Repair of metabolic processes

Jason Shipman, MD; Jeffrey Guy, MD; Naji N. Abumrad, MD, FACS

Inflammation is primarily an immune response aimed at protecting the organism from the devastating effects of pathogens, chemicals, and tissue injury. It is well established that once the body sustains an insult, local inflammatory mediators like prostaglandins and interleukins begin to circulate in the blood stream. These act both locally to direct inflammation and globally to indicate to the central nervous system the need for both added nutritional substrate directed toward vital organ systems and for blunted systemic inflammation. The hypothalamic-pituitary axis responds with cortisol and similar hormones that in the short term increase circulating carbohydrates, lipids, and proteins while simultaneously governing the immunologic response so that its potency will be precise and effective. In the acute phase of injury, these responses seem well founded and a natural answer to insult. Yet when the initial inflammatory phase wanes, the critically ill continue to show effects of protein catabolism, resulting in muscle wasting and dependence on ventilator support and immunosuppression, leaving them susceptible to severely debilitating infection, despite adequate nutritional support. Herein lies the principle paradox: despite alleviation of the initial insult, the critically ill continue to react to stress that is not there.

Without fail, critical care physicians have at some time in their career dealt with patients who survive their initial insult only to succumb to inevitable complications of protracted critical care. Their prognosis depended solely on the duration of intervention; the initial injury seemed not to matter. This was rationalized, without foundation, by transitively applying immunologic and endocrinologic principles of the initial or acute phase of injury to the heretofore unknown protracted or chronic phase of injury.

Irrespective of the nature of the insult, bacterial, viral, chemical, or physical, the initial response usually consists of damage to the immune cells with potential compromise to the blood supply of tissues. These events would then set into motion a cascade of feed-forward inflammatory responses, mediated by cytokines and hormones that aim at protecting the organism. Carl Nathan has defined these responses as a "system of information flow in response to injury and infection," whereby various cells are recruited, with each committed to release proinflammatory signals. Mast cells are recruited and release histamine, eicosanoids, tryptases, preformed tumor necrosis serum, proteases, and chemokines. Neurons release bioactive peptides in response to pain (1). Cell death ensues, and several intracellular proteins (e.g., heat shock proteins (2), high mobility group 1—HMGB1 (3)—and mitochondrial peptides bearing the N-formyl group (4)) enter into the extracellular space, which in turn triggers the production of cytokines, eicosanoids, nitric oxide, and reactive oxygen species (5, 6). Progression of the inflammatory responses requires continuous activation of macrophages and further recruitment of neutrophils. The net result is further release of cytokines, breakdown of hyaluronic acid in the extracellular matrix to low molecular weight hyaluronan that, in turn, further release chemokines and perhaps matrix metalloproteinases. Neutrophils release elastases, which with RO1 will activate matrix metalloproteinases, which in turn activate macrophage-derived latent transforming growth factor-β, the most potent known chemoattractant for neutrophils.

Interruption of the feed-forward cascade should theoretically result in transformation of the inflammatory responses to that of a healing response. Recent observations have pointed to several areas of effective interruption. A recent review (7) has identified several "checkpoints" at which early interruptions could lead to anti-inflammatory responses. Examples include a) shifting of arachidonate metabolism toward lipoxin formation in neutrophils, which in turn bind cellular receptors and block neutrophil influx; b) induction of COX2 in macrophages by microbial products and cytokines, and these in turn induce prostaglandin E_2, which at a later stage shift arachidonate metabolism to lipoxin formation; c) elevated levels of interleukin (IL)-10 and transforming growth factor-β inhibit the release of tumor necrosis factor (TNF)α and other proinflammatory mediators (8); d) presence of an intact hypothalamic-pituitary-adrenal axis is also necessary for appropriate anti-inflammatory response. Selective deficiencies of corticotropin-releasing factor, adrenocorticotropic hormone, and cortisol are all known to enhance the endotoxemic responses (9–11).

Kevin Tracey has also recently reviewed the evidence for the involvement of the autonomic nervous system as a check point at which cholinergic discharge blocks the release of TNFα from macrophages in the viscera. He termed this response as the "cholinergic anti-inflammatory pathway" that is mediated by the vagus nerve (12). Several studies have shown that vagal stimulation results in inhibition of proinflammatory cytokine synthesis, particularly TNFα, in macrophages and in liver and cardiac tissue (13). The primary effect on macrophages seems to be mediated by nicotinic, α-bungarotoxin–sensitive macrophage acetylcholine receptor (14), which is distinct from the muscarinic receptor activities identified on lymphocytes, peripheral blood mononuclear cells, and alveolar macrophages (15, 16).

