Russell, G., & Lightman, S. (2019). The human stress response. *Nature Reviews Endocrinology*, *15*(9), 525-534. https://doi.org/10.1038/s41574-019-0228-0

The human stress response

Georgina Russell[1*] and Stafford Lightman[1*]

[1]Translational Health Sciences, Dorothy Hodgkin Building, Bristol Medical School, University of Bristol, Bristol, UK.

*email: Georgina.russell@bristol.ac.uk; Stafford.Lightman@bristol.ac.uk

Abstract

The human stress response has evolved to maintain homeostasis under conditions of real or perceived stress. This objective is achieved through autoregulatory neural and hormonal systems in close association with central and peripheral clocks. The hypothalamic–pituitary–adrenal (HPA) axis is a key regulatory pathway in the maintenance of these homeostatic processes. The end product of this pathway — cortisol — is secreted in a pulsatile pattern, with changes in pulse amplitude creating a circadian pattern. During acute stress, cortisol levels rise and pulsatility is maintained. Although the initial rise in cortisol follows a large surge of adrenocorticotropic hormone (ACTH), if long-term inflammatory stress occurs, ACTH returns to near basal levels while cortisol remains raised as a result of increased adrenal sensitivity. In chronic stress, hypothalamic activation of the pituitary changes from corticotropin-releasing hormone dominant to arginine vasopressin dominant, and cortisol remains raised due at least in part to decreased cortisol metabolism. Acute elevations in cortisol are beneficial to promote survival of the fittest as part of the fight or flight response . However, chronic exposure to stress results in reversal of the beneficial effects with long-term cortisol exposure becoming maladaptive, which can lead to a broad range of problems, including the metabolic syndrome, obesity, cancer, mental health disorders, cardiovascular disease and increased susceptibility to infections. Neuroimmunoendocrine modulation in disease states and glucocorticoid-based therapeutics are also discussed.

Key points

The hypothalamic–pituitary–adrenal (HPA) axis is a key system that synchronises the stress response with circadian regulatory processes.

Regulation of the HPA axis is very dynamic with both ultradian and circadian oscillations.

Short term and longer-term stress result in different regulatory mechanisms involving hypothalamic, pituitary and adrenal activity, as well as cortisol metabolism.

Chronic elevation and non-physiological patterns of cortisol result in poor cognitive, metabolic and immune function.

[H1] Introduction

In response to a stressor the body activates multiple co-ordinated and dynamic processes to restore homeostasis, preserve life and ultimately achieve evolutionary success for the species. The importance of endocrine systems in this homeostatic regulation has been known since the early studies of Hans Selye[1] in the 1930s, when activation of the sympatho–adrenodedullary (SAM) and hypothalamic–pituitary–adrenal (HPA) axes was described in response to physical injury and exertion as well as perceived psychological threats. Interestingly, anticipation of these threats is itself a very potent activator of these systems[2]. Homeostatic processes also interact with internal and external Zeitgebers such as the light–dark cycle and internal body clocks[3,4]. These internal clocks enable the body to anticipate regular changes in the environment to ensure optimal fitness across the 24 hours and thus the best chance for survival[5]. The human stress response is an additional homeostatic mechanism that provides a better chance of survival when the body is under threat and mobilises neural and hormonal networks to optimise cognitive, cardiovascular, immunological and metabolic function (figure 1). In this Review we discuss how the HPA axis helps achieve and maintain homeostasis. We will show how it utilises rhythmic 24-hour patterns of secretion to achieve

appropriate tissue activity at different times of the day, and faster ultradian rhythms to optimise tissue specific glucocorticoid signalling and maintain the rapid reactivity necessary for a stress response system.

