor [4, 133].

The patterns of fibrous reorganization of the lung in ARDS have been described [74]. Intra-alveolar fibroblastic aggregation is continuous with a similar process in the terminal bronchioles, respiratory bronchioles, alveolar ducts, and the interstitium [8]. The bronchiolar intraluminal (bud-like) fibrosis found in patients with fibroproliferation [3] is similar to the one described in interstitial lung disorders. The coexistence of unaffected bronchioles surrounded by parenchymal interstitial and alveolar space fibrosis indicates lung injury-induced patchy bronchial epithelial and basement membrane damage. Intraluminal fibrosis as a model of inflammatory lung disease has been extensively reviewed [96]. In our experience, intraluminal fibrosis at open-lung biopsy in patients with unresolving ARDS predicts reversibility of fibroproliferation with GC rescue treatment [3], similar to the response observed in patients with bronchiolitis obliterans organizing pneumonia [96].
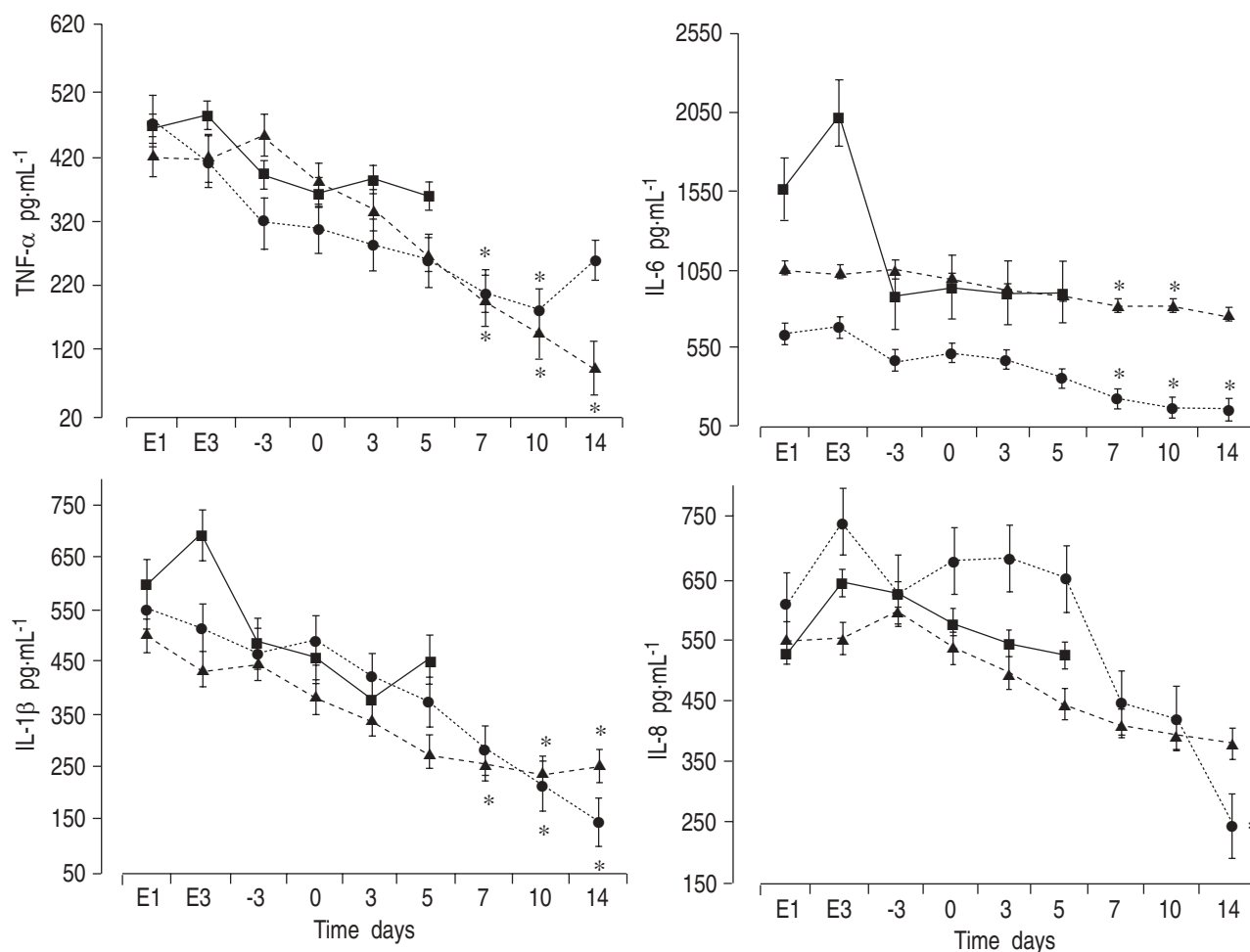
Fig. 2. – Mean changes (±SE) in plasma TNF-α, IL-1β, IL-6, and IL-8 levels before and during treatment with glucocorticoids in patients with rapid, delayed, and absent physiologic response. ⋯●⋯: rapid responders (n=5); ⋯▲⋯: delayed responders (n=2); ━■━: nonresponders (n=2); E1, E3: day 1 and 3 of ARDS. Treatment started at time 0. The mean (±SE) duration of mechanical ventilation prior to initiation of glucocorticoid treatment was 12±5 days in rapid responders, 32±8 days in delayed responders and 9±8 days in nonresponders. *: $p < 0.05$ vs start of treatment. Reproduced with permission from reference [95].

The pulmonary vascular changes occurring during the progression of ARDS are shown in table 5. Of significance, we and others have described microscopic and ultrastructural evidence of ongoing epithelial and endothelial injury in patients with advanced fibroproliferation [1, 3, 92, 113]. Despite the high turnover rate and ability of endothelial cells to repair themselves, acute endothelial injury is more pronounced in the proliferative phase than in the exudative phase of ARDS [1, 113], consistent with a continuous injury process. Fibrocellular intimal proliferation, the sequela of endothelial injury, involves predominantly the small arteries, but also the veins and lymphatics. Absence of arteriolar subintimal fibroproliferation at open-lung biopsy in patients with unresolving ARDS predicts reversibility of fibroproliferation with GC rescue treatment [3].

Clinical and physiological manifestations of the host defence response in unresolving ARDS

Morphological changes at epithelial and endothelial levels caused by recurrent injury, ongoing coagulation,

and amplified fibroproliferation can explain the physiological and laboratory findings seen in patients with unresolving ARDS.

Gas exchange and lung mechanics

Gaps in the alveolar BM in unresolving ARDS [92] allow communication between the alveoli and the interstitium for entrance of vascular and interstitial components into the airspaces (fig. 3). Myofibroblasts migrate, proliferate, and produce collagen. Progressive fibroproliferation leads to obliteration of the respiratory units, changing their mechanical properties (loss of inflection point in the pressure-volume (P-V) curve and lack of recruitability by positive end-expiratory pressure (PEEP)), increasing dead space ventilation (VD/VT) and further compromising of gas exchange [135–137]. A concomitant reduction in capillary volume and thickening of the alveolar septa additionally contributes to reducing gas transfer and increasing VD/VT [1]. In several studies, the arterial oxygen tension (Pa,O$_2$): inspiratory oxygen fraction (FI,O$_2$) ratio, although similar at the onset of

Table 5. – Pulmonary vascular changes in unresolving ARDS

	Early	Intermediate	Late
Mean PAP	36±7.3	42±9.4	42.4±3.8
Vasoconstrictor component	++(a)	-	-
Acinar*			
Acute endothelial injury	+	++	++
PMN aggregates in capillaries	+++(b)	+	-
Hyaline microthrombi[#]	+++	-	-
Organizing microthrombi[†]	+(c)	++	++
Increased medial wall thickness	+	++	++
Compression by oedema	+	-	-
Compression by fibrosis	-	++	+++
Capillary proliferation	-	++	+++
Architecture	Normal	Up to 50% reduction (c)	More than 50% reduction(c)
Preacinar			
Macrothrombi	+++	++	+
Infarction (subpleural)	-	++	+
Intimal fibroproliferation	-	++	+++
Increased medial wall thickness	+	++	+
Decreased external diameter	+	++	+++
Decreased arterial filling	+(d)	++	+++
Remodelling with tortuosity	-	++	+++
Lymphatics obstruction	+	++	++

(a): from reference [134]; (b): more prominent in ARDS induced by septicaemia; (c): from reference [1]; (d): narrowing by interstitial oedema; *: intra-acinar landmarks: terminal and respiratory bronchioles, alveolar ducts; [#]: hyaline thrombi, consisting of platelets and fibrin; [†]: organizing microthrombi, formed of red and white blood cells with layered fibrin. +: present; ++: marked; +++: very marked. Reproduced with permission from reference [10]. Above data are modified from reference [113].

ARDS, by day 3–7 clearly separated survivors (increased) from nonsurvivors (decreased or no change) [50, 108, 138–145]. In one report, mortality rate in patients with and without improvement in lung function by day 7 of ARDS was 43 and 97%, respectively [141].

Pulmonary vascular permeability

Endothelial injury [92] in unresolving ARDS favours the passage of vascular products in the alveoli (fig. 4).

Among the many cells and substances that are exuded in the airspaces, the following have been the subject of clinical investigations: albumin, proteins, neutrophils and [67]Ga.

BAL albumin and total protein are markers of pulmonary endothelial permeability in ARDS [146]. At the onset of ARDS, we have found BAL albumin and total protein levels to be similar in survivors and nonsurvivors. In agreement with others [144, 147], we noted survivors to have a progressive decline in BAL albumin and total protein levels over time (not seen in nonsurvivors),
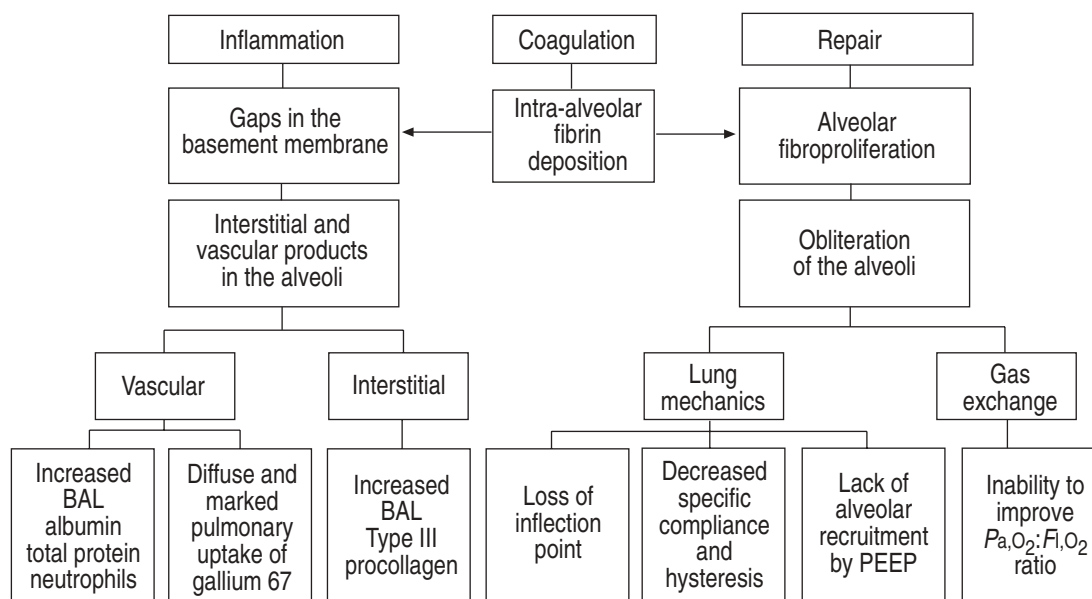

