

# Adrenocortical Stress Response during the Course of Critical Illness

Bram Peeters,<sup>1</sup> Lies Langouche,<sup>1</sup> and Greet Van den Berghe<sup>\*1</sup>

## ABSTRACT

Critically ill patients have elevated plasma cortisol concentrations, in proportion to illness severity. This was traditionally attributed exclusively to a central activation of the hypothalamus-pituitary axis. However, low rather than high plasma ACTH concentrations have been reported in critically ill patients, with loss of diurnal ACTH and cortisol rhythm. Low ACTH together with high cortisol is referred to as "ACTH-cortisol dissociation." Although cortisol production is somewhat increased with inflammation, a reduced cortisol breakdown explains to a larger extent the hypercortisolism during critical illness. Inflammation-driven decrease in cortisol binding proteins further increase the active free cortisol fraction. Several drugs administered to ICU patients suppress plasma cortisol in a dose-dependent manner.

Sustained low circulating ACTH might contribute to adrenal atrophy and dysfunction in the prolonged phase of critical illness. In the acute phase of sepsis or septic shock, a condition referred to as "relative adrenal insufficiency" has been suggested to ensue from glucocorticoid resistance and insufficiently elevated circulating cortisol to overcome such resistance, with pathological changes possibly occurring at every level of the HPA axis. However, it remains highly controversial whether tissue-specific glucocorticoid resistance is adaptive or maladaptive, how to diagnose "relative" adrenal insufficiency, and how it should be treated. Large RCTs, investigating the effect of 200 mg/d hydrocortisone treatment for sepsis or septic shock have shown conflicting, mainly negative, results. Not taking into account the reduced cortisol breakdown, which increases the risk of overdosing hydrocortisone, might have played a role. Further research on diagnostic, therapeutic and dosing aspects is urgently warranted. © 2018 American Physiological Society. *Compr Physiol* 8:283-298, 2018.

## Didactic Synopsis

### Major teaching points

- Unlike the hypothalamus-pituitary-adrenal axis response to stress outside the context of intensive care, the stress response to critical illness is hallmarked by low rather than high plasma ACTH in the face of high plasma cortisol.
- During critical illness, the diurnal rhythm of ACTH and cortisol secretion is absent.
- A normal or only slightly increased cortisol production and a consistently reduced cortisol breakdown determine the degree of hypercortisolism during critical illness.
- Sustained suppressed circulating ACTH can contribute to risk of adrenal atrophy specifically in prolonged critically ill patients.
- Drugs often given to critically ill patients such as etomidate, opioids and propofol can suppress plasma cortisol. One should consider omitting these before initiating treatment with hydrocortisone for low plasma cortisol.
- It remains controversial whether "relative" adrenal insufficiency is a clinical entity ensuing from glucocorticoid resistance with cortisol availability that is insufficiently elevated to overcome such resistance.

## Introduction

Critical illness represents any condition, evoked by major surgery, severe medical illnesses, or multiple trauma, that requires pharmacological and/or mechanical support of vital organ functions without which death would ensue. As such, critical illness is a condition of severe and sustained physical stress for the human body for which an adequate activation of several processes is required to provide necessary energy, to modulate the immune response, and to ensure hemodynamic homeostasis. The term 'stress response' indicates the combination of these closely interrelated physiological reactions to stress to maintain and restore homeostasis in the human body (137). Both neuronal and endocrine systems are involved, among which the activation of the sympathetic nervous system, the release of catecholamines from the adrenal

\* Correspondence to greet.vandenbergh@kuleuven.be

<sup>1</sup> Clinical Division and Laboratory of Intensive Care Medicine, Department of Cellular and Molecular Medicine, KU Leuven, Belgium  
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medulla, and the activation of the hypothalamus-pituitary-adrenal (HPA) axis to increase the availability of the stress hormone cortisol (32, 144).

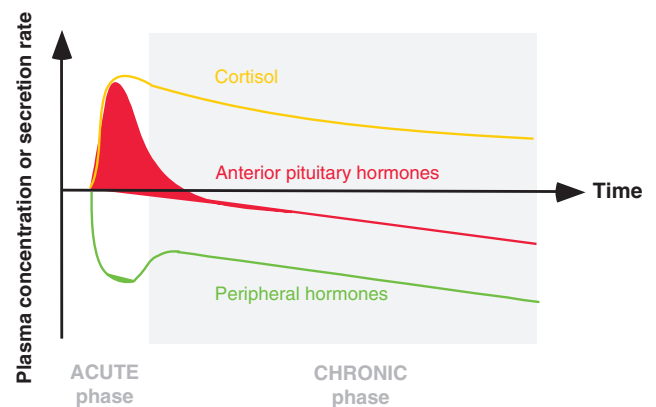
Critically ill patients indeed typically present with elevated plasma cortisol concentrations, in proportion to the severity of illness (177). Whereas this elevation in cortisol traditionally has been attributed to a central activation of the HPA axis with an increased ACTH release from the pituitary, low rather than high plasma ACTH concentrations have been reported in critically ill patients (20, 169). Low ACTH together with high cortisol is referred to as “ACTH-cortisol dissociation.” ACTH-independent mechanisms, among which direct inflammation-induced adrenocortical cortisol synthesis and release, a reduced cortisol breakdown as well as iatrogenic modulators are considered to explain this dissociation (20, 115). Recent findings have shed new light on the HPA-axis response to critical illness, with potentially important diagnostic and therapeutic implications.

In this article, we will give an overview of the adrenocortical stress response to stress, with a point-by-point discussion on the causes and consequences observed during critical illness. Next, we will discuss potential failure or dysfunction of the HPA axis, and current diagnostic and therapeutic options.

## Acute and Prolonged Critical Illness

Most patients admitted to the ICU only require a few days of intensive care, but about 25% of ICU patients receive vital organ support for a much longer period. This stage of prolonged critical illness is characterized by ongoing mechanical and pharmacological vital organ support with increased risk of organ failure and a higher risk of death. Indeed, a recent US population-based cohort study indicated an in-hospital mortality of 31% for patients with an ICU stay of at least 8 days (80). The exact timing of the transition from acute to chronic critical illness is however not clear, neither at the patient level, nor at the population level. A recent study defined this onset as the time at which severity of illness on admission was no longer predictive of mortality, which was after about 10 days (77).