[H1] Circadian clocks

In the absence of internal or external stressors, the integrity of physiological systems is maintained in a dynamic fashion over 24 h by an internal circadian clock that anticipates the changes occurring over the 24-h day. In the past, the neurocentric hierarchical view was that body rhythms were controlled from a master clock in the hypothalamic suprachiasmatic nucleus (SCN)[6]. Now, it is very clear that peripheral clocks also exist in most, if not all, tissues of the body, which have their own autonomous transcriptional autoregulatory feedback loops[7]. The core clock genes, both in the SCN and peripheral clocks, are *CLOCK* and *BMAL1*. Activation of these genes generates a heterodimer of *BMAL1* with either *CLOCK* (or NPAS2) which bind at promotor elements called E-boxes to drive genes encoding period 1-3, cryptochrome 1-2 and nuclear receptor subfamily 1-2. The resultant proteins then feedback to repress *BMAL1* by a series of feedback loops generating a 24-hour rhythm[8,9] which allows time of day dependent regulation of downstream genomic pathways. This rhythmicity is crucial for homeostasis. The body can only behave optimally when all biological rhythms are in synchrony. However, with our increasingly chaotic lifestyles this orderly physiological regulation is steadily being disrupted, which can result in chronodisruption[4,10]. This desynchronization between cellular oscillators in the SCN and peripheral tissues can manifest as negative health outcomes in the form of cardiovascular, metabolic, cognitive and immune dysfunction[4,10-14].

[H1] HPA axis and circadian rhythmicity

The HPA is critical for life and is a major part of our homeostatic regulatory system[15]. The output of this system is the endogenous glucocorticoid corticosterone (in rodents) or cortisol (in humans), which are collectively referred to as CORT. Glucocorticoids have diverse and far reaching effects, which is why they are such successful therapeutic agents; however, this diversity is a double-edged sword and excess levels of glucocorticoids result in a myriad of unwanted adverse effects including diabetes, hypertension, immune dysregulation and osteoporosis[16]. Glucocorticoids exhibit powerful anti-inflammatory functions both at a whole cell and at transcriptional level. They can induce apoptosis of T lymphocytes, neurotrophils, basophils and eosinophils[17]. They also regulate multiple proinflammatory genes encoding cytokines, chemokines and inflammatory enzymes associated with repression of AP1 and NF-Kappa B transcription[18]. Glucocorticoids also inhibit antigen presentation[19,20] and major histocompatibility complex class II expression[21] and antibodies[22], and favour T helper (T_H) 1 versus T_H2 responses[20]. They influence cytotoxic effects via cell death and oxidative stress[23], have a role in metabolic regulation through glucose utilisation and ATP production[24] and interact with the major neurotransmitters and many secondary neuropeptidergic systems. As such, glucocorticoids modulate emotion and cognition, with key examples being learning ability, performance, emotional perception and mood[25,26]. These interactions also exemplify how glucocorticoid therapy can result in multiple effects including unwanted side effects such as depression[27-30].

CORT is a homeostatic anticipatory hormone that is secreted by the adrenal glands. Consequently, under basal conditions it is released with a characteristic circadian pattern of secretion with high levels just before waking (start of active cycle), followed by a steady decline down to trough (or nadir) levels during the sleeping or inactive phase; hence anticipating the needs of the body (figure 2A). CORT's daily rhythm is regulated through indirect projections from the SCN to the paraventricular nucleus (PVN) of the hypothalamus, which inhibit corticotropin-releasing hormone (CRH) and arginine vasopressin (AVP) release during the inactive phase of the cycle[31,32]. Furthermore, to produce appropriate reactivity to physiological, cognitive and emotional stressors, the brain stem and limbic system also modulate HP activity via projections to the PVN[33,34].

Axonal terminals in the median eminence release CRH and AVP into the hypophyseal portal system where they are transported to the pituitary and stimulate pituitary corticotrophs to release adrenocorticotropic hormone (ACTH). ACTH is released into the systemic circulation and once at the adrenal cortex stimulates the production of CORT. CORT undergoes *de novo* synthesis and release back into the systemic circulation, enabling it to travel to its target tissues and produce its characteristic metabolic, cardiovascular, immunological and cognitive effects[33]. CORT also acts via an autoregulatory negative feedback loop and inhibits HPA activity via effects at the level of the pituitary, hypothalamic PVN and hippocampus[35].