Fig. 3. – Epithelial changes in unresolving acute respiratory distress syndrome (ARDS). BAL: bronchoalveolar lavage; PEEP: positive end-expiratory pressure; P_{a,O_2}: arterial oxygen tension; F_{I,O_2}: inspiratory oxygen fraction.
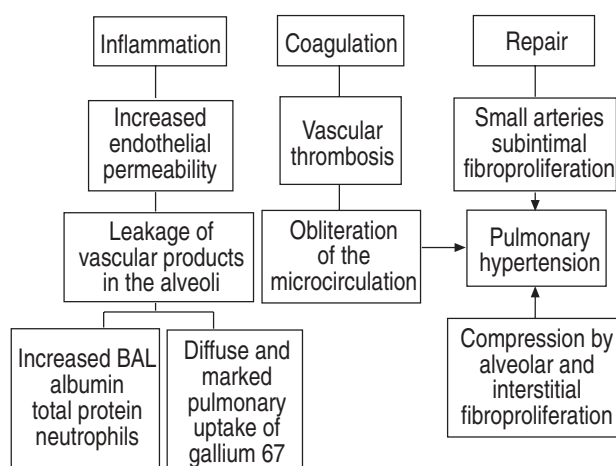
Inflammation	Coagulation	Repair
Increased endothelial permeability	Vascular thrombosis	Small arteries subintimal fibroproliferation
Leakage of vascular products in the alveoli	Obliteration of the microcirculation	Pulmonary hypertension
Increased BAL albumin total protein neutrophils / Diffuse and marked pulmonary uptake of gallium 67		Compression by alveolar and interstitial fibroproliferation

Fig. 4. – Endothelial changes in unresolving adult respiratory distress syndrome.

suggesting effective repair of the alveolar-endothelial surface [50]. We have also identified a consistent correlation between BAL albumin and total protein levels and BAL TNF-α, IL-1β, IL-6 and IL-8 levels [50].

BAL neutrophilia ($\geq$70%) is invariably found in early ARDS [147, 148]. The percentage of BAL neutrophils parallels BAL protein [75, 144] and albumin [50] concentration and correlates with the severity in gas exchange [147]. We and others have identified a positive correlation between total neutrophil count and BAL TNF-α, IL-1β and IL-8 levels [50, 69, 149] . Persistent elevation of neutrophils in the BAL by days 7 and 14 of ARDS is associated with poor outcome [150, 151]. Resolution of ARDS, on the contrary, is associated with a dramatic fall in neutrophils and an increased number of macrophages [150].

The origin of neutrophils recovered by BAL in unresolving ARDS requires a clarification. In patients with unresolving ARDS subjected both to bronchoscopy and open-lung biopsy, at an average of 14 days into respiratory failure, we have found a significant discrepancy between marked BAL neutrophilia (60% of recovered cells) and an almost complete absence of neutrophils in the airways, alveoli, and interstitium at histology [3]. It has been recognized that the alveolocapillary membrane is permeable to fluids and that in normal patients 39% of the aspirated BAL fluid originates from the circulation [152]. In conditions associated with disruption of vascular and epithelial integrity, such as unresolving ARDS, the circulatory component of BAL effluent becomes more significant, and cells such as neutrophils and erythrocytes are aspirated into the alveoli when negative pressure is applied during suction [152]. Therefore, BAL neutrophilia in unresolving ARDS is a marker of endothelial permeability [3, 75, 144], not a reflection of "neutrophilic alveolitis," and it is unlikely that neutrophils play a major role in the progression of unresolving ARDS.

^{67}Ga uptake in the lung is an additional marker of endothelial permeability [153]. In the lung, ^{67}Ga concentration is normally low. Diffuse ^{67}Ga uptake has been used as a sensitive but nonspecific test to identify patients with active pulmonary inflammation who may respond to GC treatment. Five reports (total of 44 patients) have described marked and diffuse uptake of ^{67}Ga in the lung of patients with unresolving ARDS and who subsequently responded to GC rescue treatment [3, 32–24, 154]. Diffuse and marked pulmonary uptake of ^{67}Ga in unresolving ARDS correlates with BAL neutrophilia [153]. The diagnostic role of ^{67}Ga scintigraphy in ARDS was reported [153].

Pulmonary hypertension

While in early ARDS the pulmonary vasculature is available for dilation and/or recruitment, vascular changes (table 5) in unresolving ARDS are less responsive to vasodilator treatment [134]. In these patients, vascular obstruction from thrombosis, subintimal fibroproliferation of the small arteries and compression from alveolar and interstitial expansion significantly reduces the capacity of the pulmonary microcirculation [1] and subjects the patent vascular bed to abnormally high flow, stimulating medial hypertrophy of the muscular and partially muscular arteries [155]. These changes are similar to those seen in pulmonary hypertension of thromboembolic origin [155], and they worsen with progression of fibroproliferation [108, 140, 143]. In early ARDS, there is a marked elevation in circulating levels of endothelin-1, a mediator of vascular remodelling and pulmonary hypertension [52]. With the progression of ARDS, endothelin-1 levels drop to normal values in patients improving lung function but remain elevated in patients who worsen [52]. Persistent elevation or worsening pulmonary artery pressure is a poor prognostic sign [108, 140, 143]. In the final stages of ARDS, the mean pulmonary artery pressure can exceed 40 mmHg as a result of a fourfold elevation of vascular resistance [134].

Barotrauma

In unresolving ARDS, vascular obstruction with subpleural tissue necrosis and endoluminal fibrosis with intraparenchymal pseudocyst formation contribute to alveolar rupture. Barotrauma may manifest as intraparenchymal and subpleural pneumatoceles, large compliant air collections or bullae, pulmonary interstitial emphysema, pneumomediastinum, pneumothorax, subcutaneous emphysema and, rarely, pneumoperitoneum [156]. Barotrauma in ARDS is related to the severity of lung dysfunction and is associated with a higher mortality [157–159]. An increase in the alveolar-arterial pressure gradient, from either hyperinflation and/or reduced blood flow, causes disruption at the common border between the alveolar base and the vascular sheath [155, 160]. Partial obliteration (endoluminal fibrosis) of terminal airways leads to cyst formation by valvular mechanism and by compensatory dilatation of neighbouring bronchioles [159]. Tissue necrosis distal to pulmonary artery thrombi [112] is prominent in the subpleural regions where insufficient collateral blood flow makes lung tissue particularly susceptible to ischaemia [161]. At angiography, a "picket-fence" appearance is seen, caused by dilated subpleural arteries bridging regions of oligaemia and necrosis distal to the thrombotic arterial occlusion [161]. With

progressive fibroproliferation, preferential ventilation to these hypoperfused peripheral areas may occur, contributing to the development of barotrauma [161].

Fever of infectious and noninfectious origin

Fever is caused by the systemic release of endogenous pyrogens: TNF-α, IL-1β and IL-6. Upon reaching the hypothalamic thermoregulatory centre, these cytokines induce an abrupt release of prostaglandins that increase the thermostatic set point and produce heat [162]. Fever induces production of heat-shock proteins (see later) that have an important role in cell survival under stress (cytoprotection) [163]. Fever and SIRS invariably develop during the course of unresolving ARDS, even in the absence of infections [14, 34, 164]. Clinical studies have attributed the frequent occurrence of fever and SIRS in late ARDS to VAP [15], and the assumption has been made that in these patients, nosocomial infections (mainly VAP) amplify SIRS and lead to MODS and death [5, 6, 15, 16].

Recognizing that clinical criteria are sensitive but nonspecific in diagnosing VAP or other nosocomial infections has promoted clinical investigations following recently developed guidelines [21]. Three important findings have resulted: 1) during the course of ARDS:VAP is less frequent than clinically suspected [165, 166]; 2) pulmonary host defence response itself is a frequent source of fever, clinically indistinguishable from VAP or other sources of sepsis [2, 14, 34]; and 3) during the course of ARDS, VAP or other nosocomial infections do not amplify SIRS [164]. In a prospective epidemiological study using strict diagnostic criteria, VAP was identified with protected bronchoscopic techniques in only 20% of patients with ARDS on MV for more than 48 hr (the study excluded patients with pneumonia-causing ARDS) [165]. The pathogenesis of VAP in ARDS has been reviewed [167, 168]. In a prospective study designed to identify by strict criteria all potential infectious and noninfectious sources of fever (single episode) in 20 patients with late ARDS (>48 h), we identified pneumonia in seven (35%) and noninfectious pulmonary HDR alone in five (25%) [169]. The latter diagnosis was established by open-lung biopsy in febrile patients without infection detected by an extensive and systematic evaluation [169]. Lung histology showed advanced fibroproliferation, acute epithelial and endothelial injury, vascular thrombosis, but no pneumonia.

While at autopsy histological pneumonia is most often disseminated to both lungs [170], during the course of ARDS pneumonia is frequently detectable by bronchoscopy in one lung only [171]. Among 23 patients with late ARDS and fever who were undergoing bilateral BAL, we found pneumonia to be unilateral in 47% [171]. These findings were confirmed in a larger series involving more than 100 patients (personal unpublished data). Moreover, we found that pneumonia with a low concentration of bacteria ($\leq 10^5$ colony forming units (CFU)·mL^{-1}) was usually unilateral, whereas pneumonia with a high bacterial load ($\geq 10^6$ CFU·mL^{-1}) was frequently bilateral (p=0.002) and associated with a worse short-term outcome (10 day mortality was 50%) [171]. Histological pneumonia at autopsy, therefore, may be more representative of a severe pneumonia that led to the patient's demise or of premortem dissemination of the infection in the terminal stage of the disease [21]. In agreement with prior work [172], we have found radiographic findings for pneumonia to be nonspecific in ARDS [173].

Correlation between nosocomial infections and cytokine response

In a study involving serial measurements of plasma and BAL inflammatory cytokines and careful search and diagnosis of infections, we found that nosocomial infections (including seven cases of pneumonia) developing in 34 patients with ARDS, who had been on MV for >72 h caused neither a transient nor a sustained increase in plasma and BAL inflammatory cytokines levels (TNF-α, IL-1β, IL-6 and IL-8) or SIRS score [164]. These findings are similar to those reported by others [174] and question the prior hypothesis, based on clinical criteria, that in unresolving ARDS, nosocomial infections amplify SIRS leading to MODS and death [5, 6, 15, 16]. Furthermore, two recent studies indicate that the development of MODS and outcome in surgical critical illness are related to the magnitude of SIRS and are independent of the presence of infection [175, 176].

In agreement with our findings, several recent experimental and clinical studies have shown that production of inflammatory cytokines in response to a bacterial challenge can be suppressed by prior activation of the HDR. Although, this phenomenon serves to protect the host (cytoprotection) from overwhelming cytokinaemia, an almost complete inhibition of inflammatory cytokine release may potentially cause immunodeficiency [177]. Two processes with a potentially common mechanism have been investigated: downregulation and induction of stress tolerance [178]. In agreement with experimental work [179, 180], the monocytes of patients with sepsis and ARDS show downregulation of inflammatory cytokine release in response to lipopolysaccharide (LPS) [177, 181]. Furthermore, stimulated monocytes from nonsurviving septic patients showed a significant reduction in IL-1 production when compared to survivors [182]. Pot-ential mediators of inflammatory cytokine downregulation include prostaglandin E$_2$, glucocorticoids, IL-4, IL-10, TGF-β [177] and induction of HSPs [178].