Most hypothalamus-pituitary-peripheral-hormonal axes that play a key role in the metabolic and immunological alterations accompanying critical illness typically follow a biphasic response pattern (Fig. 1) (159-163, 166). For example, in the acute phase of critical illness, plasma concentrations of the anterior pituitary hormones growth hormone (GH) and thyroid-stimulating hormone (TSH) are increased, whereas plasma concentrations of their peripheral effector hormones IGF-1 and T3 are decreased. However, when ICU-dependency continues beyond the acute time window, these pituitary hormones are typically suppressed, with a further decrease of their peripheral hormones. Whereas the acute changes can be interpreted as beneficial, bringing about the release of endogenous fatty acids and glucose into the circulation and postponing energy consuming anabolism, the uniform suppression in



**Figure 1** Shown are the biphasic neuroendocrine responses of the anterior pituitary hormones and their peripheral hormones to acute and chronic critical illness. In the acute phase of illness the growth hormone (GH) and thyrotrophin (TSH) secretory activity is amplified (red), and adrenocorticotrophic hormone (ACTH) secretory activity is increased in some cases. Plasma concentrations of their anabolic peripheral hormones (insulin-like growth factor-I, triiodothyronine) are decreased (green), but cortisol levels are elevated (yellow). In prolonged critical illness, secretion of GH, TSH, and ACTH is consistently suppressed, with a further decrease of their peripheral hormones. Plasma cortisol levels remain high, but in some cases low plasma cortisol levels appear in the chronic phase of critical illness. (Figure was reproduced from Van den Berghe (160), with permission from *The Journal of Clinical Endocrinology and Metabolism*.)

the prolonged phase of critical illness likely participates in the general wasting syndrome, with persisting hypercatabolism, causing weakness and delayed or non-recovery from intensive care dependency (105, 149).

Such bi-phasic response also applies to the HPA axis, a crucial axis in terms of acute survival. However, the anterior pituitary hormone ACTH is only very transiently elevated, and several studies have reported lower than normal plasma ACTH from quite early after admission to the ICU throughout the ICU stay. This suppression of plasma ACTH occurs while high levels of plasma cortisol are consistently observed in most ICU patients, both in the acute and the prolonged phase of critical illness (Fig. 1). This phenomenon of low ACTH together with high cortisol is referred to as ‘ACTH-cortisol dissociation’. Whether or not this dissociation points to, or can lead to a dysfunctional HPA axis, can only be interpreted if one correctly understands the pathophysiology of the HPA axis response during critical illness.

## The Adrenocortical Stress Response to Critical Illness

### The normal stress response

When the human brain senses a stressful event, it signals the paraventricular nucleus (PVN) of the hypothalamus to release corticotropin-releasing hormone (CRH) and arginine vasopressin, which both activate the anterior pituitary gland

to release ACTH. In turn, ACTH release exerts important functions on the cortex of the adrenal gland which blood samples should be collected (on ice) and processed (spun cold prior to assay) (96). In burn patients, plasma ACTH levels were found not to be elevated and did not show correlation with burn size (167). In patients undergoing surgery, plasma ACTH increased during the onset of the stress response with a subsequent fall, whereas plasma cortisol remained high (40). The first line response to stress is thus mediated by the sympathetic nervous system and the HPA axis synergistically interact with each other and are functionally interdependent (50).

In contrast with ACTH, an increase of its peripheral hormone cortisol is a hallmark of critical illness. Indeed, the more severely ill, and thus the higher the risk of dying, the higher plasma cortisol concentrations rise (177). In burn patients, plasma cortisol concentrations were shown to be elevated in proportion to burn size (167). In patients undergoing surgery, cortisol concentrations also reflect the degree of surgical stress (31, 106, 108, 177). In addition, septic shock induces

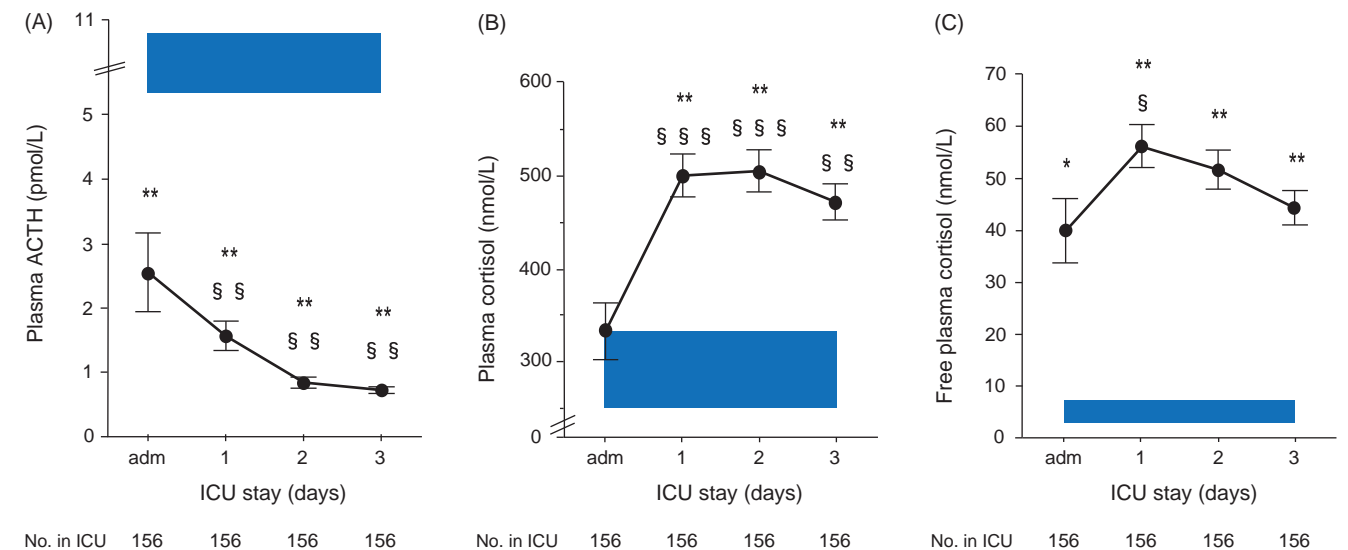


Figure 2 Mean values and standard errors for plasma ACTH (Panel A), total cortisol (Panel B), and free cortisol (Panel C) in ICU patients from admission onward until day 3 of ICU stay. The blue shaded area represents the interquartile range of morning values in healthy control subjects. \* P < 0.05, \*\* P < 0.001, for the comparison with controls. § P < 0.05, §§ P < 0.01, §§§ P < 0.0001, for the comparison of paired values of the consecutive days with the admission sample. For each day, the number of patients still in ICU is displayed below the figure. ICU denotes intensive care unit, adm denotes admission (Figure was reproduced from Peeterst(15), with permission from Clinical Endocrinology.)

elevated cortisol levels proportionally to disease severity, production rate, measured via a stable isotope technique, was found to be only slightly elevated in critically ill patients (9, 38, 104, 131).

The onset of this hypercortisolism is expected to happen instantaneously, facilitating immediate effects. However, the exact timing of the rise of plasma cortisol concentrations during the course of illness is not that clear. In patients undergoing minor or mild surgery, a cortisol increase was only observed during or near the end of surgery, with a rapid normalization during the following hours on the ward (99, 100, 158, 174). Patients undergoing elective major surgery displayed a larger cortisol response that occurred hours after, not during, surgery and remained high during the following days in ICU (59, 130). In a mixed population of severely ill medical and surgical patients, directly coming from the emergency department, the operating room or from the ward, normal cortisol plasma concentrations upon admission to the ICU were observed, which increased quickly thereafter (Fig. 2B) (115).