Further levels of circadian control exist, including splanchnic nerve innervation of the adrenal glands[36]. The adrenal glands receive autonomic (sympathetic) innervation via neuronal projections of the autonomic portion of the PVN[36,37], which alters adrenal cortical sensitivity to ACTH with a reduction of responsiveness during the circadian nadir. CORT synthesis and adrenal clock gene functioning is also influenced by a light-sensitive mechanism that occurs across the 24 h period, with a shift in irradiance threshold according to the time of day. Thus, in animals housed in complete darkness, although high intensity light activated corticosterone at all times of day, lower intensity light had no effect during the subjective day. The adrenal response therefore is dependent on both irradiance and circadian phase[38,39]. The adrenal gland itself also has an autonomous clock that regulates ACTH sensitivity and steroidogenesis, allowing it to fine tune its own homeostatic control[40,41]. Finally, peripheral CLOCK-mediated acetylation of the glucocorticoid receptor can decrease tissue sensitivity to glucocorticoids in a circadian manner[132].

These circadian fluctuations in activation of glucocorticoid receptors also have important interactions with multiple other crucial homeostatic processes including the transcriptional activity of other genes that respond to glucocorticoids and their corresponding physiological outputs, such as physical activity and body temperature[5,42]. For example, in the rat, glucocorticoid responsive tryptophan hydroxylase

2, a gene implicated in physical activity, temperature and emotional response has a circadian rhythmicity which is abolished by exogenous steroids[43].

[H1] Stress response

[H2] Acute stress.

The acute response to stress is a dynamic process that changes over time, starting with stereotypic behaviours and then changing to goal directed behaviours specific to the stressor, followed by activation of the SAM within seconds and finally recruitment of the HPA axis, with peak levels of cortisol occurring between 15 and 20 minutes after stress onset[44]. These early responses provide increased energy resources and initiate longer term and slower genomic effects that restrain inflammatory and other potentially dangerous responses[45]. The response to the acute stress of cardiac surgery can be seen in Figure 2B[46]. This response is very interesting for several reasons. First, despite the greatly increased levels of cortisol, the pattern of cortisol secretion remains pulsatile. Second, despite initial high levels of ACTH, these rapidly fall to basal levels while the cortisol remains raised. Despite this fall in ACTH levels, small changes in these basal levels of ACTH initiate large pulses of cortisol, indicating a rapidly induced increased sensitivity of the adrenal cortex to ACTH. This effect has now been investigated in reverse translation studies in rats and complemented with mathematical modelling, which enabled the importance of the dynamic adrenal steroidogenic regulatory network to be characterised[47].

[H2] Chronic stress

In response to chronic stress, a dynamic change of the ratio of AVP to CRH in the hypothalamic PVN occurs[48] as well as an associated decreased sensitivity to glucocorticoid feedback[49]. Obstructive sleep apnoea is an excellent model of chronic stress (figure 2C). In sleep apnoea, there is a marked increase in the amount of cortisol released during each secretory pulse – which normalises after continuous positive airways pressure (CPAP) treatment[50]. In critical illness, the situation is somewhat different

with the increased levels of cortisol produced by long-term stress being present for the first few days secondary to increased adrenal sensitivity to ACTH and increased cortisol synthesis[46]. During prolonged critical illness, there is a further change in HPA regulation with reduced cortisol metabolism becoming an increasingly important factor in maintaining raised levels of plasma cortisol[51,52].

[H2] Glucocorticoid signalling

Glucocorticoid receptors (GR) and mineralocorticoid receptors (MR) are the cognate intracellular nuclear receptors for CORTs[53]. The affinity of CORT for MR is approximately five-fold to ten-fold higher than that for GR[54]. Binding of CORT to its receptors leads to either transactivation or repression of genomic transcription as well as more rapid nongenomic effects[55,56]. Non genomic signalling is mediated via classic[57] or membrane bound variants of the receptors[58,59]. The membrane bound variants have lower glucocorticoid affinity than their classic nuclear counterparts[60]. Nuclear MRs are generally constantly occupied during the day and only become unoccupied at the very low levels of CORT found at night in humans or during the day in rodents. As CORT levels rise to a critical threshold (as seen during the circadian peak or following an acute or chronic stress, nuclear GR and membrane associated MR and GR occupation occurs[53]. It is important to note that the one exception to this effect is the hypothalamic SCN, which does not appear to be regulated by circulating CORT[61]. As such, the SCN is the one place where endogenous CORT cannot shift clock function. Whether this feature is due to a lack of corticosteroid receptors or altered chromatin structure is unclear.