In brief, it seems that many of the agents that are essential to induce and sustain inflammation, such as TNFα, prostaglandin E$_2$, interferon-γ, transforming growth factor-β, reactive oxygen intermediates, and reactive nitrogen intermediates are also agents that are important in sustaining the healing responses. Hence, the timing of intervention becomes important in promoting the healing processes. Thus far, most of the interventions have been nonspecific. Many of these relate to inherent advances in critical care, such as advances in ventilator support, utilization of better and more specific antimicrobial therapy, and the widespread use of enteral and parenteral nutrition. These advances have enabled physicians to improve the outcome and lessen the consequences of protracted intensive care. Recent trials concerning, corticotropin, somatotropin, insulin, and neuroimmunomodulation axes show some promise that there may be room for endocrine intervention in protracted critical illness. This review intends to incorporate these studies in a review of the metabolic processes of the critically ill.

Growth Hormone

Of all the hormones in the neuroendocrine axis, somatotropin has been the most controversial over the past decade. Its virtues were first extolled in demonstrating reduced catabolism in patients with burns (17), sepsis (18), trauma (19), previous surgery (20), and critical illness (21). It has been demonstrated to improve graft healing (22, 23), preserve muscle mass and grip strength (24), and improve weaning from ventilation (25). These results were found using supranormal continuous intravenous dosing of growth hormone (GH). Even with the above benefits, however, decreases in mortality, days on the ventilator, and days in the ICU were difficult to prove. Much of the GH debate was put to rest in 1999 when Takala et al. (26) reported the results of a randomized, double-blinded, prospective trial that showed significant increase in mortality with administration of GH. Since this trial, understandably, much skepticism surrounds medical intervention in the somatotropin axis.

GH is secreted from the pituitary gland during normal activity in a pulsatile manner. Its release is augmented by GH releasing hormone and attenuated principally by somatostatin (27, 28). Of note, some peripheral stimulators are dopamine, α- and β-agonists and high protein meals, malnourishment, and insulin-induced hypoglycemia. Peripheral inhibitors include glucocorticoid excess, hypoglycemia, leptin, and β-agonists. These demonstrate the complexity of GH regulation and represent the forces at play in critical illness. Nearly all of these factors are seen in the critically ill, particularly glucocorticoid excess, beta and alpha agonism, and malnourishment. When GH is allowed to act fully, it is known to directly increase lipolysis and lipid oxidation, antagonize insulin, and indirectly stimulate protein synthesis. GH, however, in protracted critical illness, does not act accordingly.

After trauma, GH is typically elevated. The normal pulsatile flow of GH is characterized by undetectable troughs, yet in acute insult, that trough is summarily raised to a detectable and typically constant level (29, 30). Although the peaks do not increase, the total amount of excreted hormone is increased. The causes of this are suspected to be immune in nature or perhaps nutritionally stimulated (31, 32), although this has yet to be made clear. It seems that overall GH retains its lipolytic and insulin antagonistic effect while losing its anabolic properties (33, 34). This is in accordance with the acute need to mobilize building blocks to aid in manufacturing acute phase reactants and immune modulators.

The chronic phase is somewhat different. The pulsatile phase recedes to reveal a tonic subnormal GH activity. Whereas the acute phase was characterized by GH resistance (elevated GH with limited GH activity), the chronic phase demonstrates more hypothalamic inactivity (33, 34). This has been shown true with work with GH secretagogues (35). Pulsatile GH activity was shown to return with the administration of GH releasing hormone (36, 37), demonstrating hypothalamic inactivity instead of presupposed GH resistance. Grehlin, a newly discovered biological GH releasing hormone analog produced in the stomach, has been shown to be a more potent secretagogue than GH releasing hormone, further solidifying a link between nutritional stimuli and GH activity (35). These late discoveries shed light onto controversy surrounding GH. It is possible that by using only high-dose GH to elicit anabolism instead of restoring natural pulsatile GH activity, the benefit of GH was subverted. Moreover, the acute and chronic phases of critical illness are not differentiated in many previous studies. By administering GH during the acute phase of critical illness, marked by continued pulsatile activity, the effects are unknown and possibly detrimental.

Insulin-Like Growth Factor

As intimated previously, insulin growth factor (IGF) should be included with any discussion of GH. Its involvement in the metabolic processes of the critically ill continues to evolve. IGF-1, also known as somatomedin, is an 8 kD, single-chain polypeptide with a structure similar to proinsulin. IGF-1 is produced by hepatocytes in response to GH stimulation. Many of the effects of GH have been attributed to IGF-1 (38). When IGF-1 and GH are given concurrently, the metabolic consequences are greater than if these two agents are given individually. This is explained by GH's ability to sensitize target cells to IGF-1 and stimulate hepatocytes to produce IGF-1's key binding protein, IGF binding protein-3 (IGFBP3) (39).