Hence, the dynamics of the HPA axis response to severe and prolonged life-threatening stress and to less severe stress appear to differ. Based on the general concept of the stress response, high plasma cortisol levels are predominantly attributed to an increased cortisol production. However, plasma hormone concentrations are the net result of hormone secretion, distribution, binding to plasma proteins, and plasma clearance. Hence, although cortisol production rate is not much (if not all) elevated, the reduced breakdown better explains the typically elevated plasma cortisol observed in the critically ill.

Cortisol is normally mainly broken down in the liver via A-ring reductases to the metabolites 5- and 5-tetrahydrocortisol (Fig. 3). In the kidney, cortisol can be inactivated to cortisone via 11-hydroxysteroid dehydrogenase type 2 (11-HSD2), and further degraded to tetrahydrocortisone by 5-reductase. Indeed, Boonen and colleagues showed that expression and activity of the hepatic A-ring reductases

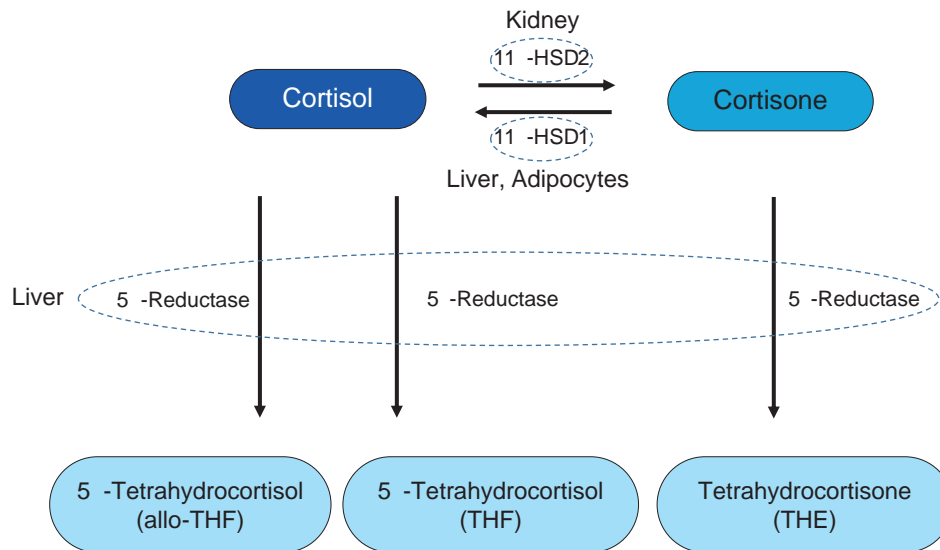


Figure 3 Cortisol metabolism in humans. Cortisol and cortisone are mainly broken down via A-ring reductases, 5-reductase and 5-reductase, in the liver to generate 5- and 5-tetrahydrocortisol. In the kidney, cortisol is metabolized by 11-hydroxysteroid dehydrogenase (11-HSD) type 2, generating cortisone, which can further be broken down to tetrahydrocortisone (THE) by 5-reductase. 11-HSD type 1 can reconvert cortisone to cortisol.

and the 11-HSD2 activity in kidney were significantly diminished in critically ill patients (20).

As a side note, the suppression of cortisol breakdown during critical illness can be interpreted as a smart adaptive and energy-efficient mechanism to rapidly increase cortisol availability in those vital organs and tissues that express these enzymes, which could be required to deal with and overcome life-threatening illnesses or trauma.

an increased ACTH sensitivity (168). This preserved dose response between ACTH and cortisol suggested that the term ACTH-cortisol dissociation (referring to total plasma concentrations) may not be entirely correct, as an association between ACTH secretion and cortisol secretion was in fact maintained, but both were lower than in healthy subjects. On the contrary, the presence of more asynchrony and irregularity in the patterns of cortisol and ACTH secretion suggested other ACTH-independent mechanisms contributing to the cortisol availability (168).

It might also be possible that critical illness induces an increase in other active splice forms of ACTH, undetected by the classic immunoenzymometric ACTH assays. However, in an observational study of septic and nonseptic ICU patients, where plasma cortisol was found to be increased in all patients, plasma ACTH was not increased, both measured by the highly specific immunoenzymometric assay as well as by a less specific single antibody competitive binding assay which would have detected other fragments or precursors of ACTH (121). These findings disproved the hypothesis that such biologically active forms of ACTH could be responsible for increased cortisol production during critical illness.

Cortisol has been interpreted as the consequence of an increased non-ACTH driven cortisol production. During critical illness, the observed moderately increased cortisol production

(20, 36, 37) could indeed theoretically be brought about by alternative stimuli, such as cytokines, neuropeptides, or certain adipokines (23). Immune cells can release cytokines that regulate cortisol secretion, but bacterial or viral pathogens can also directly interact with the adrenocortical cells via toll-like receptors (24). Toll-like-receptor-2-deficient mice had indeed an impaired adrenal corticosterone release upon stimulation with bacterial cell wall compounds, although the initial activation of cortisol production during early sepsis depended primarily on the activation of immune cells and cytokine release (81). Adipocytes situated in subcutaneous, visceral and periadrenal fat release certain adipokines, which can induce cortisol secretion but also sensitize adrenocortical cells to the release of vasoactive peptides, such as endothelin, which has shown to cause a dose-dependent stimulation of cortisol production in human and rat adrenal cells and potentiated the effect of ACTH (73).

As mentioned above, the sympathetic nervous system and the HPA axis synergistically interact with each other in the complex microenvironment of the adrenal gland (50). The addition of chromaffin cells to intact isolated perfused pig adrenals with preserved nerve supply showed that the release of corticosteroids could be stimulated through the sympathetic nervous system (22).

Also an increased ACTH sensitivity, that is, an upregulation of its melanocortin 2 receptor (MC2R), could play a role (23). However, a human study that quantified pulsatile and nonpulsatile secretion of cortisol and ACTH overnight revealed that the amount of cortisol released in response to a given ACTH level was found to be normal, arguing against

Once secreted into the bloodstream, the relatively insoluble cortisol is transported predominantly bound to corticosteroid-binding globulin (CBG, transcortin) (80%) and to a lesser extent to albumin (10%-15%). Since only free (unbound) cortisol can exert its biological and clinical effects, low CBG levels increase cortisol availability at the tissue level (117). Also CBG affinity can be modulated by pH and temperature, and by elastases produced by neutrophils at sites of inflammation, converting the high-affinity conformation of CBG to a low-affinity conformation, as such increasing free cortisol levels (69). Thus in patients with systemic infection, free rather than total cortisol correlate with the severity of disease and better reflect biologically active cortisol availability (74). In the clinical setting however, total plasma cortisol is usually measured, because ultrafiltration and equilibrium dialysis are rarely available and time-consuming. Alternatively, an estimation of free cortisol can be made with use of the validated Coolens formula, based on total plasma cortisol, CBG, and albumin levels (34).

In patients in the early stage of septic shock and multiple trauma, plasma CBG levels have shown to be immediately and significantly lowered which reflected much higher free cortisol levels than indicated by total cortisol (14). Plasma CBG levels were also transiently decreased following abdominal surgery, with a normalization on postoperative day 2 (46).