A further level of regulatory control is the tissue specificity of GR and MR distribution. GRs are present throughout the brain and peripheral tissues whilst MRs have more limited localisation, predominately being found in cardiovascular tissue, liver and kidneys, as well as corticolimbic regions of the brain[53]. Although GR is present throughout the brain, only the hippocampus, basal ganglia, lateral septum and medial amygdala neurones present a high MR:GR ratio[62]. Since these areas do not express 11 β-hydroxysteroid dehydrogenase (11 β-HSD) type 2 (discussed in detail in a subsequent section), MR is persistently occupied even during the circadian nadir and it is GR and the fast acting non-genomic

response of the lower affinity membrane-bound MR that responds when CORT levels rise in response to a stressor[28]. This response helps to prepare an individual to respond to a stressor through enhancing synaptic plasticity at a cellular level, which leads to a behavioural change in the form of altered decision making, attentional bias and risk assessment[63]. Although other brain areas can still respond to changes in CORT this will only be through their genomic and nongenomic GR[64]. Consequently, this region specificity in receptor distribution will modify corticosteroid signalling including the activation and termination of stress responses, and are likely to be important for promoting long-lasting adaptive protective mechanisms such as strategic planning, memory storage and consolidation[65].

Other layers of regulatory control arise through availability of CORT. Firstly, CORT is highly lipophilic, which means that under basal conditions 90% of CORT is tightly bound to the high-affinity, limited capacity protein carrier corticosteroid-binding globulin (CBG)[66,67]. Around 4–5% is loosely bound to albumin and the remaining 5% of CORT is 'free' and biologically active[68]. CBG levels are controlled by a variety of different factors such as genetic variability, liver and thyroid function, nutritional status, inflammation and stress. Additionally, CBG is saturated at fairly low CORT concentrations, including under basal (non-stressed) levels found at the circadian peak. Similarly to CORT, CBG exhibits homeostatic diurnal variation[69], the resultant effect being a higher proportion of free (and therefore biologically active) CORT during the circadian peak, which accentuates the diurnal profile of free CORT and the response to stress. CBG also acts as a carrier to specific target sites (such as sites of inflammation) and even acts as a protein thermocouple releasing CORT at sites of increased temperature[70]. CBG concentrations are therefore of great importance for the bioavailabity of CORT and its ability to act at a tissue level[71].

The metabolic clearance of CORT occurs through two mechanisms, enzymic degradation in the liver[71,72] and intracellular metabolism by the two iso-enzymes of 11β-HSD, which catalyse the interconversion of active CORT and the inert forms, cortisone (human) and 11-dehydrocorticosterone (rodents). 11β-HSD type 1 is predominately a reductase that catalyses the regeneration of active

glucocorticoids, which amplifies the cellular actions of CORT[72]. It is widely expressed in liver, adipose tissue, muscle, pancreatic islets, adult brain, inflammatory cells and gonads[73]. 11β-HSD type 1 therefore increases tissue levels of CORT and is thought to be a major factor in regulating tissue sensitivity to CORT, especially under conditions that can increase 11β-HSD type 1 activity such as visceral obesity[71]. In addition, 11β-HSD type 1 is thought to mediate many cognitive and metabolic effects of CORT excess. For instance, 11β-HSD type 1 deficiency in rodents protects against age and stress related cognitive decline whilst transgenic over-expression exacerbates cognitive impairment[74]. The other iso-form, 11β-HSD type 2, is a high-affinity dehydrogenase that inactivates glucocorticoids and is particularly important in the kidney, where it protects mineralocorticoid receptors from activation by the much higher concentration of cortisol[72]. More studies are needed to define how tissue ratios of cortisol and cortisone may change during stress and whether this might provide yet another mechanism to guard our homeostatic state.