IGF-1's mechanisms for promoting a beneficial metabolic profile include promoting protein synthesis and attenuating proteolysis. During conditions of catabolism, IGF-1 infusion stimulates the uptake of amino acids and promotes the assembly of these amino acids into proteins (40). A unique and beneficial property of IGF-1 is its ability to bind to both its own receptor and the insulin receptor. This feature allows the uptake of insulin and improved glucose oxidation in patients who are typically insulin resistant (41). In addition, IGF-1 stimulates cellular differentiation and cell division (40, 42).

Significant loss of lean muscle mass is a common feature after significant burn injury, which results in loss of physical strength, decreased resistance training, and diminished aerobic capabilities. Patients are predisposed to an increased rate of hospital-acquired infections and prolonged duration of mechanical ventilation. Administration of IGF-1 reduces this catabolic response after injury (43–45). Cioffi and Rue (43) documented a reduction in protein oxidation in burn patients treated with IGF-1. IGF-1 administration, both *in vivo* and *in vitro*, prevented the catabolic responses associated with thermal injury (46, 47). This IGF-1–mediated reduction in catabolism occurs by inhibiting lysosomal and ubiq-

uitin-proteasome–dependent mediated pathways of protein degradation (48).

IGF-1 given alone rapidly loses it effectiveness. IGF-1 has six binding proteins. The most relevant of these binding proteins is IGFBP3. IGF-1, when bound to IGFBP3, has a prolonged half-life and increased biological effectiveness (41). Several investigators have demonstrated improved protein balance in catabolic burn patients after administration of IGF-1/IGFBP3 complexes (41,49).

Considering these recent developments, IGF-1's role in intervention is yet to be clearly defined, but it plays an essential role in decreasing catabolism in concert with GH. Future trials must demonstrate its effectiveness in the acute vs. prolonged phase of critical illness.

Insulin

Recent developments concerning insulin demand its inclusion in a discussion of the metabolic processes of the critically ill. It has previously been viewed as a metabolic force only. Not only has its significance burgeoned over time, but its scope as an anti-inflammatory agent has as well (50). Insulin is a pancreas-derived hormone that regulates the uptake of glucose by adipose and muscle tissues. During periods of stress, the liver's ability to generate glucose, gluconeogenesis, is under the influence of insulin. Hyperglycemia is commonly seen in critical illness or after serious injury in patients without any history of diabetes mellitus (48, 51, 52). Insulin resistance is characterized by elevated levels of: serum glucose free fatty acids, glycerol, and lactate levels. The impaired actions of insulin are a result of decreased insulin-stimulated phosphorylation of the insulin receptor and many of the secondary messengers (53). As alluded to previously, insulin's ability to regulate blood glucose levels has been overshadowed by some fascinating and important anabolic and anti-inflammatory properties.

Multiple investigations in normal, human volunteers have demonstrated that insulin possesses anabolic properties (54–56). Insulin promotes the uptake of amino acids and synthesis of new proteins (34). The administration of insulin induces these beneficial metabolic changes with only minimal hypoglycemic response (30). In addition to augmenting anabolism, researchers have determined that the administration of insulin will decrease the rate of proteolysis (57, 58).

The exciting recent observations by Van den Berghe et al. (59) demonstrated that intensive insulin therapy resulted in significant decreases in mortality and morbidity in critically ill patients maintained on mechanical ventilatory support. Insulin therapy with maintenance of plasma glucose levels between 80 and 100 mg/dL resulted in a near 50% drop in mortality (from 8.0% to 4.6%) compared with patients whose blood glucose was maintained at 180–200 mg/dL. Intensive insulin therapy also reduced mortality in patients with bacteremia from 29.5% to 12.5%, and it also reduced the frequency of episodes of sepsis by 46%.

Infusion of insulin improves outcomes in cardiac patients. Both the American College of Cardiology and the American Heart Association have recommended a regiment of glucose, insulin, and potassium, also known as GIK therapy, to patients with acute myocardial ischemia, especially for patients who are high risk for anticoagulation or thrombolytics (53). Both insulin and glucose inhibit the effects of TNFα and production of migration inhibitory factor in a dose-dependent fashion (60–62). In models of cardiac ischemia, glucose, insulin, and potassium may prevent coronary artery thrombosis by enhancing the activity of nitric oxide synthesis (49). Insulin increases the production of NO that subsequently promotes vasodilatation and decreased platelet aggregation.

The mechanism of this protective effect has not been well identified. In animal studies, treatment with insulin resulted in beneficial changes in leukocyte adherence and migration (43, 42) and could inhibit TNFα production and the proinflammatory effects of TNFα in a dose-dependent manner (63). Nuclear factor-κβ is the principle mediator of TNFα induced inflammation. TNFα and IL-1 produce a breakdown of inhibitor factor-κβ, and a subsequent increase in nuclear factor-κβ. Nuclear factor-κβ is the mediator responsible for the proinflammatory actions attributed to TNFα. Insulin acts as an anti-inflammatory molecule by decreasing nuclear factor-κβ in mononuclear cells and increasing inhibitor factor-κβ. These molecular changes result in a net decrease in the production of oxygen free-radicals (64) and to stimulate endothelial NO synthesis (65) Insulin also suppresses the production of the pro-inflammatory molecule macrophage migration inhibitory factor. Once the in-

sulin is removed, the anti-inflammatory changes revert back to normal (66).