In experimental models of lung injury and sepsis, induction of HSPs prior to an otherwise lethal stress, protected animals (stress tolerance) during the time course of HSP elevation with reduced organ damage and improved survival [163, 178]. Cytokines, TNF, IL-1, IL-2, and IL-6 can induce the synthesis of HSPs in a dose-dependent manner [183]. In a negative feedback, HSPs can downregulate LPS-induced monocyte production of TNF and IL-1 [184, 185], while other macrophage functions (*i.e.* phagocytosis) remain intact [185]. Although the precise mechanism of action is not known, ample evidence indicates that many HSPs act as chaperones and assist in the process of protein folding and translocation across membranes [186]. Furthermore, HSPs are directly responsible for binding TNF intracellularly and preventing its release from the macrophage [187]. The importance of HSPs in the host defence response and their intimate association with activation of the HPA axis and sympathetic nervous system have recently been appreciated [24].

Expression of HSPs is regulated primarily through the activity of transcription factor HSFs [24]. Experimental work has shown that HSP70 levels are increased in the adrenal gland and in the vasculature of stressed animals [24]. Adrenal expression is mediated *via* ACTH, while vasculature response is subject to α_1-adrenergic control [188]. Long-term glucocorticoid treatment significantly decreases adrenal HSP70 expression [189]. Of interest, experimental work indicates that GC may reinstate cell responsiveness and offset downregulation of inflammatory cytokine production [190, 191].

Systemic effects of the host defence response

Figure 5 displays the local and systemic effects of a protracted HDR in ARDS. Systemic release of HDR mediators causes: 1) SIRS; 2) development and progression of MODS (by a mechanism similar to the one causing progression of ARDS); and 3) increased systemic endothelial permeability. Fever is a common clinical denominator to noninfectious SIRS and nosocomial infections.

Relationship between exaggerated host defence response and endogenous glucocorticoid function

The pathophysiology of septic shock is similar to that of ARDS, and reference to this literature is frequently necessary to explain the complex relationship between exaggerated HDR and glucocorticoid function in unresolving ARDS. Although glucocorticoids are the most important modulator of the HDR, nonsurvivors of septic shock have an exaggerated and protracted release of inflammatory cytokines (table 4) despite a marked elevation in baseline plasma ACTH and cortisol levels [192]. Furthermore, the degree of cortisolaemia frequently correlates with severity of illness and mortality rate [193, 194] and is associated with an altered response of the HPA axis to suppression by dexamethasone and stimulation by corticotrophin-releasing hormone (CRH)

and ACTH [194]. Cortisol elevation is achieved by several mechanisms including: 1) activation of the HPA axis; 2) GC resistance resulting from alterations in GCR binding; 3) failure of pituitary and hypothalamus glucocorticoid negative feedback; 4) decreased binding to CBG [195–197]; and 5) decreased cortisol extraction from the blood [192, 193].

In critically ill patients, dexamethasone infusion, at a dose that easily suppresses ACTH and cortisol secretions in normal subjects, is minimally effective [194, 195], with pre- and postinfusion cortisol and ACTH levels remaining significantly higher in nonsurvivors [194]. In addition, the negative feedback of endogenous glucocorticoid is clearly altered, and stimulation with CRH causes a significant increase (7–450%) in ACTH response, in spite of cortisol levels sufficient to abolish a CRH-induced ACTH surge in normal subjects [194]. Following CRH stimulation, nonsurvivors have a smaller increase in cortisol levels, despite a percentage increase in ACTH production similar to survivors [194]. Other studies have also found a lower cortisol response to ACTH in nonsurvivors of septic shock [36, 198, 199].

Although several potential mechanisms have been described [40], glucocorticoid resistance from altered GCR function may be partially responsible for the changes in the HPA axis of critically ill patients and could result in an augmented ACTH response to CRH [194]. It is now recognized that cytokines can cause a concentration-dependent resistance to glucocorticoids by reducing GCR binding affinity [200, 201]. Many cytokines produce their cellular effects by activating transcription factors, AP-1 and NF-κB [42]. For example, TNF-α activates both AP-1 and NF-κB in human lung [202]. These activated transcription factors may then form complexes with activated GCR, preventing GCR interaction with DNA [42]. When T-cells are incubated with either the combination of IL-2 and IL-4 [201], or IL-1, IL-6 and IFN-γ [200], GC resistance is induced in a concentration-dependent fashion. Resistance results from a marked reduction in GCR binding affinity that is reversed by removal of cytokines [201]. In our patients with
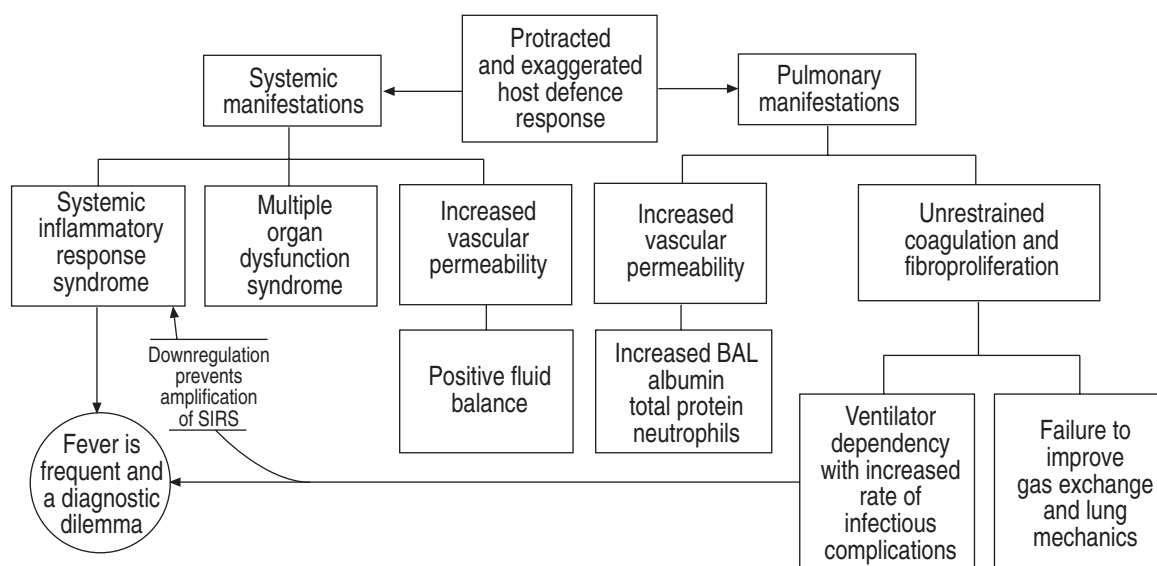

Fig. 5. – Pathophysiology of unresolving adult respiratory distress syndrome. BAL: bronchoalveolar lavage; SIRS: systemic inflammatory response syndrome.

ARDS, we have found significantly higher (p<0.001) plasma levels of IL-2 and IL-4 (fig. 2) as well as plasma (fig. 1) and BAL IL-1β, IL-6 and TNF-α (p<0.0001) in nonsurvivors than in survivors [49]. This line of evidence is reinforced by the results of recent experimental studies [131, 203, 204]. In a sheep model of sepsis-induced ARDS, maximal binding capacity of GCR decreased continuously after endotoxin infusion, despite marked elevation in cortisol levels [203]. Furthermore, reduced GCR binding correlated negatively (r=-0.87; p<0.01) with PLA_2 activity. In a rat model of septic shock, GCR blockade exacerbated the pathological changes induced by endotoxaemia, especially in the lung and small intestine [204]. PLA_2 activity in rats with 80% GCR blockade was more marked than in those with 50% GCR blockade [204]. Monocytes of patients with sepsis have decreased GCR affinity [131]. TNF-α and IFN-γ also block, in a concentration-dependent manner, the stimulatory effects of CRH and ACTH on the pituitary and adrenal cortex, respectively [205, 206]. Any reduction in GC responsiveness, therefore, would be greater as the intensity of the HDR increases. The ACTH and cortisol profile observed over time in critically ill patients [40] can be reproduced in cancer patients by daily administrtaion of recombinant IL-6 [207]. Syndromes of glucocorticoid resistance respond to treatment with exogenous glucocorticoids [44].

Additional factors contributing to hypercortisolaemia include decreased binding to CBG [195–197] and decreased cortisol extraction from the blood [192, 193]. Septic patients have a rapid depletion in CBG with increased free (nonbound) cortisol [195–197]. The mechanism for the high rate of degradation or removal (protein leak?) of CBG is unknown, but it parallels an increase in C-reactive protein levels [197]. In survivors, in contrast to nonsurvivors, CBG and cortisol levels return to normal during recovery [197] and correlate positively with a reduction in PLA_2 levels (r>0.833; p<0.0001) [88]. Furthermore, nonsurvivors of sepsis, in contrast to survivors, also have impaired removal of injected cortisol from the plasma [193].

Response to glucocorticoid therapy in unresolving ARDS

At present, there is no established treatment to halt the progression of ARDS [208]. In theory, however, a treatment aimed at arresting an exaggerated and sustained HDR should promote epithelial and endothelial cell repair, halt fibroproliferation, and decrease extracellular matrix deposition while stimulating its net resorption. Glucocorticoids can potentially fulfil these requirements through a broad range of inhibitory effects on the HDR, including modulation of macrophage and fibroblast activity (previously reviewed in [34]). Although, large randomized clinical trials have clearly shown no benefit when a short course (≤48 h) of high-dose intravenous GC is administered in early sepsis and ARDS [209, 210], no randomized study has yet evaluated prolonged GC treatment in this patient population.

Three groups of investigators have reported a significant improvement in lung function during prolonged GC administration in patients with unresolving ARDS (rescue treatment) [2, 3, 32–34]. We have investigated the effects of *i.v.* methylprednisolone sodium succinate (2–3 mg·kg⁻¹ daily on days 1–14, 1 mg·kg⁻¹ daily on days 15–28, 0.5 mg·kg⁻¹ daily on days 28–35, followed by tapering over 7 days for a total treatment duration of 6 weeks) administered after 15±7 days of MV in 25 patients with unresolving ARDS and without active pulmonary or extrapulmonary nosocomial infection (confirmed in 13 patients by open-lung biopsy) [3]. Most patients had fever (56%), leucocytosis (88%), significant BAL neutrophilia (86%), diffuse alveolar densities on chest radiograph (83%) and diffuse marked uptake of ⁶⁷Ga in both lung fields (100%). A significant physiological improvement, defined as a reduction in lung injury score (LIS) [211] of more than one point or an improvement in Pa,O_2:FI,O_2 ratio of >100, was seen within 7 days of therapy in 15 patients (rapid responders) and 14 days in 7 (delayed responders) and was not seen in 3 (nonresponders) [3]. In rapid responders, the mean LIS decreased from 2.97±0.13 (day treatment was started) to 2.22± 0.14 (day 3) (p=0.0002) and 1.55±0.13 (day 7) (p=0.0001); the Pa,O_2:FI,O_2 increased from 170±17 to 237±18 (day 3) (p=0.008) and 280±18 (day 7) (p= 0.0001); the mean pulmonary artery pressure decreased from 34±3.1 to 24±4 (day 5) (p=0.04) and 14.5±9 (day 14) [3]. Treatment was also associated with resolution of fever, clearing of densities on chest radiograph, normalization of ⁶⁷Ga pulmonary uptake, reduction in BAL neutrophilia (from 60± 6 to 29±12% in 14 days) and albumin and restoration of normal alveolar architecture (documented by follow-up histology in two patients) [3, 95]. The three patients failing to improve LIS died of respiratory disease, while mortality was only 14% in the 22 responders, indicating that reversal of fibroproliferation improves outcome. Surveillance bilateral BAL with quantitative bacterial cultures for early recognition and treatment of VAP was an integral part of our protocol and may have contributed to improved outcome [3]. Earlier extubation decreased the incidence of nosocomial infections [3]. Normalization of surfactant production with corticosteroid treatment of late adult ARDS has been described [212].