[H2] Ultradian rhythm

Underlying the circadian rhythm there is a complex and dynamic ultradian rhythm comprised of discrete pulses of secretion of both ACTH and cortisol (figure 2A). This pulsatile pattern is produced at an approximate frequency of 60–90 min with increased pulse amplitude and frequency at the circadian peak[75]. Patterns of secretion are influenced by a multitude of factors including genetics, sex hormones, epigenetic influences, environmental stressors and age, which leads to individual diversity[76,77]. Unlike circadian rhythms, the origin of the ultradian rhythm is not from central SCN regulation; rodents with SCN lesions show an isolated loss of circadian rhythmicity, but the underlying pulsatility of CORT remains[78]. Nor does the ultradian rhythm arise as one would expect from a central hypothalamic 'pulse generator', as pulses of CRH do not induce downstream pulses of ACTH and CORT[79]. Mathematical modelling techniques were consequently utilised, which predicted that the ultradian rhythm has a sub-hypothalamic origin[61]. Here, a dynamic feedforward–feedback communication exists between ACTH on the adrenal cortex and endogenous glucocorticoids on the

pituitary corticotrophs, together driving ultradian rhythmicity with a natural delay between ACTH activating adrenal MC2 receptors, glucocorticoid biosynthesis and cortisol release (figure 3). This prediction was subsequently confirmed in our rat model[61].

The adrenal gland itself preferentially responds to oscillatory ACTH signals; in rodents whose endogenous ACTH is suppressed with exogenous glucocorticoids, pulsatile ACTH infusions result in pulsatile corticosterone secretion whereas constant ACTH infusions of the same total dose result in no response[80]. Additionally, the adrenal gland has its own internal feedforward–feedback system that amplifies the response to rapid changes of ACTH, sensitising the adrenal glands to oscillating levels of ACTH and hence alterations in CORT[81].

[H2] Impact of hormone oscillation

Both circadian and ultradian oscillations in hormone levels matter. Between 5% and 20% of cells show clear circadian oscillations in mRNA levels[12] and circadian fluctuations are found at almost every stage of gene expression[82] and have been associated with important biological effects. In transgenic mice, if circadian peak levels of CORT are administered during a motor learning task, these mice show increased memory of motor skills[83]. This effect arises secondary to a rapid GR-mediated non-genomic mechanism via LIM kinase signalling that increases numbers of new neuronal spines. Low trough levels of CORT are then needed to retain and stabilise the memory. This retention and stabilisation involves 'pruning' of old spines via a MR-mediated pathway that alters gene expression, which implies that circadian fluctuations are required for plasticity in learning and memory[83]. Indeed, clinically this phenomenon is nothing new as in patients taking glucocorticoid-based therapeutics and in Cushing syndrome, both situations with chronically elevated glucocorticoid levels and/or no oscillations, there are marked impairments in learning and memory[25].

What we are beginning to understand is that circadian rhythmicity is only the tip of the iceberg and functionally important dynamic changes are seen on a shorter time frame than simply circadian alterations. In the HPA axis, ultradian hormone presentation is essential for physiological gene

functioning[84]. Both *in vitro* and *in vivo* very different patterns of gene activation are seen depending on the pattern of hormone presentation[85]. Ultradian oscillations in glucocorticoid levels result in a phenomenon known as gene pulsing (figure 4A). This phenomenon is specific for the endogenous glucocorticoid hormones as synthetic glucocorticoids have long binding characteristics at the GR and cannot cycle on and off[85]. As CORT levels rise, as is seen in endogenous glucocorticoid pulsatility, GR binding and activation occurs followed by nuclear translocation, dimerization and interaction with deoxyribonucleic acid (DNA) glucocorticoid response elements (GRE), which initiates transcription. Next, rapid cycling of GR and transcription factors on and off chromatin occurs. Once CORT levels decline, as seen in pulse termination, GR dissociates from DNA and is released back into the nucleoplasm ready to rapidly respond to subsequent CORT pulses. If the same dose of CORT is used but presented as a constant as opposed to pulsatile pattern of presentation, genomic reactivity changes lead to altered transcriptional activity[85,86] as well as differential gene responses (figure 4B)[85]. In addition to the classic GRE-dependent interactions, GR can also interact with a variety of other transcription factors and influence transcription via protein–protein interactions[87,88] and chromatin remodelling[87,89]. These pattern dependent interactions have serious implications for therapeutics based on continuous exposure to glucocorticoids, and is likely to contribute to the adverse effects of these drugs[16].