Hyperglycemia has also been implicated in the pathogenesis of the inflammatory responses. For example, exposure of monocytes to high plasma glucose levels induced IL-6 and TNFα expression and secretion (67), and the alteration of the redox status in muscle cells by decreasing the expression of the rate-limiting enzyme of glutathione synthesis, γ-glutamylcystein synthetase (68). In humans, hyperglycemia is known to impair the phagocytic function of neutrophils, and this might be attributed to induction of a monocyte adhesion molecule, mac-1 (69). Irrespective of the etiology, the study by Van den Berghe et al. (59) attests to the importance of tight control of plasma glucose levels in the critically ill patient. Additional studies to determine the optimal doses of insulin used and the optimal blood glucose levels to be achieved are needed.

Neuroimmunomodulation

This concept was briefly discussed in the introduction but has, during the last 5 yrs, received more acclaim and, in turn, deserves more profound exposure. The endocrine system is known to interact with the immune system, but the extent to which this happens is much more than at first apparent. The hypothalamic-pituitary axis reacts to systemic inflammatory mediators released during initial stress. One of the effects of this reaction is to increase systemic corticosteroids. This in effect mitigates the immune response so that the entire body is not mutilated in its wake. However, the interaction between the neurologic and immune system is quite complex. The major immunoregulatory organs have immense autonomic innervations (70). Animals with known hypothalamic deficiency are more susceptible to chronic immunologic insults like arthritis. Intraventricular injection of endotoxin inhibits function of systemic immunologic machinery (71). Deficiency of GH leads to atrophic thymus that is reversed by administration of GH (72).

Most of the previous discourse about neuroimmunomodulation concerns humoral interaction between the neuroendocrine framework and the peripheral immune system. A second route of exchange has been demonstrated by direct neural-catecholaminergic stimulation (73). In the acute phase of stress re-

> *M*ore work is needed to answer questions about catabolism, cachexia in the face of adequate nutrition, and stunted immune response, but we believe this review begins to shed light on these problems.

sponse, increased sympathetic outflow stimulates a surge of catecholamines, increasing as much as 10-fold (74, 75). They are a well-known participant in acute stress response and help to increase cardiac output (76), increase basal energy expenditure (77) and attenuate normal anabolic activity, and increase breakdown of skeletal muscle (78, 79) for the manufacturing of acute-phase proteins. Herndon et al. (80, 81) demonstrated that beta blockade can interfere with long-term catabolism of severely burned patients and decreasing heart rate (82, 83). In addition, norepinephrine and epinephrine have been demonstrated in animals and human models to have immune capabilities by enhancing expression of immune mediators and exhibiting effects on T-helper cells (84–90).

Sympathetic activity, primarily because of its association with the stress response, has received most of the attention from researchers, but parasympathetic activity during stress has recently been discovered to attenuate the stress response as well. Afferent activity has been demonstrated to enhance cytokine to brain activation of the immune response (91–93). Efferent activity has also been shown to decrease systemic levels of TNFα and prevent lethal hypotension (94). Acetylcholine, just like catecholamines, has immuno-active capabilities. It actively inhibits release of inflammatory interleukins like IL-6 and IL-18, but not IL-10, which is an anti-inflammatory protein (94). In addition, studies show that nicotine might be an effective tool in fighting inflammatory bowel disease, and there is less inflammatory activity in the bowel mucosa of smokers (95–97). The real impact of sympathetic and parasympathetic innervation to the immune response is yet to be illuminated, but there is ample activity trying to do so.

Even less is known about the long-term effects of autonomic stimulation during a stress response. Because of the difficulty of studying long-term critical illness in both human and animal models, it is easy to see why there is more information about the acute phase. Some have generally postulated that continued stimulation of reactionary systems by long-term stress results in an attenuated response (98, 99). This has been demonstrated in lifetime stress and aging (99), but diminished activity of the autonomic nervous system during critical illness has yet to be shown.

Critical illness continues to present challenges, even when therapies improve. If physicians were not successful in treating initial insults, protracted critical illness would not be an issue. It is previously mentioned experimentation that differentiates acute from chronic illness and the array of metabolic processes involved. More work is needed to answer questions about catabolism, cachexia in the face of adequate nutrition, and stunted immune response, but we believe this review begins to shed light on these problems.