In our study, no physiological variables recorded at day 1 of ARDS or at the time of GC administration could predict the type of physiological response or final outcome. Outcome could be predicted only by: 1) open-lung biopsy histological findings; 2) the presence of liver failure; and 3) the type of physiological response. At histology, the presence of preserved alveolar architecture (p=0.045), myxoid cellular type fibrosis (p=0.045), coexistent endoluminal bronchiolar fibrosis (p=0.0045), and lack of arteriolar subintimal fibroproliferation (p= 0.045) separated survivors from nonsurvivors. These histological findings indicate that GC treatment can halt progression of fibroproliferation if given before end-stage fibrosis develops. In models of tissue repair, GC treatment is more effective during the rapid phase of wound healing (approximately 21 days), when collagen biosynthesis and degradation are accelerated [213].

Host defence response with supplemental glucocorticoid administration

We provided further evidence of the important relationship between HDR and ARDS outcome by measuring

plasma and BAL inflammatory cytokine levels during GC treatment in nine patients with unresolving ARDS (day 15±9 of MV) [95] and having physiological parameters and (plasma and BAL) inflammatory cytokine levels identical to the previously reported group of ARDS nonsurvivors [49, 50]. During GC treatment, effective reduction in inflammatory cytokine levels separated survivors from nonsurvivors [95]. Significant reductions in plasma inflammatory cytokine levels were seen by day 5 (almost no change was observed in the first 2 days) of treatment in responders, while no reduction in inflammatory cytokine levels was detected in the two nonresponders (respiratory death). Moreover, improvements in lung physiology (LIS, $P_{a,O_2}:F_{I,O_2}$) and indices of pulmonary vascular permeability (BAL albumin) paralleled the reductions in both plasma and BAL TNF-α, IL-1β, IL-6 and IL-8 levels. When treatment was ineffective in lowering plasma and BAL inflammatory cytokine levels, lung function did not improve and patients died from refractory respiratory failure [95]. The striking parallelism between improvements in lung function and reduction in cytokine levels during GC treatment support the link between HDR and progression of ARDS. Of importance is that, when effective, GC treatment suppressed production of all measured inflammatory cytokines. A similar degree of anti-inflammatory action may not be achieved by specific (expensive) antimediator therapy (*i.e.* monoclonal antibody directed at TNF or antagonist directed at IL-1 receptors).

We also found that intensity and duration of the host defence response prior to initiating GC treatment may have been an important factor in determining response to therapy. At initiation of treatment, rapid responders had significantly lower inflammatory cytokine values than delayed responders or nonresponders. The only observed difference between the two groups of patients was a significantly higher plasma IL-6 level in nonresponders in the early phase of ARDS (fig. 3). Nonresponders also had physiological evidence of accelerated fibroproliferation. It has been shown that IL-6 is a potent inducer of *de novo* synthesis of tissue inhibitor of metalloproteinases (TIMP), which inhibits collagenase [214]. It is interesting to speculate that elevated IL-6 levels in nonresponders could play a significant role in developing pulmonary fibrosis by upregulating TIMP, which would inhibit the normal remodelling and removal of newly formed collagen by collagenase. Because cytokine levels at initiation of treatment were similar between delayed and nonresponders, it is also possible that a more advanced form of fibroproliferation may have prevented adequate GC tissue penetration in the latter group. In these patients, GC administration earlier in the course of ARDS or in higher doses could have been more effective.

Our findings suggest that GC treatment should be prolonged to achieve and sustain suppression of the HDR necessary to reverse progression of ARDS. This is supported by prior animal [215–217] and clinical studies [2, 32]. In experimental acute lung injury, GC treatment is effective in decreasing lung collagen and oedema formation as long as treatment is prolonged, but withdrawal rapidly negates the positive effects of therapy [216–218]. Limiting GC treatment to the first 6 days after acute lung injury enhances accumulation of collagen after discontinuing therapy, whereas GC given later, on days 7

until 12, have an alleviating effect [217]. Furthermore, a short-course GC treatment may negatively affect the host response by significantly enhancing cytokine response to LPS for up to 6 days [219]. Two clinical studies have shown that premature discontinuation of GC administration in late ARDS was associated with deterioration in lung function that resolved with the reinstitution of treatment [2, 32].

Results similar to those reported by our group [50] were also obtained in patients with sepsis and early ARDS [35]. In 12 patients with septic shock (eight with ARDS), a 5-day continuous low-dose hydrocortisone infusion (10 mg·h^{-1}) was associated with a statistically significant (p<0.01) reduction (in comparison to 45 control patients with similar condition) in SIRS score and circulatory levels of C-reactive protein and PLA$_2$ by day 3 of treatment [35]. Upon withdrawing treatment, however, PLA$_2$ and C-reactive protein levels increased and the SIRS amplified. Amplification of SIRS was associated with a second bout of hypotension in four patients that resolved upon reinstituting GC therapy [35]. During GC treatment, suppression of the HDR was associated with a much lower mortality than in the control group (8 *vs* 40%). Measurement of cortisol levels during this study provided additional evidence of the central role of the HPA axis in controlling the host defence response. During hydrocortisone infusion, plasma cortisol concentration and SIRS score decreased, but returned toward baseline values after discontinuing therapy.

Conclusion

The host defence response to insults is similar regardless of the tissue involved and results from an interactive network of specialized and interconnected pathways. An exaggerated and protracted host defence response plays a key role in ARDS outcome and is accountable for the histological, laboratory, clinical, and physiological findings seen during the course of unresolving ARDS. Continued elevated production of host defence response mediators, such as TNF-α, IL-1β, and IL-6, prevents effective restoration of lung anatomy and function by sustaining inflammation, coagulation and fibroproliferation. The degree of initial host defence response may determine the progress of ARDS. Elevated levels of inflammatory cytokines cause glucocorticoid resistance, offsetting the modulatory activity of the HPA axis. Predictors of poor outcome in ARDS as reported in the literature are, in actuality, manifestations of a persistent and exaggerated host defence response. During ARDS, downregulation of the host defence response is responsible for the lack of laboratory and clinical amplification of SIRS with nosocomial infections. These findings dispute the hypothesis, based on clinical criteria, that in unresolving ARDS, nosocomial infections amplify SIRS and lead to MODS and death. Spontaneous or treatment-induced suppression of the exaggerated and autonomous pulmonary host defence response is associated with improvement in lung function and outcome. Glucocorticoid resistance in ARDS and severe sepsis may respond to exogenous glucocorticoid administration. Clinical and experimental studies monitoring laboratory and clinical markers of the host defence response have shown that

effective containment of the host defence response in ARDS and sepsis may be achieved only if glucocorticoid administration is prolonged. In ARDS, glucocorticoid treatment may be highly effective when given in noninfected patients before end-stage fibrosis develops. Glucocorticoid treatment of unresolving ARDS cannot be recommended unless proven effective in a double-blind randomized study (in progress).

Recent appreciation of the complex relationship between the HPA axis and modulation of the host defence response in critical illness, and the lack of randomized studies evaluating prolonged glucocorticoid administration in this patient population, necessitate a reappraisal of the role of exogenous glucocorticoid treatment in ARDS and sepsis. Lack of financial incentive for the pharmaceutical industry, the blind acceptance that prior randomized studies have conclusively proven the lack of efficacy for glucocorticoids and the hope of finding a "magic bullet" among the many sites of possible interaction on the host defence response web, has discouraged this type of clinical investigation. It is my hope that the information provided here will encourage a critical re-evaluation of this inexpensive form of immunomodulation, and stimulate additional research.