Downstream, constant and pulsatile patterns of CORT result in differential neuronal and behavioural responses. For instance, noise stress induces neuronal activation of c-fos in the amygdala, hippocampus and hypothalamic PVN. Interestingly, pulse phase is crucial for this response. If the noise is administered during the rising phase, the c-fos response in the amygdala is increased (Figure 4C). This effect was paralleled by differential behavioural responses to the stressor. Importantly, the effect is reproducible with the pulse phase dependency pattern being recorded in aggressive and novelty behaviour responses to glucocorticoids, as well as in the neuroendocrine mechanism of rapid HPA negative-feedback[90,91]. Translational human studies have found similar results (figure 4D). Healthy male volunteers on a block and replace regimen of endogenous glucocorticoid blockade with

metyrapone and replacement therapy with either circadian or ultradian subcutaneous hydrocortisone or standard oral hydrocortisone therapy (three times a day) at the same total daily dosages exhibit altered cognitive and behavioural effects that are dependent on the cortisol pattern. Specifically, a lack of physiological pulsatility was associated with poorer quality of sleep, poorer working memory performance and altered accuracy in recognition and attentional bias toward or away from emotional faces[92].

[H1] Stress and disease

Stress is any stimulus that disrupts or threatens homeostatic balance. The stress response is a mechanism that can restore homeostatic processes and promote self-preservation through a complex interaction between the HPA axis, central and peripheral autonomic nervous systems and immune systems[15]. The HPA axis response to stress by rapidly releasing CORT. As CORT is highly lipophilic it is not prestored in vesicles and is synthesised *de novo* in response to ACTH. During long-term stress a dissociation between ACTH and CORT secretion develops[93,94], predominantly via increased adrenal sensitivity to ACTH[47] or non-ACTH mediated mechanisms[94]. This effect is seen in patients undergoing cardiac surgery, who show a marked shift in adrenal responsiveness (figure 1)[46], reverse translation animal studies suggest this shift arises secondary to changes in the regulation of both stimulatory and inhibitory intra-adrenal signalling pathways[46]. In models of inflammatory bowel disease and viral challenges, IL-6 and ACTH independent immune–adrenal pathways have also been linked to this dissociation[95,96].

The rapid rise in CORT seen under conditions of stress increases availability of supplemental energy stores by promotion of gluconeogenesis and inhibition of insulin production combined with vasoconstriction, which aids delivery of blood to muscles and the brain[97]. Centrally, CORT promotes decision making and alertness and stimulates cognitive functioning. CORT also has a pro-inflammatory effect, resulting in an increased response to infection[98]. Chronic exposure to stress results in reversal of the beneficial effects with long-term cortisol exposure becoming maladaptive, which leads to a

broad range of symptoms and disease states, including the metabolic syndrome, obesity, cancer, mental health disorders, cardiovascular disease and increased susceptibility to infections[10,11,53,99,100].

With regards to immune functioning, components of both the adaptive and innate immune system undergo circadian variations in levels and functionality to optimise response and recovery to a pathogen[101]. Autonomous circadian clocks are also found on different subsets of immune cells, including macrophages and lymphocytes[102,103]. CORT also has a multitude of effects on the immune system[20]. Chronic increases in CORT, either as a result of the effect of the steroid or the development of glucocorticoid resistance[100], increases susceptibility to infection[20]. Chronic stress can impair GR function as a result of long-term exposure to the resultant release of pro-inflammatory cytokines. Indeed, cytokines (along with their signalling pathways including but not limited to mitogen-activated protein (MAP) kinases, nuclear factor-kappaB (NF-κB) and cyclooxygenase) inhibit GR function[98]. Potential mechanisms underlying this inhibition include disruption of GR translocation, GR–DNA binding, GR inflammatory mediator interactions and alterations in GR phosphorylation status[104-106]. Glucocorticoid resistance in turn interferes with HPA downregulation of the very same pro-inflammatory cytokines and creates a vicious cycle. Thus, in the case of the common cold, an exaggerated response to the symptoms and signs of the upper respiratory tract illness is generated by the resultant pro-inflammatory response[107].