REFERENCES

1. Steinhoff M, Vergnolle N, Young SH: Agonists of proteinase-activated receptor 2 induce inflammation by a neurogenic mechanism. *Nat Med* 2000; 6:151–158
2. Basu S, Srivastava PK: Heat shock proteins: The fountainhead of innate and adaptive immune responses. *Cell Stress Chaperones* 2000; 5:443–451
3. Scaffidi P, Misteli T, Bianchi ME: Release of chromatin protein HMGB1 by necrotic cells triggers inflammation. *Nature* 2002; 418:191–195
4. Carp H: Mitochondrial N-formylmethionyl proteins as chemoattractants for neutrophils. *J Exp Med* 1982; 155:264–275
5. Tracey KJ: Anti-cachectin/TNF monoclonal antibodies prevent septic shock during lethal bacteraemia. *Nature* 1987; 330:662–664
6. Tracey KJ: Shock and tissue injury induced by recombinant human cachectin. *Science* 1986; 234:470–474
7. Carl Nathan: Points of control in inflammation. *Nature* 2002; 420:846–852
8. Tsunawaki S, Sporn M, Ding A, et al: Deactivation of macrophages by transforming growth factor-α. *Nature* 1988; 334:260–262
9. Bertini R, Bianchi M, Ghezzi P: Adrenalectomy sensitizes mice to the lethal effects of interleukin 1 and tumor necrosis factor. *J Exp Med* 1988; 167:1708–1712
10. Butler LD: Neuroendocrine regulation of *in vivo* cytokine production and effects: I. *In vivo* regulatory networks involving the neuroendocrine system, interleukin-1 and tumor necrosis factor-a. *J Neuroimmunol* 1989; 24:143–153
11. Bloom O: Hypophysectomy, high tumor necrosis factor levels, and hemoglobinemia in lethal endotoxemic shock. *Shock* 1998; 10:395–400
12. Bellinger DL, Lorton D, Lubahn C: Psychoneuroimmunology. San Diego, Academic, 2001, pp 55–112
13. Kevin J: The inflammatory reflex. *Nature* 2002; 420:853–859
14. Borovikova LV, Ivanova S, Zhang M, et al: Differential effects of hemorrhage and LPS on tissue TNF-alpha, IL-1 and associated neurohormonal and opioid alterations. *Science* 2000; 66:399–409
15. Sato KZ: Diversity of mRNA expression for muscarinic acetylcholine receptor subtypes and neuronal nicotinic acetylcholine receptor subunits in human mononuclear leukocytes and leukemic cell lines. *Neurosci Lett* 1999; 266:17–20
16. Sato E, Koyama S, Okubo Y, et al: Acetylcholine stimulates alveolar macrophages to release inflammatory cell chemotactic activity. *Am J Physiol* 1998; 274:L970–L979
17. Gore DC, Honeycutt D, Jahoor F, et al: Effect of exogenous growth hormone on whole-body and isolated-limb protein kinetics in burned patients. *Arch Surg* 1991; 126:38–44
18. Voerman BJ, van Schijndel RJM, Groenveld ABJ, et al: Effects of recombinant human growth hormone in patients with severe sepsis. *Ann Surg* 1992; 216:648–655
19. Jeevanandam M, Ali MR, Holaday NJ, et al: Adjuvant recombinant human growth hormone normalizes plasma amino acids in parenterally fed trauma patients. *JPEN J Parenter Enteral Nutr* 1995; 19:137–144
20. Ponting GA, Halliday D, Teae JD, et al: Postoperative positive nitrogen balance with intravenous hyponutrition and growth hormone. *Lancet* 1988; 1:438–440
21. Voerman BJ, Strack van Schijndel RJM, Groeneveld AB, et al: A prospective randomized, placebo-controlled trial. *Crit Care Med* 1995; 23:665–673
22. Gilpin DA, Barrow RE, Rutan RL, et al: Recombinant human growth hormone accelerates wound healing in children with large cutaneous burns. *Ann Surg* 1994; 220:19–24
23. Herndon DN, Hawkins HK, Nguyen TT, et al: Characterization of growth hormone enhanced donor site healing in patients with large cutaneous burns. *Ann Surg* 1995; 221:649–659
24. Jiang ZM, He GZ, Zhang SY, et al: Low-dose growth hormone and hypocaloric nutrition attenuate the protein-catabolic response after major operation. *Ann Surg* 1989; 210:513–525
25. Knox JB, Wilmore DW, Demling RH, et al:

Use of growth hormone for postoperative respiratory failure. *Am J Surg* 1996; 171: 576–580

26. Takala J, Ruokonen E, Webster N, et al: Increased mortality associated with growth hormone treatment in critically ill adults. *N Engl J Med* 1999; 341:785–792