Also in a mixed population of medical and surgical ICU patients, plasma CBG and albumin were acutely downregulated already upon ICU admission, causing a several fold increase in plasma free cortisol concentrations, in the face of unaltered total plasma cortisol (Fig. 2C) (115). In this set of patients, from the morning after ICU admission, free plasma

concentrations increased further mainly due to the rise in septic patients (63). In contrast to liver GR expression, muscle total cortisol levels, and remained high during the following GR expression was not lower in patients receiving exogenous days. In an observational cohort study in patients with sepsis, glucocorticoids in tissue samples of patients who died in the ICU and septic shock, total CBG levels decreased in proportion to disease severity (109). This was explained by an increase in down-regulating effects of glucocorticoids in critical illness in cleaved low-affinity CBG, which was associated with the (116). In vitro and animal research indeed suggested increased plasma neutrophil concentration.

Elevated free plasma cortisol during severe stress in liver tissue during critical illness (4, 151). GR expression in muscle tissue, but decreased GR expression mainly determined by the combined effect of a decrease in A tissue-specific regulation of glucocorticoid signaling high-affinity CBG due to elastase cleavage and reduced CBG may limit undue cortisol exposure in vulnerable vital organs and albumin synthesis by the liver, and increased total cortisol that would suffer from an excess of cortisol and increase levels (109). In theory, cleavage of CBG can be interpreted as a beneficial response that may target increased cortisol in cells that might require more cortisol action. However, as a beneficial response that may target increased cortisol, further research regarding tissue-specific changes is needed to bioavailability to sites of interest during critical illness (110). Unravel whether this phenomenon is adaptive or maladaptive. However, a depletion of high-affinity CBG, possibly worsened by reduced synthesis, can evoke a loss of the circulating cortisol reservoir, resulting in a failure of cortisol supply to the inflammatory sites and a loss of ability to dampen inflammation (109).

Local cortisol activity is also further regulated by tissue-specific alterations of glucocorticoid signaling (29, 62). At the levels of the target cells, free cortisol diffuses the cell membrane where it can bind to the cytoplasmic glucocorticoid receptor (GR), which form dimers that translocate to the nucleus and act as a ligand-dependent transcription factor to regulate target gene expression, to exert its effects (165). Alternative splicing of the GR gene can generate different isoforms of the receptor, of which GR $\alpha$  the active positive isoform, and GR $\beta$ , the negative isoform, are the most important ones (62). Other common GR isoforms are GR $\gamma$  and GR $\delta$ . Expression of the GR receptor is normally downregulated by cortisol to maintain homeostasis (43). Cortisol also binds to the mineralocorticoid receptor (MR) with a 10-fold higher affinity (127). In contrast with the GR, which is widely expressed in all tissues, the MR is expressed only in certain tissues, such as the kidneys, where it mainly regulates salt and water homeostasis. Although affinity to cortisol is higher, cortisol signaling through the MR is limited by the activity of 11 $\beta$ -HSD2 in cells in which MR is expressed (55).

The hypothalamic-pituitary feedback regulation is central in the physiological response to maintain and restore homeostasis during stress. Cortisol exerts fast (seconds to minutes), intermediate (hours) and slow (days) feedback inhibition at the level of the hypothalamus and the pituitary to tune its own release (84). Fast feedback exerts negative feedback by inhibiting ACTH and CRH release, and does not in turn regulate target gene expression, to exert its effects (165). The hypothalamic-pituitary feedback regulation appears much more complex than the initially proposed simple closed loop system (176). For example, whereas TSH becomes completely unresponsive to the hypothalamic thyrotropin-releasing hormone when thyroid hormone levels are high (57), CRH can override the feedback inhibition on ACTH exerted by cortisol to maintain homeostasis (43). Cortisol can also bind to the mineralocorticoid receptor (MR) with a 10-fold higher affinity (127). In contrast with the GR, which is widely expressed in all tissues, the MR is expressed only in certain tissues, such as the kidneys, where it mainly regulates salt and water homeostasis. Although affinity to cortisol is higher, cortisol signaling through the MR is limited by the activity of 11 $\beta$ -HSD2 in cells in which MR is expressed (55). Evidence from animal and human studies indicate that such explaining the low plasma ACTH concentrations. This besides alternative splicing of the GR, also GR expression and GR affinity and GR translocation are regulated and could be similar to the inhibition of ACTH and CRH synthesis and secretion in response to a prolonged exposure (24 h or more) to high doses of exogenous corticosteroids (84, 121). GR expression was found to be transiently increased in white blood cells of adult septic patients (63). White blood cell binding capacity of labeled dexamethasone was markedly reduced in ventilated critically ill patients with the lowest GR receptor which suggests that increased central stress inputs might maintain some degree of ACTH secretion and partially overcome the feedback inhibition (176). In white blood cells of critically ill children, suffering from trauma and sepsis, lower total and cytoplasmic GR levels than in healthy individuals have been reported (75). A suppression of GR expression in white blood cells has also been reported in adults, however currently unclear. One could speculate that the longer

the feedback inhibition persists, the more CRH and/or ACTH characteristics. Other, yet unknown, mechanisms might have synthesis and secretion would be suppressed. However, a suppressed plasma ACTH, which may have hidden any additional progressive loss of responsiveness of the HPA axis to central pharmacological suppression on ACTH release, negative feedback regulation, probably due to degenerative changes such as negative feedback inhibition exerted by the elevated changes in the hippocampus, could also play a role (136). On the other hand, long-term administration of exogenous glucocorticoids or endogenously elevated plasma cortisol concentrations in patients with Cushing syndrome have been shown to cause tertiary, and not secondary, adrenal insufficiency. From the current available literature, it cannot be concluded whether the suppressive effect of drugs such as opioids, propofol or a single dose of etomidate on plasma cortisol is beneficial or harmful. However, when a patient and/or its higher regulatory inputs (61). Alternatively, hypoxia displays low plasma cortisol during critical illness and treatment in amputation might also directly damage the pituitary and hypothalamus with hydrocortisone is considered, it should also be thereby lower ACTH in the critically ill (21, 120). Additional well-controlled studies on central HPA axis changes over time in which case plasma cortisol should be reassessed. will be highly informative to understand illness evolution.