References

1. Bachofen M, Weibel ER. Alterations of the gas exchange apparatus in adult respiratory insufficiency associated with septicemia. *Am Rev Respir Dis* 1977; 116: 589–615.
2. Ashbaugh DG, Maier RV. Idiopathic pulmonary fibrosis in adult respiratory distress syndrome. Diagnosis and treatment. *Arch Surg* 1985; 120: 530–535.
3. Meduri GU, Chinn AJ, Leeper KV, *et al*. Corticosteroid rescue treatment of progressive fibroproliferation in late ARDS. Patterns of response and predictors of outcome. *Chest* 1994; 105: 1516–1527.
4. Hill JD, Ratliff JL, Parrott JCW, *et al*. Pulmonary pathology in acute respiratory insufficiency: lung biopsy as a diagnostic tool. *J Thorac Cardiovasc Surg* 1976; 71: 64–71.
5. Bell RC, Coalson JJ, Smith JD, Johanson WG Jr. Multiple organ system failure and infection in adult respiratory distress syndrome. *Ann Intern Med* 1983; 99: 293–298.
6. Ashbaugh DG, Petty TL. Sepsis complicating the acute respiratory distress syndrome. *Surgery* 1972; 135: 865–869.
7. Zapol WM, Trelstad RL, Coffey JW, Tsai I, Salvador RA. Pulmonary fibrosis in severe acute respiratory failure. *Am Rev Respir Dis* 1979; 119: 547–554.
8. Collins JF, Smith JD, Coalson JJ, Johanson WG Jr. Variability in lung collagen amounts after prolonged support of acute respiratory failure. *Chest* 1984; 85 (5): 641–646.
9. Pratt PC, Vollmer RT, Shelburne JD, Crapo JD. Pulmonary morphology in a multihospital collaborative extracorporeal membrane oxygenation project. *Am J Pathol* 1979; 95: 191–214.
10. Meduri GU. Late ARDS. *New Horizons* 1993; 1: 563–577.
11. Snyder LS, Hertz MI, Harmon KR, Bitterman PB. Failure of lung repair following acute lung injury: Regulation of the fibroproliferative response. *Chest* 1990; 98: 733–738.
12. Calandrino FS Jr, Anderson DJ, Mintun MA, Schuster DP. Pulmonary vascular permeability during the adult respiratory distress syndrome: a positron emission tomographic study. *Am Rev Respir Dis* 1988; 138: 421–428.
13. Bone RC, Balk RA, Cerra FB, *et al*. Definitions for sepsis and organ failure and guidelines for the use of innovative therapies in sepsis. *Chest* 1992; 101: 1644–1655.
14. Andrews CP, Coalson JJ, Smith JD, Johanson WG Jr. Diagnosis of nosocomial bacterial pneumonia in acute, diffuse lung injury. *Chest* 1981; 80: 254–258.
15. Montgomery AB, Stager MA, Carrico CJ, Hudson LD. Causes of mortality in patients with the adult respiratory distress syndrome. *Am Rev Respir Dis* 1985; 132: 485–489.
16. Seidenfeld JJ, Pohl DF, Bell RC, Harris GD, Johanson WG Jr. Incidence, site, and outcome of infections in patients with the adult respiratory distress syndrome. *Am Rev Respir Dis* 1986; 134: 12–16.
17. Suchyta MR, Clemmer TP, Elliott CG, Orme JF Jr, Weaver LK. The adult respiratory distress syndrome. A report of survival and modifying factors. *Chest* 1992; 101: 1074–1079.
18. Mancebo J, Artigas A. A clinical study of the adult respiratory distress syndrome. *Crit Care Med* 1987; 15: 243–246.
19. Raffin TA. Intensive care unit survival of patients with systemic illness. *ARRD* 1989; 140: 528–535.
20. Wheeler AP, Jesmok G, Brigham KL. Tumor necrosis factor's effects on lung mechanics, gas exchange, and airway reactivity in sheep. *Am Physiol Soc* 1990; 2542–2549.
21. Meduri GU, Chastre J. The standardization of bronchoscopic techniques for ventilator-associated pneumonia. *Chest* 1992; 102: 557–564S.
22. Baselski V, El-Torky M, Coalson SS, Griffin J. The standardization of criteria for processing and interpreting laboratory specimens. *Chest* 1992; 102: 571–579S.
23. Raghow R. The role of extracellular matrix in postinflammatory wound healing and fibrosis. *FASEB J* 1994; 8: 823–831.
24. Udelsman R, Holbrook NJ. Endocrine and molecular responses to surgical stress. *Curr Probl Surg* 1994; 31: 653–720.
25. Baumann H, Gauldie J. The acute phase response. *Immunology Today* 1994; 15: 74–80.
26. Fischer JE, Hasselgren PO. Cytokines and glucocorticoids in the regulation of the "Hepato-Skeletal Muscle Axis" in sepsis. *Am J Surg* 1991; 161: 266–271.
27. Cerami A. Inflammatory cytokines. *Clinical Immunol and Immunopathol* 1992; 62: S3–S10.
28. Kovacs EJ, Dipietro LA. Fibrogenic cytokines and connective tissue production. *FASEB J* 1994; 8: 854–861.
29. Hart PH, Whitty GA, Burgess DR, Croatto M, Hamilton JA. Augmentation of glucocorticoid action on human monocytes by interleukin-4. *Lymphokine Research* 1990; 9: 147–153.
30. Munck A, Guyre PM, Holbrook NJ. Physiological functions of glucocorticoids in stress and their relation to pharmacological actions. *Endocrine Reviews* 1984; 5: 25–44.
31. Zuckerman SH, Shellhaas J, Butler LD. Differential regulation of lipopolysaccharide-induced interleukin 1 and

tumor necrosis factor synthesis: effects of endogenous and exogenous glucocorticoids and the role of the pituitary-adrenal axis. *Eur J lmmunol* 1989; 19: 301–305.

32. Hooper RG, Kearl RA. Established ARDS treated with a sustained course of adrenocortical steroids. *Chest* 1990; 97: 138–143.

33. Hooper RG, Kearl RA. Treatment of established ARDS - steroids, antibiotics, and antifungal therapy. *Chest* 1991; 100: 137S.

34. Meduri GU, Belenchia JM, Estes RJ, Wunderink RG, ElTorky M, Leeper KV Jr. Fibroproliferative phase of ARDS. Clinical findings and effects of corticosteroids. *Chest* 1991; 100: 943–952.

35. Briegel J, Kellermann W, Forst H, *et al.* The Phospholipase A2 Study Group. Low-dose hydrocortisone infusion attenuates the systemic inflammatory response syndrome. *Clin Invest* 1994; 72: 782–787.

36. Soni A, Pepper GM, Wyrwinski PM, *et al.* Adrenal insufficiency occurring during septic shock: incidence, outcome, and relationship to peripheral cytokine levels. *Am J Med* 1995; 98: 266–271.

37. Schneider AJ, Voerman HJ. Abrupt hemodynamic improvement in late septic shock with physiological doses of glucocorticoids. *Inten Care Med* 1991; 17: 436–437.

38. Perlstein RS, Whitnall MH, Abrams JS, Mougey EH, Neta R. Synergistic roles of interleukin-6, interleukin-1, and tumor necrosis factor in the adrenocorticotropin response to bacterial-lipopolysaccharide *in vivo. Endocrinology* 1993; 132: 946–952.

39. Hermus ARMM, Sweep CGJ. Cytokines and the hypothalamic-pituitary-adrenal axis. *J Steroid Biochem Molec Biol* 1990; 37: 867–871.

40. Vermes I, Beishuizen A, Hampsink RM, Haanen C. Dissociation of plasma adrenocorticotropin and cortisol levels in critically ill patients: possible role of endothelin and atrial natriuretic hormone. *J Clin Endocrinol Metab* 1995; 80: 1238–1242.

41. Hammond GL. Potential functions of plasma steroid-binding proteins. *Trends Endocrinol Metab* 1995; 6: 298–304.

42. Barnes PJ, Greening AP, Crompton GK. Glucocorticoid resistance in asthma. *Am J Respir Crit Care Med* 1995; 152: S125–S142.

43. Pratt WB. The role of heat shock proteins in regulating the function, folding, and trafficking of the glucocorticoid receptor. *J Biol Chem* 1993; 268: 21455–21458.

44. Detera-Wadleigh SD, Karl M. The glucocorticoid receptor gene. *Ann Intern Med* 1993; 119: 1113–1124.

45. Pruzanski W, Vadas P. Phospholipase A2 - a mediator between proximal and distal effectors of inflammation. *Immunology Today* 1991; 12: 143–146.

46. Goulding NJ, Guyre PM. Glucocorticoids, lipocortins and the immune response. *Curr Opin Immunol* 1993; 5: 108–113.

47. Santos AA, Scheltinga MR, Lynch E, *et al.* Elaboration of interleukin 1-receptor antagonist is not attenuated by glucocorticoids after endotoxemia. *Arch Surg* 1993; 128: 138–144.

48. Tutor JD, Mason CM, Dobard E, Beckerman RC, Summer WR, Nelson S. Loss of compartmentalization of alveolar tumor necrosis factor after lung injury. *Am J Respir Crit Care Med* 1994; 149: 1107–1111.

49. Meduri GU, Headley S, Kohler G, *et al.* Persistent elevation of inflammatory cytokines predicts a poor outcome in ARDS. Plasma IL-1β and IL-6 are consistent and efficient predictors of outcome over time. *Chest* 1995; 107: 1062–1073.

50. Meduri GU, Kohler G, Headley S, Tolley E, Stentz F, Postlethwaite A. Inflammatory cytokines in the BAL of patients with ARDS. Persistent elevation over time predicts poor outcome. *Chest* 1995; 108: 1303–1314.

51. Dofferhoff ASM, De Jong HJ, Bom VJJ, *et al.* Complement activation and the production of inflammatory mediators during the treatment of severe sepsis in humans. *Scand J Infect Dis* 1992; 24: 197–204.

52. Langleben D, Demarchie M, Laporta D, Spanier AH, Schlesinger RD, Stewart DJ. Endothelin-1 in acute lung injury and the adult respiratory distress syndrome. *Am Rev Respir Dis* 1993; 148: 1646–1650.

53. Waage A, Halstensen A, Espevik T. Association between tumour necrosis factor in serum and fatal outcome in patients with meningococcal disease. *Lancet* 1987; 355–357.

54. Girardin E, Grau GE, Dayer JM, Roux-Lombard P, The J5 Study Group, Lambert PH. Tumor necrosis factor and interleukin-1 in the serum of children with severe infectious purpura. *N Engl J Med* 1988; 319: 397–400.

55. Marano MA, Fong Y, Moldawer LL, *et al.* Serum cachectin/tumor necrosis factor in critically ill patients with burns correlates with infection and mortality. *Surg Gynecol & Obstet* 1990; 170: 32–38.

56. Sullivan JS, Kilpatrick L, Costarino AT Jr, Lee SC, Harris MC. Correlation of plasma cytokine elevations with mortality rate in children with sepsis. *J Pediatr* 1992; 120: 510–515.

57. Ertel W, Scholl FA, Gallati H, Bonaccio M, Schildberg FW, Trentz O. Increased release of soluble tumor necrosis factor receptors into blood during clinical sepsis. *Arch Surg* 1994; 129: 1330–1337.

58. Damas P, Ledoux D, Nys M, *et al.* Cytokine serum level during severe sepsis in human IL-6 as a marker of severity. *Ann Surg* 1992; 215 (4): 356–362.

59. Calandra T, Gerain J, Heumann D, Baumgartner JD, Glauser MP, Swiss-Dutch J5 Immunoglobulin Study Group. High circulating levels of interleukin-6 in patients with septic shock: evolution during sepsis, prognostic value, and interplay with other cytokines. *Am J Med* 1991; 91: 23–29.

60. Pinsky MR, Matuschak GM. Multiple systems organ failure: failure of host defense homeostasis. *Crit Care Clin* 1989; 5: 199–219.

61. Fisher CJ Jr, Opal SM, Dhainaut JF, *et al.*, The CB0006 Sepsis Syndrome Study Group. Influence of an anti-tumor necrosis factor monoclonal antibody on cytokine levels in patients with sepsis. *Crit Care Med* 1993; 21 (3): 318–327.

62. Casey LC, Balk RA, Bone RC. Plasma cytokine and endotoxin levels correlate with survival in patients with the sepsis syndrome. *Ann Intern Med* 1993; 119: 771–778.

63. Hack Ce, DeGroot ER, Felt-Bersma RJF, *et al.* Increased plasma levels of interleukin-6 in sepsis. *Blood* 1989; 74: 1704–1710.

64. Moscovitz H, Shofer F, Mignott H, Behrman A, Kilpatrick L. Plasma cytokine determinations in emergency department patients as a predictor of bacteremia and infectious disease severity. *Crit Care Med* 1994; 22: 1102–1107.

65. Marty C, Misset B, Tamion F, Fitting C, Carlet J, Cavaillon JM. Circulating interleukin-8 concentrations in patients with multiple organ failure of septic and non-septic origin. *Crit Care Med* 1994; 22 (4): 673–679.

66. Suter PM, Suter S, Girardin E, Roux-Lombard P, Grau GE, Dayer JM. High bronchoalveolar levels of tumor

necrosis factor and its inhibitors, interleukin-1, interferon, and elastase, in patients with adult respiratory distress syndrome after trauma, shock, or sepsis. *Am Rev Respir Dis* 1992; 145: 1016–1022.

67. Romaschin AD, DeMajo WC, Winton T, *et al*. Systemic phospholipase A_2 and cachectin levels in adult respiratory distress syndrome and multiple-organ failure. *Clin Biochem* 1992; 25: 55–60.

68. Roumen RMH, Hendriks T, van der Ven-Jongekrijg J, *et al*. Cytokine patterns in patients after major vascular surgery, hemorrhagic shock, and severe blunt trauma. Relation with subsequent adult respiratory distress syndrome and multiple organ failure. *Ann Surg* 1993; 218 (6): 769–776.