27. Hartman ML, Veldhuis JD, Thorner MO: Normal control growth hormone secretion. *Horm Res* 1993; 40:37

28. Wehrenberg WB, Janowski BA, Piering AW, et al: Glucocorticoids: Potent inhibitors and stimulators of growth hormone secretion. *Endocrinology* 1990; 126:3200

29. Noel GL, Suh HK, Stone SJG, et al: Human prolactin and growth hormone release during surgery and other conditions of stress. *J Clin Endocrinol Metab* 1972; 35:840–851

30. Ross R, Miell J, Freeman E, et al: Critically ill patients have high basal growth hormone levels with attenuated oscillatory activity associated with low levels of insulin-like growth factor-1. *Clin Endocrinol* 1991; 35: 47–54

31. Hartman ML, Veldhuis JD, Johnson ML, et al: Augmented growth hormone secretory burst frequency and amplitude mediate enhanced GH secretion during a two day fast in normal men. *J Clin Endocrinol Metab* 1992; 74: 757–765

32. Thissen JP, Ketelslegers JM, Underwood LE: Nutritional regulation of insulin-like growth factors. *Endocr Rev* 1994; 15:80–101

33. Van den Berghe G, de Zegher F, Velduis J, et al: The somatropic axis in critical illness: Effect of continuous growth hormone-releasing hormone and growth hormone-releasing peptide-2 infusion. *J Clin Endocrinol Metab* 1997; 82:590–599

34. Van den Berghe G, de Zegher, Bouillon R: Acute and prolonged critical illness as different neuroendocrine paradigms. *J Clin Endocrinol Metab* 1998; 83:1827–1834

35. Inui A: Ghrelin: An orexigenic and somatotrophic signal from the stomach. *Nature* 2001; 2:1–10

36. Van den Berghe G, de Zegher F, Bowers CY, et al: Pituitary responsiveness to growth hormone releasing hormone, GH-releasing peptide-2 and thyrotropin releasing hormone in critical illness. *Clin Endocrinol* 1996; 45: 341–351

37. Van den Berghe G, Wouters P, Weekers F, et al: Reactivation of pituitary hormone release and metabolic improvement by infusion of growth hormone-releasing peptide and thyrotropin-releasing hormone in patients with protracted critical illness. *J Clin Endocrinol Metab* 1999; 84:1311–1323

38. Schmid CM, Binz JZ: The Endocrine/Paracrine Actions of IGFs on Bone. Modern Concepts of Insulin-Growth Factors. New York, Elsevier, 1991, pp 129–141

39. Meyer NA, Herndon DN: Combined insulin-like growth factor-1 and growth hormone improved weight loss and wound healing in burned rats. *J Trauma* 1996; 41:1008–1012

40. Lieberman SA, Harrison D, Hoffman AR: Anabolic effects of recombinant human insulin like growth factor-I in cachectic patients with acquired immunodeficiency syndrome. *J Clin Endocrinol Metab* 1994; 78:404–410

41. Debroy MA, Zhang XJ, Chinkes DL, et al: Anabolic effects of insulin-like growth factor in combination with insulin-like growth factor binding protein-3 in severely burned adults. *J Trauma* 1996; 47:904–910

42. Froesch ER, Schmid C, Zapf J: Metabolic and growth effects of IGF-1 in vitro and *in vivo*. *Dev Endocrinol* 1990; 67:141–153

43. Cioffi WG, Rue LW: Insulin-like growth factor-I lowers protein oxidation in patients with thermal injury. *Ann Surg* 1994; 220: 310–317

44. Fang CH, Wang JJ, Fischer JE, et al: Treatment of rats with insulin-like growth factor I inhibits the catabolic response in skeletal muscle following burn injury. *Am J Physiol* 1998; 275:R1091–R1098

45. Fang CH, Wray CJ, Hasselgren PO: Insulin-like growth factor-I inhibits lissome and proteasome-dependent proteolysis in skeletal muscle after burn. *J Burn Care Rehabil* 2002; 23:318–325

46. Ferrando AA, Wolf SE, Martin S, et al: A submaximal dose of insulin promotes net skeletal muscle protein synthesis in patients with severe burns. *Ann Surg* 1999; 229: 11–18

47. Herndon DN, Kunkle KR, Broemeling L, et al: Effects of recombinant growth hormone on donor-site healing in severely burned children. *Ann Surg* 1990; 212:424–429

48. Wolfe RR, Jahoor F, Miyoshi H, et al: Effect of severe burn injury on substrate cycling by glucose and fatty acids. *N Engl J Med* 1987; 317:403

49. Herndon DN, DebRoy MA, Zehng M, et al: Muscle protein catabolism after severe burns: Effects IGF-1/IGFBP-3 treatment. *Ann Surg* 1999; 229:713–720

50. Das UN: Is insulin an anti-inflammatory molecule? *Nutrition* 2001, 17:409–413

51. Wolfe RR, Burke JF: Glucose metabolism in man: Responses to intravenous glucose infusion. *Metabolism* 1979; 28:210–217