During surgery, at the emergency ward and during stay in the ICU, patients receive a broad variety of drugs. Importantly, many of these drugs can theoretically affect the HPA axis activity, either directly at the level of the hypothalamus, pituitary, and/or adrenal gland, or indirectly via a modulation of the activity of the sympathetic nervous system, thereby explaining at least part of the acute ACTH-cortisol dissociation, as observed already upon admission to the ICU (25, 94, 119). A well-known suppressor of adrenocortical cortisol production, by inhibiting 11-beta-hydroxylase, is etomidate (72). Prolonged etomidate infusion has been shown to be associated with an increased mortality and was therefore abandoned as a sedative from all ICUs (175). A single induction dose of etomidate, however, was not related with an increase in mortality, but still lowered plasma cortisol concentrations (26). Also opioids, frequently used as strong painkillers that act on the opioid receptor to produce morphine-like effects, have shown to result in suppressed plasma ACTH and/or cortisol concentrations when administered to healthy individuals by a multisynaptic neural pathway (27). The SCN was typically regarded as the only self-sustained clock to act as a master pacemaker for the entire organism, influenced by the light-dark cycle, physical activity, and food intake and fasting (3). Remarkably, many peripheral tissues, including endocrine glands such as the adrenal gland, are capable of generating self-sustained oscillations independently of the master SCN clock (56). Indeed, there is evidence that an intrinsic adrenocortical circadian oscillator drives the adrenal response to ACTH, defining a time window in which the cortisol response to ACTH is the highest (145). This sensitivity is also increased by the SCN, especially during the rising part of the diurnal rhythm, mediated through autonomic pathways (135). In addition, the sensitivity of the pituitary to negative feedback from cortisol, and dobutamine had a stimulatory effect, but none of these drugs independently affected plasma ACTH (115). The higher effect during the nadir of the diurnal rhythm (60). Although plasma ACTH levels are low in all ICU patients, ACTH secretion is not completely suppressed. The dynamics and interaction of cortisol and ACTH during critical illness

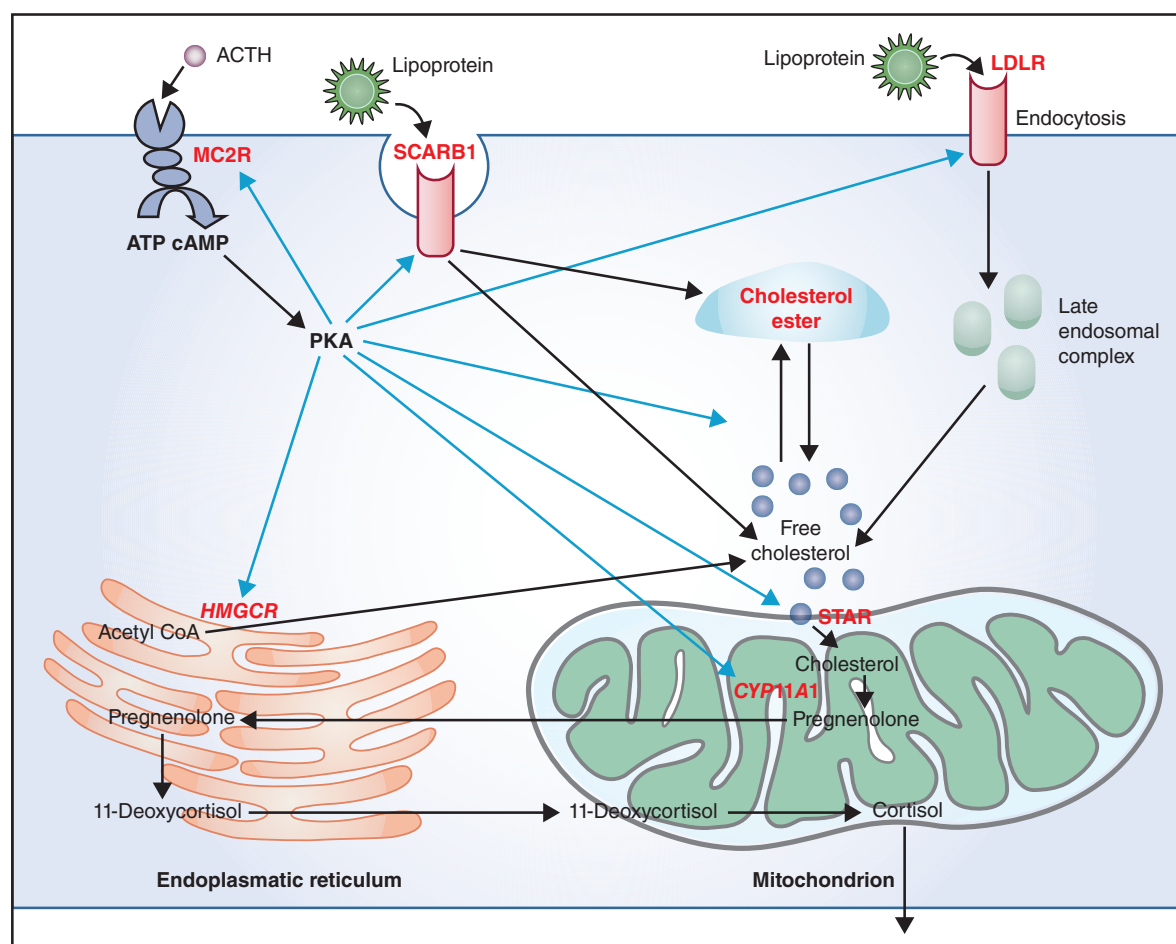
It has been generally assumed that anesthetic drugs may predominantly suppress the stress response via a central inhibition of the HPA axis and of the sympathetic nervous system (25). In a recent study of a mixed population of critically ill patients, however, it was shown that opioids, etomidate, and propofol had a suppressive effect on plasma cortisol, and dobutamine had a stimulatory effect, but none of these drugs independently affected plasma ACTH (115). The associations between these drugs and cortisol levels were independent of the medical or surgical nature of ICU admission, severity of illness, occurrence of sepsis, or other patient

have been assessed with use of repeated sampling time series and through stimulation of vascular endothelial growth of plasma levels in a mixed set of 40 surgical and medical ICU patients (53, 139). Finally, ACTH has a direct stimulatory effect on patients as compared with 8 healthy matched volunteers (19). The expression of its own receptor (MC2R) which amplifies Hormonal secretory profiles were created by deconvolution of the responsiveness to ACTH (89). The extensive acute analysis, which took into account the substantially prolonged and chronic impact of ACTH on the adrenal cortex ensures cortisol half-life, and which allowed to quantify pulsatile and normal adrenal gland structure and functioning. As such, ICU non-pulsatile secretion rates of cortisol and ACTH (168). This acquired adrenal failure could be the consequence of continuous study indicated that nocturnal ACTH as well as cortisol pulsatilely low plasma ACTH, which negatively affects the adrenal satile secretion rates were reduced in patients, attributed to the cortex. Indeed, continuously low plasma ACTH negatively reduction of pulse masses rather than a reduction of number affects the adrenal cortex, as evidenced by POMC knockout of pulses. No diurnal rhythm was present for ACTH, nor for mice, which suffer from adrenal atrophy and hypofunction cortisol, and plasma (total and free) cortisol concentrations (33, 82). Also in human patients with POMC deficiency, a were constantly high and ACTH levels constantly low.

When cortisol availability is insufficient, this has immediate potentially lethal consequences, as demonstrated by the phenotype of patients with Addison's crisis and Addison's disease (155). Also in adrenalectomized mice, it was shown that mortality strongly increased when sepsis was induced by bacterial endotoxin administration (16).