Long-term exposure to CORT also increases blood levels of glucose[108] and stimulates adipocyte precursor maturation, which promotes adiposity[109]. Adipose tissue is now seen as an active endocrine gland rather than a passive energy store as it produces a wide variety of hormones and regulatory factors[71]. The levels of these factors in turn reflect the metabolic status of the adipocytes and interact with homeostatic connections between adipose tissue and the brain as well as other organs, which together control energy intake and expenditure[71,110]. Obesity is also linked to chronic low-grade inflammation[107] via TNF and IL-6. TNF inhibits intracellular insulin signalling pathways and IL-6 promotes atherosclerosis and metabolism of glucose and lipids[110,111] all of which interact with the HPA

axis[20]. Studies of an increasingly common stress associated problem — sleep deprivation — confirm elevated nadir CORT levels, which then acts as another risk factor for insulin resistance[112]. Behaviourally, chronic sleep deprivation is associated with increased appetite and energy expenditure[113]. Chronic CORT exposure also stimulates mesolimbic reward pathways within the brain, leading to increased intake of palatable food such as sucrose solutions[110]. Additionally, increased feeding might be stimulated by leptin and ghrelin levels[114,115]. During chronic stress it is thought that these two hormones can no longer accurately signal caloric need[116]. The effect in this situation is a misperception of insufficient available energy stores[115]. This effect is in combination with increased pro-inflammatory cytokine levels[28], which makes it is unsurprising that chronic stress and chronodisruption are linked with the development of metabolic complications.

Hypercortisolaemia is also linked with affective disorders[10,27]. Mathematical modelling techniques have shown that individuals with major depressive disorder have disordered cortisol secretion with increased mass of cortisol in each pulse and increased pulse frequency[117]. Patients with major depressive disorder have long been known to exhibit glucocorticoid resistance in the form of resistance of endogenous cortisol to suppression in the dexamethasone and dexamethasone–CRH suppression tests[118], the results of which can predict clinical outcome[119]. Chronic exposure to stress results in a degree of limbic system atrophy, a key region involved in regulation of emotion[120]. This effect is also closely interlinked with immune function; stress induced proinflammatory cytokines, including IL-1β, IL-6 and TNF, are implicated in depressive behaviour in rodent models of depression and in depressed patients[100,104,121,122]. The mechanisms underlying this are complex. It is possible that inflammation and glucocorticoid signalling may act on the same processes to cause cumulative damage or that the dual pro and anti-inflammatory effects of glucocorticoids disrupt the brains immune system in favour of inflammatory effects[123]. In patients with depression, decreased T cell availability might arise secondary to increased apoptosis sensitivity combined with impaired glucocorticoid responsiveness and reduced brain trafficking capacity in response to the appropriate neuroendocrine or immune stimuli[100,121]. Stress also enhances NF-κB activation[105,106,124], indeed stress-

induced anhedonia has been found to be directly linked to increased NF-κB activity[104]. Interestingly, antidepressants can enhance GR functioning[125] and inhibit cytokine pathways[126].

These effects of glucocorticoids are usually accepted as simply being due to the high levels found in the blood. However, this theory might be an oversimplification. In primary hypocortisolaemia (or Addison disease), patients are treated with what we believe to be optimal replacement of hydrocortisone three times a day to mimic the circadian rhythm[127,128]. Despite this effort to give physiological replacement these patients have increased mortality as a result of cardiovascular disease, infectious disease and cancer and increased morbidity from fatigue and difficulties in concentrating[129-131]. This therapy fails to provide the anticipatory rise of cortisol prior to wakening and thus gives a shifted circadian profile while also resulting in acute nonphysiological levels of cortisol in the middle of the day[132]. Patients have altered immune responses in the form of altered circadian patterns of circulating monocytes and natural killer cells[133], impaired natural killer cell function[134] and hypomethylated CD4+ T cells[135]. In an attempt to rectify these adverse outcomes, modified and dual release preparations[133,136,137], as well as continuous[138] and pulsatile pump (The Pulses study, ISRCTN67193733) preparations are being trialled. Initial results are encouraging, with closer mirroring of circadian profiling resulting in alterations in clock gene function[139] and improvements in immune functioning[133] and quality of life[133,136,140,141]. These issues are not only limited to endogenous CORT, but also synthetic glucocorticoids[16]. These are some of the most widely prescribed medications[142,143] but despite their clinical efficacy over three quarters of patients experience adverse effects even at what would be considered a fairly low dose[144,145]. Chronic glucocorticoid treatment can also induce glucocorticoid resistance in susceptible patients with glucocorticoid responsive conditions such as asthma or rheumatoid arthritis[146].