52. Shangraw RE, Miyoshi H: Differentiation between septic and post burn insulin resistance. *Metabolism* 1989; 38:983–991

53. Ryan TJ, Brooks NH: Update: ACC/AHA guidelines for the management of patients with acute myocardial infarction: Executive summary and recommendations. A report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines. *Circulation* 1999; 100:1016

54. Bennet WM, Scrimgeour CM: Euglycemic hyperinsulinemia augments amino acid uptake by human leg tissue during hyperaminoacidemia. *Am J Physiol* 1990; 259: E185–E194

55. Biolo G, Wolfe RR: Physiologic hyperinsulinemia stimulates protein synthesis and enhances transport of select amino acids in human skeletal muscle. *J Clin Invest* 1995; 95:811–819

56. Fryburg DA, Hill SA: Insulin and insulin-like growth factor-I enhance human skeletal muscle protein anabolism during hyperaminoacidemia by different mechanisms. *J Clin Invest*, 1995; 96:1722–1729

57. Abumrad NN, Jefferson LS, Rannels SR, et al: Role of insulin in the regulation of leucine kinetics in the conscious dog. *J Clin Invest* 1982; 70:1031–1041

58. Flakoll PJ, Kulaylat M, Frexes-Steed M, et al: Amino acids augment insulin's suppression of whole body proteolysis. *Am J Physiol* 1989; 257:E839–E847

59. Van den Berghe G, Wouters P, Weekers F, et al: Intensive insulin therapy in critically ill patients. *N Engl J Med* 2001; 345:1359–67

60. Herndon D, Hart D, Wolf S, et al: Reversal of catabolism by beta-blockade after severe burns. *N Engl J Med* 2001; 345:1223–1229

61. Herndon DN, Barrow RE, Rutan TC, et al: Effect of propranolol administration on hemodynamic and metabolic responses of burned pediatric patients. *Ann Surg* 1988; 208:494–482

62. Sinha AK, Acharya K, Mazumdar S: Stimulation of nitric oxide synthesis and protective role of insulin in acute thrombosis *in vivo*. *Life Sci* 1999; 65:2687

63. Satomi N, Haranaka K: Relationship of hypoglycemia to tumor necrosis factor production and antitumor activity: Role of glucose, insulin, and macrophages. *J Natl Cancer Inst* 1985; 74:1255

64. Dandona P, Mohanty P: Insulin inhibits intranuclear factor kappa-β and stimulates I kappa-β in mononuclear cells in obese subjects: Evidence for an anti-inflammatory effect? *J Clin Endocrinol Metab* 2001; 86:3257

65. Das UN: Possible beneficial action(s) of glucose-insulin-potassium regimen in acute myocardial infarction and inflammatory conditions: A hypothesis. *Diabetologia* 2000, 43: 1081–1082

66. Fraker DL, Norton JA: Reversal of the toxic effects of cachectin by concurrent insulin administration. *Am J Physiol* 1989; 2566: E725

67. Morohoshi M, Fujisawa K, Uchimura I, et al: Glucose-dependent interleukin 6 and tumor necrosis factor production by human peripheral blood monocytes *in vitro*. *Diabetes* 1996; 45:954–959

68. Powell LA, Nally SM, McMaster D, et al: Restoration of glutathione levels in vascular smooth muscle cells exposed to high glucose conditions. *Free Radic Biol Med* 2001; 31: 1149–1155

69. Sampson MJ, Davies IR, Brown JC, et al: Monocyte and neutrophil adhesion molecule expression during acute hyperglycemia and after antioxidant treatment in type 2 diabetes and control patients. *Arterioscler Thromb Vasc Biol* 2002; 22:1187–1193

70. Felten DL, Felten SY, Carlson SL, et al: Noradrenergic and peptidergic innervation of lymphoid tissue. *J Immunol* 1985; 135(Suppl):755s–765s

71. Sternberg EM, Young WS III, Bernadini R, et

al: A central nervous system defect in biosynthesis of corticotropin-releasing hormone is associated with susceptibility to streptococcal cell wall-induced arthritis in Lewis rats. *Proc Nat Acad Sci U S A* 1989; 86:4771–4775

72. Weigent DA, Blalock JE: Growth hormone and the immune system. *Prog Neuroendocrinimmunol* 1990; 3:231–241

73. Reichlin S: Neuroendocrine-immune interactions *N Engl J Med* 1993; 329:1246–1253

74. Goodall MC, Stone C Haynes BW Jr: Urinary output of adrenaline and noradrenaline in severe thermal burns. *Ann Surg* 1957; 145:479–487