69. Miller EJ, Cohen AB, Magao S, *et al*. Elevated levels of NAP-1/interleukin-8 are present in the airspaces of patients with the adult respiratory distress syndrome and are associated with increased mortality. *Am Rev Respir Dis* 1992; 146: 427–432.

70. Gunther K, Baughman RP, Pattishall E, Whitsell J. Evaluation of intrapulmonary interleukin-8 in patients with sepsis-induced adult respiratory distress syndrome. *Am J Respir Crit Care Med* 1994; 149: A427.

71. Chollett-Martin S, Montravers P, Gibert C, *et al*. Relationship between polymorphonuclear neutrophils and cytokines in patients with adult respiratory distress syndrome. *Ann NY Acad Sci* 1994; 725: 354–366.

72. Calandra T, Baumgartner JD, Grau GE, *et al.*, the Swiss-Dutch J5 Immunoglobulin Study Group. Prognostic values of tumor necrosis factor/cachectin, interleukin-1, interferon-α, and interferon-γ in the serum of patients with septic shock. *J Infect Dis* 1990; 161: 982–987.

73. Pinski MR, Vincent JL, Deviere J, Alegre M, Kahn RJ, Dupont E. Serum cytokine levels in human septic shock. Relation to multiple-system organ failure and mortality. *Chest* 1993; 103: 565–575.

74. Burkhardt A. Alveolitis and collapse in the pathogenesis of pulmonary fibrosis. *Am Rev Respir Dis* 1989; 140: 513–524.

75. Harris TR, Bernard GR, Brigham KL, *et al*. Lung microvascular transport properties measured by multiple indicator dilution methods in patients with adult respiratory distress syndrome. *Am Rev Respir Dis* 1990; 141: 272–280.

76. Kawamura M, Yamasawa F, Ishizaka A, *et al*. Serum concentration of 7S collagen and prognosis in patients with the adult respiratory distress syndrome. *Thorax* 1994; 49 (2): 144–146.

77. Clark JG, Milberg JA, Steinberg KP, Hudson LD. Type III procollagen peptide in the adult respiratory distress syndrome. Association of increased peptide levels in bronchoalveolar lavage fluid with increased risk for death. *Ann Intern Med* 1995; 122: 17–23.

78. Raponi G, Antonelli M, Gaeta A, *et al*. Tumor necrosis factor in serum and in bronchoalveolar lavage of patients at risk for the adult respiratory distress syndrome. *J Crit Care* 1992; 17 (3): 183–188.

79. Siler TM, Swierkosz JE, Hyers TM, Fowler AA, Webster RO. Immunoreactive interleukin-1 in bronchoalveolar lavage fluid of high-risk patients and patients with the adult respiratory distress syndrome. *Exp Lung Res* 1989; 15: 881–894.

80. Chollett-Martin S, Montravers P, Gibert C, Desmonts JM, Chastre J, Gougerot-Pocidalo MA. Increased IL-8 levels in plasma and bronchoalveolar lavage fluid during the adult respiratory distress syndrome. *Infection and Immunity* 1993; 61: 4553–4559.

81. Torre D, Zeroli C, Giola M, *et al*. Levels of interleukin-8 in patients with adult respiratory distress syndrome. *J Infect Dis* 1993; 167: 505–506.

82. Van Nhieu JT, Misset B, Lebargy F, Carlet J, Bernaudin JF. Expression of tumor necrosis factor-α gene in alveolar macrophages from patients with the adult respiratory distress syndrome. *Am Rev Respir Dis* 1993; 147: 1585–1589.

83. Li XY, Donaldson K, Brown D, MacNee W. The role of tumor necrosis factor in increased airspace epithelial permeability in acute lung inflammation. *Am J Respir Cell Mol Biol* 1995; 13: 185–195.

84. Jacobs RF, Tabor DR, Burks AW, Campbell GD. Elevated interleukin-1 release by human alveolar macrophages during the adult respiratory distress syndrome. *Am Rev Respir Dis* 1989; 140: 1686–1692.

85. Donnelly SC, Strieter RM, Kunkel SL, *et al*. Interleukin-8 and development of adult respiratory distress syndrome in at-risk patient groups. *Lancet* 1993; 341: 643–647.

86. Xing Z, Jordana M, Kirpalani H, Driscoll KE, Schall TJ, Gauldie J. Cytokine expression by neutrophils and macrophages *in vivo*: Endotoxin induces tumor necrosis factor-α, macrophage inflammatory protein-2, interleukin-1β, and interleukin-6 but not RANTES or transforming growth factor-β1 mRNA expression in acute lung inflammation. *Am J Respir Cell Mol Biol* 1994; 10: 148–153.

87. Monkemuller K, Meduri GU, Headley S, Kohler G, Leeper KV. Serum and BAL inflammatory cytokines patterns at the onset of ARDS. Comparison between direct and indirect lung injury (Abstract). *Am Rev Respir Dis* 1994; 150: A427.

88. Vadas P, Pruzanski W, Stefanski E, Ruse J, Farewell V, McLaughlin J, Bombardier C. Concordance of endogenous cortisol and phospholipase A_2 levels in gram-negative septic shock: a prospective study. *J Lab Clin Med* 1988; 111: 584–590.

89. Bernard GR, Korley V, Chee P, Swindell B, Ford-Hutchinson AW, Tagari P. Persistent generation of peptido leukotrienes in patients with the adult respiratory distress syndromes. *Am Rev Respir Dis* 1991; 144: 263–267.

90. Robbins RA, Russ WD, Rasmussen JK, Clayton MM. Activation of the complement system in the adult respiratory distress syndrome. *Am Rev Respir Dis* 1987; 135: 651–658.

91. Crouser ED, Dorinsky PM. Gastrointestinal tract dysfunction in critical illness: pathophysiology and interaction with acute lung injury in adult respiratory distress syndrome/multiple organ dysfunction syndrome. *New Horizons* 1994; 2: 476–487.

92. Meduri GU, ElTorky M, Winer-Muram HT. The fibroproliferative phase of late adult respiratory distress syndrome. *Semin Respir Infect* 1995; 10: 154–175.

93. Leff JA, Bodman ME, Cho OJ, Rohrbach S, Reiss OK, Vannice JL, Repine JE. Post-insult treatment with interleukin-1 receptor antagonist decreases oxidative lung injury in rats given intratracheal interleukin-1. *Am J Respir Crit Care Med* 1994; 150: 109–112.

94. Donnelly SC, Strieter RM, Kunkel SL, *et al*. Prognostic implications of interleukin-10 (IL-10) in bronchoalveolar lavage (BAL) samples in the adult respiratory distress syndrome (ARDS) (Abstract). *Am J Respir Crit Care Med* 1995; 151: A75.

95. Meduri GU, Headley S, Tolley E, Shelby M, Stentz F, Postlethwaite A. Plasma and BAL cytokine response to

corticosteroid rescue treatment in late ARDS. *Chest* 1995; 108: 1315–1325.

96. Cordier J-F, Peyrol S, Loire R. Bronchiolitis obliterans organizing pneumonia as a model of inflammatory lung disease. *In*: Epler GR, Ed. Diseases of the Bronchioles. New York, Raven Press Ltd. 1994; pp. 313–345.

97. Idell S. Extravascular coagulation and fibrin deposition in acute lung injury. *New Horizons* 1994; 2: 566–574.

98. Bone RC. Modulators of coagulation: a critical appraisal of their role in sepsis. *Arch Intern Med* 1992; 151: 1381–1389.

99. Hasegawa N, Husari AW, Hart WT, Kandra TG, Raffin TA. Role of the coagulation system in ARDS. *Chest* 1994; 105: 268–277.

100. Kadas L, Szell K. The role of megakaryocytes and tissue mast cells in the respiratory distress syndrome of adults. *Acta Morphologica Acad Sci Hung* 1981; 29: 395–404.

101. Idell S, Koenig KB, Fair DS, Martin TR, McLarty J, Maunder RJ. Serial abnormalities of fibrin turnover in evolving adult respiratory distress syndrome. *Am J Physiol* 1991; 261: L240–L248.

102. Bertozzi P, Astedt B, Zenzius L, *et al*. Depressed bronchoalveolar urokinase activity in patients with adult respiratory distress syndrome. *N Engl J Med* 1990; 322: 890–897.

103. Moalli R, Doyle JM, Tahhan HR, Hasan FM, Braman SS, Sadeen T. Fibrinolysis in critically ill patients. *Am Rev Respir Dis* 1989; 140: 287–293.

104. Fuchs-Buder T, de Moerloose P, Ricou B, *et al*. Time course of procoagulant activity and D dimer in bronchoalveolar fluid of patients at risk for or with acute respiratory distress syndrome. *Am J Respir Crit Care Med* 1996; 153: 163–167.

105. Kirschstein W, Heene DL. Fibrinolysis inhibition in acute respiratory distress syndrome. *Scand J Clin Lab Invest* 1985; 45: 87–94.

106. de Boer JP, Creasy AA, Chang A, *et al*. Activation patterns of coagulation and fibrinolysis in baboons following infusion with lethal or sublethal dose of *Escherichia coli*. *Circulatory Shock* 1993; 39: 59–67.

107. Carvalho AC. Blood alterations during ARDS. *In*: Zapol WM, Falke KJ, Eds. Acute respiratory failure. New York, Marcel Dekker 1985; pp. 303–346.

108. Bone RC, Balk R, Slotman G, *et al.*, the Prostaglandin E1 Study Group. Adult respiratory distress syndrome. Sequence and importance of development of multiple organ failure. *Chest* 1992; 101: 320–326.

109. Schneider RC, Zapol WM, Carvalho AC. Platelet consumption and sequestration in severe acute respiratory failure. *Am Rev Respir Dis* 1980; 122: 445–451.

110. Quinn DA, Carvalho AC, Geller E, *et al*. ^{99m}Tc-fibrinogen scanning in adult respiratory distress syndrome. *Am Rev Respir Dis* 1987; 135: 100–106.

111. Taylor FB. The inflammatory-coagulant axis in the host response to gram-negative sepsis: regulatory roles of proteins and inhibitors of tissue factor. *New Horizons* 1994; 2: 555–565.

112. Greene R, Zapol WM, Snider MT, *et al*. Early bedside detection of pulmonary vascular occlusion during acute respiratory failure. *Am Rev Respir Dis* 1981; 124: 593–601.

113. Tomaseski JF, Davies P, Boggis C, Greene R, Zapol WM, Reid LM. The pulmonary vascular lesions of the adult respiratory distress syndrome. *Am J Pathol* 1983; 112: 112–126.

114. Malik AB. Pulmonary microembolism and lung vascular injury. *Eur Respir J* 1990; 3: 499S–506S.

115. Leavell KJ, Peterson MW, Gross TJ. The role of fibrin degradation products in neutrophil recruitment to the lung. *Am J Respir Cell Mol Biol* 1996; 14: 53–60.

116. Schapira M, Gardaz JP, Py P, Lew PD, Perrin LH, Suter PM. Prekallikrein activation in the adult respiratory distress syndrome. *Bull Eur Physiopathol Respir* 1985; 21: 237–247.

117. McDonald JA. The yin and yang of fibrin in the airways. *N Engl J Med* 1990; 322: 929–931.

118. Kuhn C III, Boldt J, King TE, Crouch E, Vartio T, McDonald JA. An immunohistochemical study of architectural remodeling and connective tissue synthesis in pulmonary fibrosis. *Am Rev Respir Dis* 1989; 140: 1693–1703.