ACTH is responsible for both the short- and long-term regulation of cortisol synthesis from the adrenal gland. Insufficient cortisol availability during critical illness on the membrane of the adrenocortical cells, it activates protein kinase A (PKA) (Fig. 4) (142). PKA activates cholesterol esterase through phosphorylation of droplets (intracellular vesicles) into the cytoplasm of these adrenocortical cell (156). Furthermore, ACTH rapidly increases the expression of the steroidogenic acute regulatory protein (STAR), which is responsible for the transport of cholesterol from the cytoplasm to the inner membrane of the mitochondria where steroidogenesis takes place (142). STAR is indispensable for cortisol production (93). Next, cholesterol is converted into different steroid hormones by their respective catalyzing enzymes, in which the first step of the synthesis of cortisol is the hydroxylation of 11-deoxycortisol by 11 $\beta$ -hydroxylase (150). This process, which in total takes only a few minutes, does not depend on new mRNA synthesis but on the activation of several proteins, primarily caused by phosphorylation through PKA (150). The long-term impact of sustained ACTH activity on its receptor involves increased transcription of genes important for cholesterol uptake, cholesterol synthesis, and steroidogenesis as such enhancing the synthetic capacity of the cells (91, 92, 95, 142, 150). In addition, increased availability of ACTH affects adrenal gland structure and growth, by first inducing hypertrophy and hyperplasia later on, and by increasing blood flow to the adrenal cells could be caused by an increase of the expression

However, the causal relationship between sustained ACTH deprivation, critical illness-associated acquired loss of adrenal function and low plasma cortisol still has to be established. Insufficient cortisol availability during critical illness could also be the result of failure at any level of the HPA axis, from low CBG or altered CBG binding capacity with a loss of the circulating cortisol reservoir, to an inadequate cortisol production (102). Pro-inflammatory cytokines, such as TNF- and IL-1, may reduce cortisol synthesis during sepsis by inhibiting stimulatory actions of ACTH (146). Furthermore, ischemia or hemorrhage within the adrenal cortex during severe stress or sepsis can cause changes that impair cortisol production. Also, a decreased blood supply to the pituitary can evoke ischemia, followed by accumulation of nitric oxide and impaired ACTH secretion (30). Decreased cortisol production during acute illness may theoretically also be due to substrate deficiency, since HDL cholesterol has been shown to be substantially reduced during sepsis (164). Other possible causes of ICU acquired adrenal failure, are the administration of drugs that interfere with steroidogenesis, such as etomidate and the antifungal agent ketoconazole, and chronic exogenous corticosteroid therapy (35).



**Figure 4** Adrenocorticotropic hormone (ACTH) binds to its receptor, the melanocortin 2 receptor (MC2R), on the membrane of the adrenocortical cells, which increases cyclic AMP (cAMP) and stimulates protein kinase A (PKA). PKA causes the release of cholesterol from the lipid droplets into the cytoplasm and de novo production from acetyl coenzyme A (acetyl CoA). ACTH increases the expression of the steroidogenic acute regulatory protein (STAR) to transport cholesterol from the cytoplasm to the inner membrane of the mitochondria where steroidogenesis takes place. Cholesterol is converted into different steroid hormones. The long-term impact of ACTH involves increased transcription of genes important for cholesterol uptake [scavenger-receptor class B, member 1 (SCARB1), LDL receptor (LDLR)] and cholesterol synthesis [3-hydroxy-3-methylglutaryl-CoA reductase (HMGCR)], and for steroidogenesis (STAR and CYP11A1). ACTH has a direct stimulatory effect on the expression of its own receptor (MC2R). Blue lines represent ACTH effects. (Figure was reproduced from Boonen (17), with permission from *The Lancet Diabetes & Endocrinology*.)

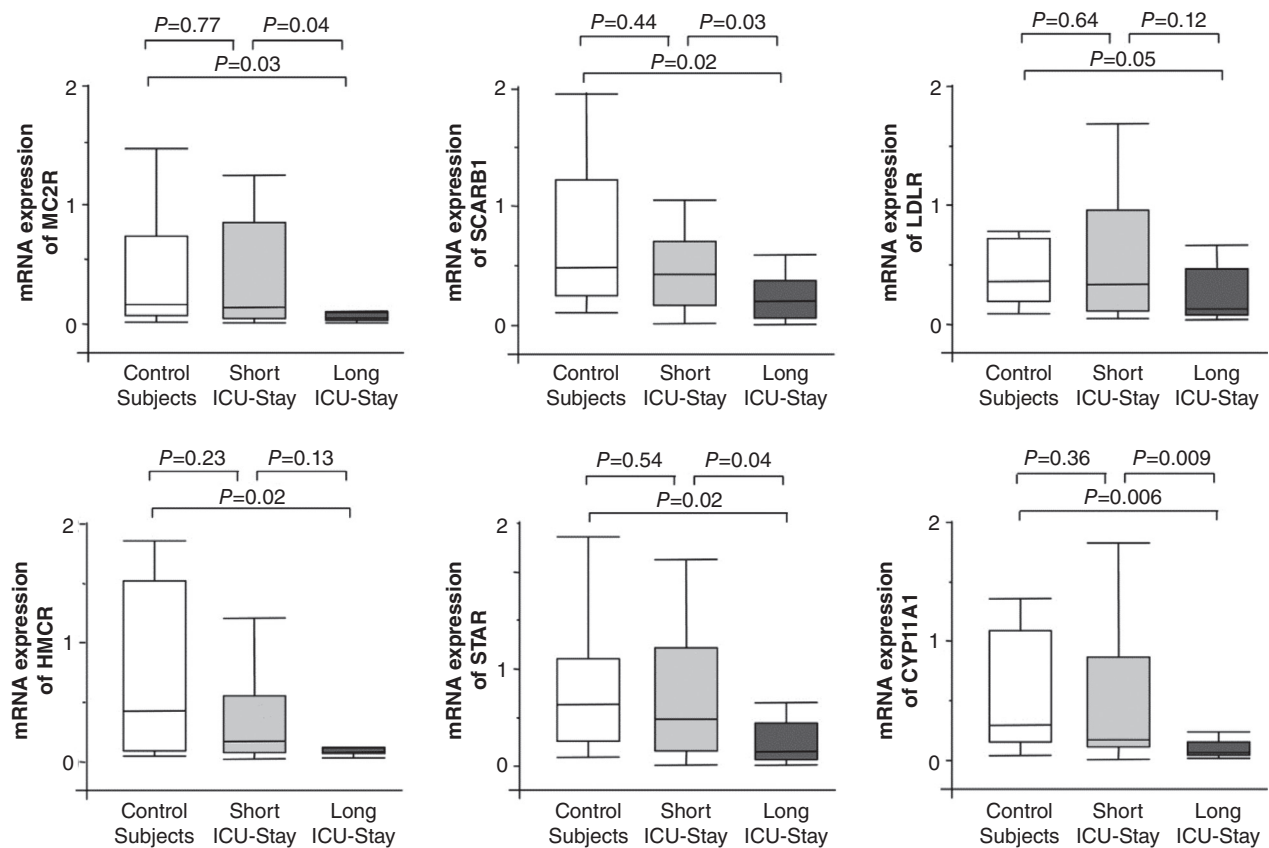
of GR $\beta$ , and/or by downregulation of GR $\alpha$ , which could be mediated by micro RNA124 (75, 90). Furthermore, reduced translocation to the nucleus or the presence of less functional GR polymorphisms may also play a role. Rodents with a general dimerization-deficient GR are indeed highly susceptible to adverse outcome when sepsis is induced (85). However, it remains unclear whether the changes documented in peripheral blood cells are adaptive, to safeguard the function of immune cells, or, instead, maladaptive and a sign of generalized insufficient GR signaling.