75. Wilmore DW, Aulick LH: Metabolic changes in burned patients. *Surg Clin North Am* 1978; 58:1173–1187

76. Asch MJ, Feldman RJ, Walker HL, et al: Systemic and pulmonary hemodynamic changes accompanying thermal injury. *Ann Surg* 1973; 178:218–221

77. Reiss W, Pearson E, Artz CP: The metabolic response to burns. *J Clin Invest* 1956; 35:62–77

78. Newsome TW, Mason AD Jr, Pruitt BA Jr: Weight loss following thermal injury. *Ann Surg* 1973; 178:215–217

79. Hart DW, Wolf SE, Mlcak RP, et al: Persistence of muscle catabolism after severe burn. *Surgery* 2000; 128:312–319

80. Herndon D, Hart D, Wolf S, et al: Reversal of catabolism by beta-blockade after severe burns. *N Engl J Med* 2001; 345:1223–1229

81. Herndon DN, Barrow RE, Rutan TC, et al: Effect of propranolol administration on hemodynamic and metabolic responses of burned pediatric patients. *Ann Surg* 1988; 208:494–482

82. Minifee PK, Barrow RE, Abston S, et al: Improved myocardial oxygen utilization following propranolol infusion in adolescents with post burn hyper metabolism. *J Pediatr Surg* 1989; 24:806–810

83. Baron PW, Barrow RE, Pierre EJ, et al: Prolonged use of propranolol safely decreases cardiac work in burned children. *J Burn Care Rehabil* 1997; 18:223–227

84. Panina-Bordignon P, Mazzeo D, Lucia PD, et al: Beta2-agonists prevent Th1 development by selective inhibition of interleukin-12. *J Clin Invest* 1997; 100:1513–1519

85. Hasko G, Szabo C, Nemeth ZH, et al: Stimulation of beta-adrenoreceptors inhibits endotoxin-induced IL-12 production in normal and IL-10 deficient mice. *J Neuroimmunol* 1998; 88:57–61

86. Sanders VM, Baker RA, Ramer-Quinn DS, et al: Differential expression of the beta2-adrenergic receptor by TH1 and Th2 clones: Implications for cytokine production and B cell help. *J Immunol* 1997; 158:4200–4210

87. Borger P, Hoekstra Y, Esselink MT, et al: Beta-adrenoceptor-mediated inhibition of IFN-gamma, IL-3, and GM-CSF mRNA accumulation in activated human T lymphocytes is solely mediated by the beta2-adrenoceptor subtype. *Am J Respir Cell Mol Biol* 1998; 19:400–407

88. Hasko G, Elenkov IJ, Kvetan V, et al: Differential effect of selective block of alpha 2-adrenoreceptors on plasma levels of tumor necrosis factor-alpha, interleukin-6 and corticosterone induced by bacterial lipopolysaccharide in mice. *J Endocrinol* 1995; 144:457–462

89. Kovacs KJ, Elenkov IJ: Differential dependence of ACTH secretion induced by various cytokines on the integrity of the paraventricular nucleus. *J Neuroendocrinol* 1995; 7:15–23

90. Woiciechowsky CK, Asadullah K, Nestler D, et al: Sympathetic activation triggers systemic interleukin-10 release in immunodepression induced by brain injury. *Nat Med* 1998; 4:808–813

91. Fleshner M, Goehler LE, Schwartz BA, et al: Thermogenic and corticosterone responses to intravenous cytokines are attenuated by sub diaphragmatic vagotomy. *J Neuroimmunol* 1998; 86:134–141

92. Maier SF, Goehler LE, Fleshner M, et al: The role of the vagus nerve in cytokine-to-brain communication. *Ann N Y Acad Sci* 1998; 840:289–300

93. Watkins LR, Goehler LE, Relton JK, et al: Blockade of interleukin-1 induced hyperthermia by sub diaphragmatic vagotomy: Evidence for vagal mediation of immune-brain communication. *Neurosci Lett* 1995; 183:27–31

94. Borovikova LV, Ivanova S, Zhang M, et al: Vagus nerve stimulation attenuates the systemic inflammatory response to endotoxin. *Nature* 2000; 405:458–462

95. Guslandi M: Nicotine treatment for ulcerative colitis. *Br J Physiol* 1998; 274:L970–L979

96. Sandborn WJ: Transdermal nicotine for mildly to moderately active ulcerative colitis: A randomized, double-blind, placebo-controlled trial. *Ann Intern Med* 1997; 126:364–371

97. Sher ME: The influence of cigarette smoking on cytokine levels in patients with inflammatory bowel disease. *Inflamm Bowel Dis* 1999; 5:73–78

98. McEwen B: Protective and damaging effects of stress mediators. *N Engl J Med* 1998; 338:171–179

99. Seeman TE, Robbins RJ: Aging and hypothalamic-pituitary-adrenal response to challenge in humans. *Endocr Rev* 1994; 15:233–260