119. Colvin RB, Gardner PI, Roblin RO, Verderber EL, Lanigan JM, Mosesson MW. Cell surface fibrinogen-fibrin receptors on cultured human fibroblasts. *Lab Invest* 1979; 41: 464–473.

120. Cherington PV, Pardee AB. Synergistic effects of epidermal growth factor and thrombin on the growth stimulation of diploid chinese hamster fibroblasts. *J Cellular Physiology* 1980; 105: 25–32.

121. Fukuda Y, Ishizaki M, Masuda Y, Kimura G, Kawanami O, Masugi Y. The role of intra-alveolar fibrosis in the process of pulmonary structural remodeling in patients with diffuse alveolar damage. *Am J Pathol* 1987; 126: 171–182.

122. Lamy M, Fallat RL, Koeniger E, Dieterich HP, Ratliff JL, Eberhart RC, Tucker HJ, Hill JD. Pathologic features and mechanisms of hypoxemia in adult respiratory distress syndrome. *Am Rev Respir Dis* 1976; 114: 267–284.

123. Zapol WM, Snider MT, Rie M. Pulmonary circulation in adult respiratory distress syndrome. *In*: Artigas A, Lemaire F, Suter PM, Zapol WM, Eds. Adult Respiratory Distress Syndrome. Singapore, Churchill Livingstone 1992; pp. 259–278.

124. Bitterman PB, Henke CA. Fibroproliferative disorders. *Chest* 1991; 99: 81S–84S.

125. Shen AS, Haslett DC, Feldsien C, Henson PM, Cherniack RM. The intensity of chronic lung inflammation and fibrosis after bleomycin is directly related to the severity of acute injury. *Am Rev Respir Dis* 1988; 137: 564–571.

126. Kuhn C III, Boldt J, King TE Jr, Crouch E, Vartio R, McDonald JA. An immunohistochemical study of architectural remodeling and connective tissue synthesis in pulmonary fibrosis. *Am Rev Respir Dis* 1989; 140: 1693–1703.

127. Elias JA, Freundlich B, Kern JA, Rosenbloom J. Cytokine networks in the regulation of inflammation and fibrosis in the lung. *Chest* 1990; 97: 1439–1445.

128. Entzian P, Huckstadt A, Kreipe H, Barth J. Determination of serum concentrations of type III procollagen peptide in mechanically ventilated patients. Pronounced augmented concentrations in the adult respiratory distress syndrome. *Am Rev Respir Dis* 1990; 142: 1079–1082.

129. Kropf J, Grobe E, Knoch M, Lammers M, Gressner AM, Lennartz H. The prognostic value of extracellular matrix component concentrations in serum during treatment of adult respiratory distress syndrome with extracorporeal CO_2 removal. *Eur J Clin Chem Clin Biochem* 1991; 29: 805–812.

130. Waydhas C, Nast-Kolb D, Trupka A, Lenk S, Duswald KH, Schweiberer L, Jochum M. Increased serum concentrations of procollagen type III peptide in severely

injured patients: an indicator of fibrosing activity. *Crit Care Med* 1993; 21: 240–247.

131. Molijn GJ, Koper JW, van Uffelen CJ, de Jong FH, Brinkmann AO, Bruining HA, Lamberts SWJ. Temperature-induced down-regulation of the glucocorticoid receptor in peripheral blood mononuclear leucocyte in patients with sepsis or septic shock. *Clin Endocrinol* 1995; 43: 197–203.

132. Farjanel J, Hartmann DJ, Guidet B, Luquel L, Offenstadt G. Four markers of collagen metabolism as possible indicators of disease in the adult respiratory distress syndrome. *Am Rev Respir Dis* 1993; 147 (5): 1091–1099.

133. Martin C, Papazian L, Payan MJ, Saux P, Gouin F. Pulmonary fibrosis correlates with outcome in established ARDS. Usefulness and safety of the transbronchial lung biopsy in mechanically-ventilated patients. *Chest* 1995; 107: 196–200.

134. Zapol WM, Snider MT. Pulmonary hypertension in severe acute respiratory failure. *N Engl J Med* 1977; 296: 476–480.

135. Matamis D, Lemaire F, Harf A, Brun-Buisson C, Ansquer JC, Atlan G. Total respiratory pressure-volume curves in the adult respiratory distress syndrome. *Chest* 1984; 86: 58–66.

136. Klose R, Osswald PM. Effects of PEEP on pulmonary mechanics and oxygen transport in the late stages of acute pulmonary failure. *Inten Care Med* 1981; 7: 165–170.

137. deLatorre FF. Study of gas exchange as a prognostic factor in adult respiratory distress syndrome. *In*: Artigas A, Lemaire F, Suter PM, Zapol WM, Eds. Adult Respiratory Distress Syndrome. Singapore, Churchill Livingstone 1992; pp. 371–378.

138. Heffner JE, Zamora CA. Clinical predictors of prolonged translaryngeal intubation in patients with the adult respiratory distress syndrome. *Chest* 1990; 97: 447–452.

139. Maunder RJ, Kubilis PS, Anardi DM, Hudson LD. Determinants of survival in the adult respiratory distress syndrome (ARDS). *Am Rev Respir Dis* 1989; A220.

140. Shimada Y, Yoshiya I, Tanaka K, Sone S, Sakurai M. Evaluation of the progress and prognosis of adult respiratory distress syndrome: simple respiratory physiologic measurement. *Chest* 1979; 76: 180–186.

141. Bernard GR, Luce JM, Sprung CL, *et al*. High-dose corticosteroids in patients with the adult respiratory distress syndrome. *N Engl J Med* 1987; 317: 1565–1570.

142. Simmons RS, Berdine GG, Seidenfeld JJ, *et al*. Fluid balance and the adult respiratory distress syndrome. *Am Rev Respir Dis* 1994; 135: 924–929.

143. Sloane PJ, Gee MH, Gottlieb JE, *et al*. A multicenter registry of patients with acute respiratory distress syndrome. *Am Rev Respir Dis* 1992; 146: 419–426.

144. Sinclair DG, Braude S, Haslam PL, Evans TW. Pulmonary endothelial permeability in patients with severe lung injury. *Chest* 1994; 106: 535–539.

145. Heffner JE, Brown LK, Barbieri CA, Harpel KS, DeLeo J. Prospective validation of an acute respiratory distress syndrome predictive score. *Am J Respir Crit Care Med* 1995; 152: 1518–1526.

146. Holter JF, Weiland JE, Pacht ER, Gadek JE, Davis WB. Protein permeability in the adult respiratory distress syndrome. Loss of size selectivity of the alveolar epithelium. *J Clin Invest* 1986; 78: 1513–1522.

147. Weiland JE, Davis WB, Holter JF, Mohammed JR, Dorinsky PM, Gadek JE. Lung neutrophils in the adult respiratory distress syndrome. Clinical and pathophysiologic significance. *Am Rev Respir Dis* 1986; 133: 218–225.

148. Martin TR, Pistorese BP, Hudson LD, Maunder RJ. The function of lung and blood neutrophils in patients with the adult respiratory distress syndrome. Implications for the pathogenesis of lung infections. *Am Rev Respir Dis* 1991; 144: 254–262.

149. Jorens PG, VanDamme J, DeBacker W, *et al*. Interleukin 8 (IL-8) in the bronchoalveolar lavage fluid from patients with the adult respiratory distress syndrome (ARDS) and patients at risk for ARDS. *Cytokine* 1992; 4 (6): 592–597.

150. Steimberg KP, Milberg JA, Martin TR, Maunder RJ, Cockrill BA, Hudson LD. Evolution of bronchoalveolar lavage cell population in the adult respiratory distress syndrome. *Am J Respir Crit Care Med* 1994; 150: 113–122.

151. Gunther K, Baughman RP, Rashkin M, Pattishall E. Bronchoalveolar lavage results in patients with sepsis-induced adult respiratory distress syndrome: evaluation of mortality and inflammatory response. *Am Rev Respir Dis* 1993; 147: A346.

152. Kelly CA, Fenwick JD, Corris PA, Fleetwood A, Hendrick DJ, Walters EH. Fluid dynamics during bronchoalveolar lavage. *Am Rev Respir Dis* 1988; 138: 81–84.

153. Meduri GU, Belenchia JM, Massey JD, ElTorky M, Tolley, EA. [67]Ga scintigraphy in diagnosing sources of fever in ventilated patients. *Inten Care Med* 1996; 22: 395–403.

154. Passamonte PM, Martinez AJ, Singh A. Pulmonary gallium concentration in the adult respiratory distress syndrome. *Chest* 1984; 85: 828–830.

155. Snow RL, Davies P, Pontoppidan H, Zapol WM, Reid L. Pulmonary vascular remodeling in adult respiratory distress syndrome. *Am Rev Respir Dis* 1982; 126: 887–892.

156. Unger JM, England DM, Bogust GA. Interstitial emphysema in adults: recognition and prognostic implications. *J Thorac Imaging* 1989; 4: 86–94.

157. Woodring JH. Pulmonary interstitial emphysema in the adult respiratory distress syndrome. *Crit Care Med* 1985; 13: 786–791.

158. Gattinoni L, Bombino M, Pelosi P, *et al*. Lung structure and function in different stages of severe adult respiratory distress syndrome. *JAMA* 1994; 271: 1722–1729.

159. Rouby JJ, Lherm T, de Lassale EM, *et al*. Histologic aspects of pulmonary barotrauma in critically ill patients with acute respiratory failure. *Inten Care Med* 1993; 19: 383–389.

160. Macklin MT, Macklin CC. Malignant interstitial emphysema of the lungs and mediastinum as an important occult complication in many respiratory diseases and other conditions: an interpretation of the clinical literature in light of laboratory experiments. *Medicine* 1944; 23: 281–358.

161. Tomasheski JR Jr. Pulmonary pathology of the adult respiratory distress syndrome. *Clin Chest Med* 1990; 11: 593–619.

162. Dinarello CA, Cannon JG, Wolff SM. New concepts on the pathogenesis of fever. *Rev Infect Dis* 1988; 10: 168–189.

163. Villar J, Ribeiro SP, Mullen BM, Kuliszewski M, Post M, Slutsky AS. Induction of the heat shock response reduces mortality rate and organ damage in a sepsis-induced acute lung injury model. *Crit Care Med* 1994; 22: 914–921.

164. Headley S, Tolley F, Meduri GU. Infections and the inflammatory response in ARDS. *Chest* 1997; (in press).

165. George DL, Falk PS, Meduri GU, *et al*. The epidemiology of nosocomial pneumonia in medical intensive

care unit (MICU) patients: a prospective study based on protected bronchoscopic sampling. *Infect Control Hosp Epidemiol* 1992; 21: 496–497.

166. Sutherland KR, Steinberg KP, Maunder RJ, Milberg JA, Allen DL, Hudson LD. Pulmonary infection during the acute respiratory distress syndrome. *Am J Respir Crit Care Med* 1995; 152: 550–556.

167. Estes R, Meduri GU. Pathogenesis of ventilator-associated pneumonia: mechanisms of bacterial colonization and airway inoculation. *Inten Care Med* 1995; 21: 365–383.