In the past, experts have suggested the presence of a phenomenon that comprises a “relative exhaustion” or “insufficiently activated” adrenal cortex, insufficient to cope with the level of stress of septic shock in particular (138). In patients suffering from such presumed “relative” adrenal failure, plasma (free) cortisol concentrations are still much higher than normal, but it is assumed that this is not enough to cope

with the level of stress and inflammation, and therefore to negatively affect outcome (9). However, the term “relative” adrenal insufficiency is currently quite controversial and many experts now challenge its existence (97).

### Diagnosis of adrenal failure during critical illness

Diagnosis of adrenal failure is complex and prevalence among ICU patients varies widely from 0% to 77% depending on the definition and criteria used (103). Diagnosis of adrenal failure starts with a clinical suspicion such as hypotension that is resistant to vasopressors, unexplained coma, hyponatremia and hyperkalemia. Outside the ICU, this clinical suspicion can be confirmed by the presence of a low total morning plasma cortisol (<3  $\mu\text{g}/\text{dL}$  or <80  $\text{nmol}/\text{L}$ ), although this can be highly variable (45, 65). Therefore, plasma cortisol concentrations <18  $\mu\text{g}/\text{dL}$  or <500  $\text{nmol}/\text{L}$  upon stimulation with



**Figure 5** mRNA expression of ACTH-regulated proteins in adrenal glands, harvested from individuals dying suddenly out of hospital (control subjects), from patients dying after short critical illness and from patients after prolonged critical illness. The mRNA data are expressed, normalized to RNA18S as a fold difference from the mean of the controls. Boxes represent medians and interquartile ranges and whiskers represent firstquartile-1.5\*IQR and thirdquartile+1.5\*IQR. (Figure was reproduced from Boonen (18), with permission from *The Journal of Clinical Endocrinology and Metabolism*.)

250 µg of synthetic ACTH(1-24) (short ACTH stimulation test, Synacthen), are more indicative of adrenal failure (125).

Using the same cut-off levels for cortisol as used in healthy individuals has potential pitfalls, since basal cortisol levels are much higher in ICU patients. Furthermore, low CBG and albumin levels might further increase free cortisol levels, which makes total cortisol levels less relevant for this diagnostic question (66). Indeed, a study in 66 critically ill patients reported that hypoproteinemia results in low total plasma cortisol levels with a low total cortisol response to Synacthen, indicating adrenal insufficiency, while free plasma cortisol levels were consistently elevated and several times higher than in healthy volunteers (67). Therefore, the free cortisol response to Synacthen in critically ill patients might be a more valuable clinical determination than the total cortisol response, to avoid treatment of patients with a normal adrenal function (98). Some authors even doubt that an increase in total cortisol would be essential to survive acute stress, given that the free fraction is so much higher (97). Studies of critically ill patients, investigating the association between plasma cortisol and mortality, failed to show a minimum level of plasma cortisol concentration below which mortality clearly increased (78, 79, 147). Hence, there is currently no consensus

on a cut-off for plasma cortisol to diagnose adrenal failure in ICU patients and even less to indicate the need for treatment with hydrocortisone.

Salivary cortisol levels might be a surrogate for free plasma cortisol in the diagnosis of adrenal failure in critically ill patients, but has not been validated extensively. Salivary cortisol is in close equilibrium with free cortisol and might offer an accurate measure of the biologically active cortisol availability (10, 171). However, local conversion to cortisone through 11β-HSD2 presence in the salivary gland, and reduced salivary flow due to stress, hypovolemia, and opioids effects might limit the use of this technique (13, 42, 118). Also potential blood contamination, by presence of mucositis and/or pathogenic microorganisms, constitutes a major challenge during sampling of pure saliva in critically ill patients (13, 42). Nevertheless, several studies in both adult and pediatric critically ill patients found excellent correlations of salivary and free plasma cortisol, strengthening its potential clinical use (10, 52, 64, 121). In the diagnosis of adrenal failure, in accordance with morning plasma cortisol, morning salivary cortisol levels vary widely and are not advised to be used (101). But ACTH-stimulated free plasma and salivary cortisol concentrations increased in parallel in both adult and

pediatric critically ill patients (10, 12). However, also opposing results between free and salivary cortisol were measured in patients with severe sepsis (12, 51). Interestingly, in a study of 28 acutely ill patients with a clinical suspicion of adrenal insufficiency, 13 patients had a similar response to ACTH in peak serum total and salivary cortisol, whereas 15 patients displayed a subnormal serum total cortisol response, but a normal salivary cortisol response (122). Salivary cortisol measurements can thus potentially identify patients with a normal adrenal function but an abnormal total cortisol response. As such, salivary cortisol might be a clinically useful and easily obtainable parameter to exclude adrenal failure in ICU patients and thus avoid unnecessary treatment. This possibility should be further investigated.

Experts have advised to diagnose “relative” adrenal failure in ICU patients by the incremental cortisol response to an ACTH stimulation test, irrespective of the baseline plasma cortisol. A cortisol increase of less than 9 µg/dL or 240 nmol/L after stimulation with 250 µg of synthetic ACTH(1-24), irrespective of the baseline plasma cortisol, or a high baseline plasma cortisol levels >34 µg/dL or 907 nmol/L, have been proposed, as these were most discriminative for increased risk of death (9). A low cortisol response to exogenous ACTH was also associated with a higher baseline cortisol and ACTH and with more severe disease and presence of sepsis and septic shock (39, 54). However, in critically ill patients, a low rise in plasma cortisol in response to an ACTH stimulation test was associated both with a low cortisol production rate (but still equal to healthy individuals), and, more importantly, with low clearance of plasma cortisol (20). This suggested that a suppression of cortisol breakdown may explain a reduced cortisol response to ACTH, and actually may reflect the degree of negative feedback inhibition exerted by supranormal cortisol availability. This mechanism is also observed in patients treated with exogenous glucocorticoids, with a lower response to an ACTH stimulation test (134). Therefore, in the presence of increased plasma cortisol and suppressed cortisol metabolism, a reduced cortisol response to ACTH may not necessarily point to an insufficient cortisol availability. Most recent practice guidelines therefore do not advise to use the ACTH stimulation test to guide treatment, in line with the lack of consensus on how to diagnose “relative” adrenal failure in the ICU (128). Clearly, more research on this topic is needed.