168. Meduri GU, Estes R. Pathogenesis of ventilator-associated pneumonia: the lower respiratory tract. *Inten Care Med* 1995; 21: 452–461.

169. Meduri GU, Mauldin GL, Wunderink RG, *et al*. Causes of fever and pulmonary densities in patients with clinical manifestations of ventilator-associated pneumonia. *Chest* 1994; 106: 221–233.

170. Rouby JJ, deLassale EM, Poete P, *et al*. Nosocomial bronchopneumonia in the critically ill. Histologic and bacteriologic aspects. *Am Rev Respir Dis* 1992; 146: 1059–1066.

171. Brunson M, Meduri GU, Leeper KV, Wunderink RG, Stanley T, Tolley E. Pneumonia in ARDS: diagnostic and prognostic value of bilateral bronchoscopic sampling. *Am Rev Respir Dis* 1993; 147: A355.

172. Wunderink RG, Woldenberg LS, Zeiss J, *et al*. Radiologic diagnosis of autopsy-proven ventilator-associated pneumonia. *Chest* 1992; 101: 458–463.

173. Winer-Muram HT, Rubin SA, Ellis JV, *et al*. Pneumonia and ARDS in the ventilated patient: diagnostic accuracy of the chest radiograph. *Radiology* 1993; 188: 479–485.

174. Kristiansson M, Soop M, Saraste L, Sundqvist KG. Postoperative circulating cytokine patterns - the influence of infection. *Inten Care Med* 1993; 19: 395–400.

175. Marshall J, Sweeney D. Microbial infection and the septic response in critical surgical illness. *Arch Surg* 1990; 125: 17–23.

176. Poole GV, Muakkassa FF, Griswold JA. The role of infection in outcome of multiple organ failure. *Am Surg* 1993; 59 (11): 727–732.

177. Ertel W, Kremer JP, Kenney J, *et al*. Downregulation of proinflammatory cytokine release in whole blood from septic patients. *Blood* 1995; 85: 1341–1347.

178. Ribeiro SP, Villar J, Slutsky AS. Induction of the stress response to prevent organ injury. *New Horizons* 1995; 3: 301–311.

179. Nelson S, Mason C, Bagby GJ, *et al*. Lipopolysaccharide-induced inhibition of intrapulmonary tumor necrosis factor and lung antibacterial defenses. *Am Rev Respir Dis* 1990; 141: A512.

180. Wilson JD Jr, Stewart RM, Fabian TC, Weinberg JA, Trenthem LL, Proctor KG. Plasma tumor necrosis factor and post-traumatic hyperdynamic sepsis evoked by endotoxin. *Shock* 1994; 1: 176–183.

181. Simpson SQ, Modi HN, Balk RA, Bone RC, Casey LC. Reduced alveolar macrophage production of tumor necrosis factor during sepsis in mice and men. *Crit Care Med* 1991; 19: 1060–1066.

182. Luger H, Graf H, Schwarz HP, Stummvoll HK, Luger TA. The serum interleukin 1 activity and monocyte interleukin 1 production in patients with fatal sepsis. *Crit Care Med* 1986; 14: 458–461.

183. Low-Friedrich I, Weisensee D, Mitrou P, Schoeppe W. Cytokines induce stress protein formation in cultured cardiac myocytes. *Basic Res Cardiol* 1992; 87: 12–18.

184. Klostergaard J, Barta M, Tomasovic SP. Hyperthermic modulation of tumor necrosis factor-dependent monocyte/macrophage tumor cytotoxicity *in vitro*. *J Biol Response Mod* 1989; 8: 262–277.

185. Snyder YM, Guthrie L, Evans GF, Zuckerman SH. Transcriptional inhibition of endotoxin-induced monokine synthesis following heat shock in murine peritoneal macrophages. *J Leucoc Biol* 1992; 51: 181–187.

186. De Maio A. The heat shock response. *New Horizons* 1995; 3: 198–207.

187. Riberio SP, Downey GP, Edelson JD, *et al*. Evidence that heat shock protein-72 (HSP72) participates in post-translational control of tumor necrosis factor in alveolar macrophages exposed to the stress response (Abstract). *Am J Respir Crit Care Med* 1995; 151: A166.

188. Udelsman R, Blake MJ, Stagg CA, Li D, Putney DJ, Holbrook NJ. Vascular heat shock protein expression in response to stress. Endocrine and autonomic regulation of this age-dependent response (Abstract). *J Clin Invest* 1993; 91: 465–473.

189. Udelsman R, Blake MJ, Stagg CA, Holbrook NJ. Endocrine control of stress-induced heat shock protein 70 expression *in vivo*. *Surgery* 1994; 115: 611–616.

190. Schooltink H, Van de Leur HS, Heinrich PC, Rose-John S. Up-regulation of the interleukin-6-signal transducing protein (gp130) by interleukin-6 and dexamethasone in HegG2 cells. *FEBS* 1992; 297: 263–265.

191. Komaki G, Gottshall PE, Somogyvari-Vigh A, Tatsuno I, Yathohgo T, Arimura A. Rapid increase in plasma IL-6 after hemorrhage, and posthemorrhage reduction of the IL-6 response to LPS, in conscious rats: interrelation with plasma corticosterone levels. *Neuroimmunomodulation* 1994; 1: 127–134.

192. Melby JC, Egdahl RH, Spink WW. Secretion and metabolism of cortisol after injection of endotoxin. *J Lab Clin Med* 1960; 56: 50–62.

193. Melby JC, Spink WW. Comparative studies on adrenal-cortical function and cortisol metabolism in healthy adults and in patients with shock due to infection. *J Clin Invest* 1958; 37: 1791–1798.

194. Reincke M, Allolio B, Wurth G, Winkelmann W. The hypothalamic-pituitary-adrenal axis in critical illness: response to dexamethasone and corticotropin-releasing hormone. *J Clin Endocrinol Metab* 1993; 77: 151–156.

195. Perrot D, Bonneton A, Dechaud H, Motin J, Pugeat M. Hypercortisolism in septic shock is not suppressible by dexamethasone infusion. *Crit Care Med* 1993; 21: 396–401.

196. Savu L, Zouaghi H, Carli A, Nunez EA. Serum depletion of corticosteroid binding activities: an early marker of human septic shock. *Biochem Biophysical Res Comm* 1981; 102: 411–419.

197. Pugeat M, Bonneton A, Perrot D, *et al*. Decreased immunoreactivity and binding activity of corticosteroid-binding globulin in serum in septic shock. *Clin Chem* 1989; 35: 1675–1679.

198. Rothwell PM, Udwadia ZF, Lawler PG. Cortisol response to corticotropin and survival in septic shock. *Lancet* 1991; 337: 1230–1231.

199. Sibbald WJ, Short A, Cohen MP, Wilson RF. Variations in adrenocortical responsiveness during severe bacterial infections. Unrecognized adrenocortical insufficiency in severe bacterial infections. *Ann Surg* 1977; 186: 29–33.

200. Almawi WY, Lipman ML, Stevens AC, Zanker B, Hadro Et, Strom TB. Abrogation of glucocorticosteroid-mediated inhibition of T cell proliferation by the synergistic

action of IL-1, IL-6, and IFN-γ. *J Immunol* 1991; 146: 3523–3527.

201. Kam JC, Szefler SJ, Surs W, Sher ER, Leung DYM. Combination IL-2 and IL-4 reduces glucocorticoid receptor-binding affinity and T cell response to glucocorticoids. *J Immunol* 1993; 151: 3460–3466.

202. Adcock IM, Shirasaki H, Gelder CM, Peters MJ, Brown CR, Barnes PJ. The effects of glucocorticoids on phorbol ester and cytokine stimulated transcription factor activation in human lung. *Life Sci* 1994; 55: 1147–1153.

203. Liu LY, Sun B, Tian Y, Bao-Zhang L, Wang J. Changes of pulmonary glucocorticoid receptor and phospholipase A₂ in sheep with acute lung injury after high dose endotoxin infusion. *Am Rev Respir Dis* 1993; 148: 878–881.

204. Fan J, Gong X, Wu J, Zhang Y, Xu R. Effect of glucocorticoid receptor (GR) blockade on endotoxemia in rats. *Circulatory Shock* 1994; 42: 76–82.

205. Gaillard RC, Turnill D, Sappino P, Muller AF. Tumor necrosis factor α inhibits the hormonal response of the pituitary gland to hypothalamic releasing factors. *Endocrinology* 1990; 127: 101–106.

206. Vankelecom H, Carmeliet P, Heremans H, *et al*. Interferon-γ inhibits stimulated adrenocorticotropin, prolactin, and growth hormone secretion in normal rat anterior pituitary cell cultures. *Endocrinology* 1990; 126: 2919–2926.

207. Mastorakos G, Chrousos GP, Webber JS. Recombinant interleukin-6 activates the hypothalamic pituitary-adrenal axis in humans. *J Clin Endocrinol Metab* 1993; 77: 1690–1694.

208. Kollef MH, Schuster DP. The acute respiratory distress syndrome. *N Engl J Med* 1995; 332: 27–37.

209. Lefering R, Neugebauer EAM. Steroid controversy in sepsis and septic shock: a meta-analysis. *Crit Care Med* 1995; 23: 1294–1303.

210. Cronin L, Cook DJ, Carlet J, *et al*. Corticosteroid treatment for sepsis: a critical appraisal and meta-analysis of the literature. *Crit Care Med* 1995; 23: 1430–1439.

211. Murray JF, Matthay MA, Luce JM, Flick MR. An expanded definition of the adult respiratory distress syndrome. *Am Rev Respir Dis* 1988; 138: 720–723.

212. Braude S, Haslam P, Hughes D, MacNaughton P, Evans TW. Chronic adult respiratory distress syndrome - a role for corticosteroids. *Crit Care Med* 1992; 20: 1187–1189.

213. Salvador RA, Fiedler-Hagy C, Coffey JW. Biochemical basis for drug therapy to prevent pulmonary fibrosis in ARDS. *In*: Zapol WM, Falke KJ, Eds. Acute Respiratory Failure. New York, Dekker, 1985; pp. 477–506.

214. Lotz M, Guerne PA. Interleukin-6 induces the synthesis of tissue inhibitor of metalloproteinases-1/erythroid potentiating activity (TIMP-1/EPA). *J Biol Chem* 1991; 266 (4): 2017–2020.

215. Jones RL, King EG. The effects of methylprednisolone on oxygenation in experimental hypoxemic respiratory failure. *J Trauma* 1975; 15: 297–303.

216. Hesterberg TW, Last JA. Ozone-induced acute pulmonary fibrosis in rats - prevention of increased rates of collagen synthesis by methylprednisolone. *Am Rev Respir Dis* 1981; 123: 47–52.

217. Hakkinen PJ, Schmoyer RL, Witschi HP. Potentiation of butylated-hydroxytoluene-induced acute lung damage by oxygen. Effects of prednisolone and indomethacin. *Am Rev Respir Dis* 1983; 128: 648–651.

218. Kehrer JP, Klein-Szanto AJP, Sorensen EMB, Pearlman R, Rosner MH. Enhanced acute lung damage following corticosteroid treatment. *Am Rev Respir Dis* 1984; 130: 256–261.

219. Barber AE, Coyle SM, Marano MA, *et al*. Glucocorticoid therapy alters hormonal and cytokine responses to endotoxin in man. *J Immunol* 1993; 150: 1999–2006.