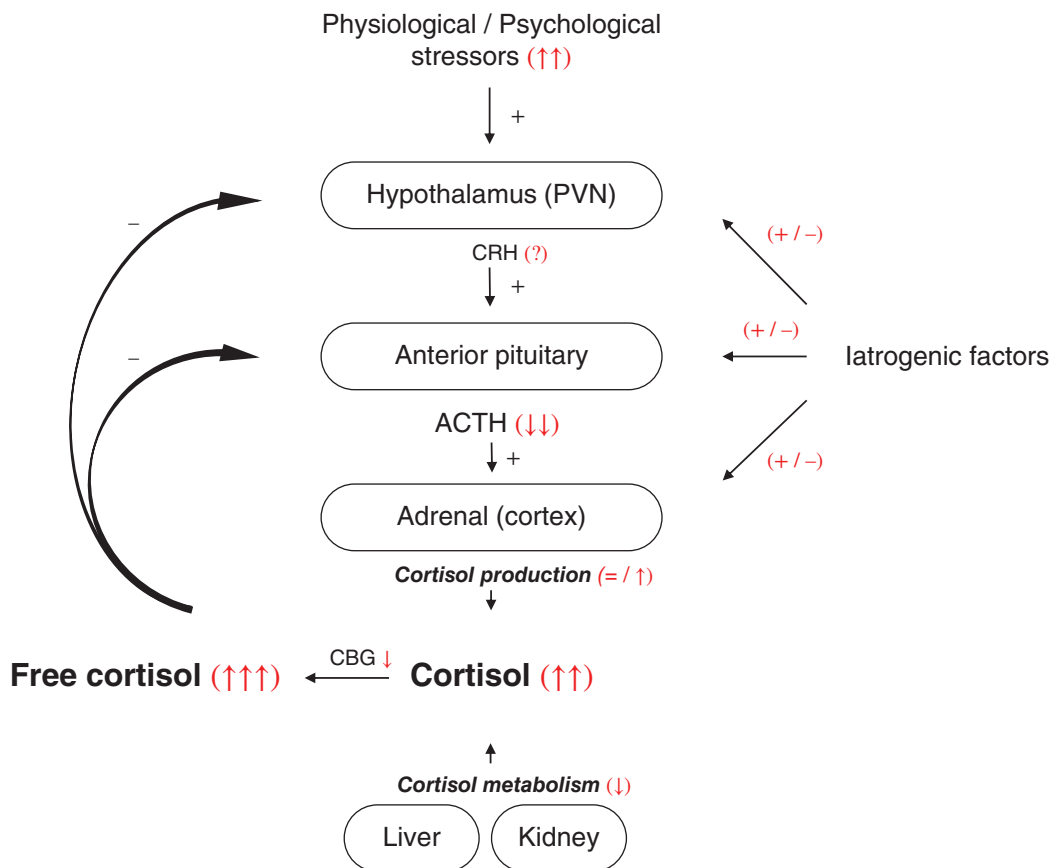
## Treatment

Evidently, ICU patients suffering from adrenal failure should receive coverage to cope with the stress (41). Currently, it is recommended to treat adrenal failure during critical illness with a bolus of 100 mg hydrocortisone, followed by 50 mg every 6 hours on the first and second day, followed by 25 mg every 6 h on day 3, tapering to a maintenance dose by days 4 to 5. As such, this dose is the equivalent of a several-fold increased daily cortisol production, which is normally around 25 to 30 mg per day.

Patients with presumed “relative” adrenal failure during critical illness with signs of shock should not be treated with hydrocortisone, if adequate fluid resuscitation and vasopressor therapy are able to restore hemodynamic stability during shock (128). However, there is a weak recommendation, with low quality of evidence, that if this is not achievable, this condition can be treated with hydrocortisone at a dose of 200 mg per day. Indeed, a rise in blood pressure following treatment with hydrocortisone has been used as a proof for underlying adrenal failure (66). However, this might be related to a pharmacological effect of these doses of hydrocortisone on the vasculature rather than indicating a successful treatment of any form of suspected adrenal failure (97, 148). Empiric treatment with hydrocortisone, which results in hemodynamic improvement in some patients, does not assume a previous diagnosis of “relative” adrenal failure. A large French randomized controlled trial (RCT) investigated the impact of 200 mg hydrocortisone (in combination with 50 µg fludrocortisone) in patients with septic shock (8). This pioneer study showed reduction in mortality with this treatment only in patients who did not have an incremental cortisol response to synacthen above 9 µg/dl (8). In contrast, a subsequent large European RCT failed to show any mortality benefit with use of hydrocortisone (148). Other smaller RCTs and systematic reviews also generated conflicting results (6, 76, 83, 172). Subgroups of ICU patients, more specifically patients with acute respiratory distress syndrome (ARDS) and patients with severe community acquired pneumonia appear to benefit from treatment with corticosteroids (107, 152). A speculative explanation for the conflicting results of these studies might be that, given the reduced breakdown and prolonged cortisol half-life during critical illness (20), doses of 200 mg hydrocortisone could be too high, and induce side effects such as myopathy, muscle wasting, whereby extending the intensive care dependency (70, 71). It is reasonable to assume that lower doses of hydrocortisone, for any indication during critical illness, might actually be sufficient (114). It was demonstrated that cortisol production during critical illness is only moderately increased, more or less doubled but only in patients with excessive inflammation, whereas in other critically ill patients, cortisol production is not different from that in healthy subjects (20). Hence, during critical illness, cortisol production rates range from about 30 to 60 mg per day. It is thus possible that 60 mg hydrocortisone per day could suffice as a substitution dose during critical illness, but further research is needed to determine the optimal therapeutic dose and potential benefits.

## Conclusion

During critical illness, normal to slightly increased cortisol production and a substantially reduced cortisol breakdown appear to be the main drivers of hypercortisolemia during critical illness (Fig. 6). Besides total plasma cortisol levels, the dynamics of increased biologically active free plasma



**Figure 6** Overview of the regulation of hypercortisolism during critical illness. ↑, elevated plasma concentrations; ↓, decreased plasma concentrations; ?, no univocal data available; +, stimulates; -, inhibits; PVN, paraventricular nucleus; ACTH, adrenocorticotropic hormone; CBG, corticosteroid-binding globulin.

cortisol, and tissue-specific alterations of glucocorticoid signaling, further characterize these changes. Clinicians should be aware that several drugs that are often administered to ICU patients, such as opioids, etomidate, and propofol, suppress cortisol in a dose-dependent manner. While plasma cortisol levels are increased, plasma ACTH levels, however, are decreased, which implies that critical illness is not hallmarked by a full central activation of the HPA-axis, but by an “ACTH cortisol dissociation,” with loss of the diurnal rhythm of ACTH and cortisol. These findings have revived the ongoing debate about which level of cortisol availability is sufficient in the struggle for survival of the critically ill, about the concept of “relative” adrenal failure, and about how to correctly interpret diagnostic laboratory tests. The ongoing controversy clearly indicates the need for further research on this important clinical problem.

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