[H1] Conclusions

Homeostatic processes are dynamic and interactive. As society changes with development of a 24 h world and its use of social media and changes in interpersonal communication, many of our

evolutionarily adaptive processes are in danger of becoming maladaptive. If we are to understand the growing epidemic of stress-related human disease, we need to go back to first principles and understand not only the mechanisms underlying the regulation of our patterns of physiological activity and hormone secretion — particularly over the important and notoriously difficult nadir period during sleep — but also why they are important for the maintenance of health. If we are going to aspire to an objective of optimal personalised medicine, we need to devise methods not only to measure dynamic basal patterns of hormonal metabolic and immune functioning over the whole day, but also novel therapeutic interventions to counteract the causes of environmental and/or stress related illness.

Glossary terms

Zeitgebers - Cues that entrain or synchronise the bodies 24-hour cycle

Circadian clock – Biochemical oscillator with phases synchronised with solar time

Circadian rhythm – Any biological process that displays an oscillation of approximately 24 hours

Ultradian rhythm – A biological rhythm that occurs with a frequency of <24 hours

Indirect projections – Neural pathways involving at least one relay

Hypophyseal portal system – The microcirculation allowing transport of hypothalamic hormones to the pituitary gland

Irradiance threshold – The threshold power of (solar) electromagnetic radiation need to exert an effect

Stereotypic behaviours – Repetitive body movements that serve no biological function

Goal directed behaviours – Behaviour engaged for a specific functional purpose

1. Szabo S, Tache Y, Somogyi A. The legacy of Hans Selye and the origins of stress research: a retrospective 75 years after his landmark brief "letter" to the editor# of nature. Stress 2012;15:472-8.
2. Levine S. Influence of psychological variables on the activity of the hypothalamic-pituitary-adrenal axis. European journal of pharmacology 2000;405:149-60.
3. Brown SA, Azzi A. Peripheral circadian oscillators in mammals. Handb Exp Pharmacol 2013:45-66.
4. Roenneberg T, Merrow M. The Circadian Clock and Human Health. Curr Biol 2016;26:R432-43.
5. Bass J, Lazar MA. Circadian time signatures of fitness and disease. Science 2016;354:994-9.
6. Turek FW. Circadian neural rhythms in mammals. Annu Rev Physiol 1985;47:49-64.
7. Skene DJ, Skornyakov E, Chowdhury NR, et al. Separation of circadian- and behavior-driven metabolite rhythms in humans provides a window on peripheral oscillators and metabolism. Proc Natl Acad Sci U S A 2018;115:7825-30.
8. Buhr ED, Takahashi JS. Molecular components of the Mammalian circadian clock. Handb Exp Pharmacol 2013:3-27.
9. Takahashi JS. Transcriptional architecture of the mammalian circadian clock. Nat Rev Genet 2017;18:164-79.
10. Baron KG, Reid KJ. Circadian misalignment and health. Int Rev Psychiatry 2014;26:139-54.
11. Potter GD, Skene DJ, Arendt J, Cade JE, Grant PJ, Hardie LJ. Circadian Rhythm and Sleep Disruption: Causes, Metabolic Consequences, and Countermeasures. Endocr Rev 2016;37:584-608.
12. Zhang R, Lahens NF, Ballance HI, Hughes ME, Hogenesch JB. A circadian gene expression atlas in mammals: implications for biology and medicine. Proc Natl Acad Sci U S A 2014;111:16219-24.
13. Smarr BL, Schirmer AE. 3.4 million real-world learning management system logins reveal the majority of students experience social jet lag correlated with decreased performance. Sci Rep 2018;8:4793.
14. Gardner M, Lightman S, Kuh D, et al. Dysregulation of the hypothalamic pituitary adrenal (HPA) axis and cognitive capability at older ages: individual participant meta-analysis of five cohorts. Sci Rep 2019;9:4555.
15. Selye H. Stress and the general adaptation syndrome. British medical journal 1950;1:1383-92.
16. Russell GM, Lightman SL. Can side effects of steroid treatments be minimized by the temporal aspects of delivery method? Expert opinion on drug safety 2014;13:1501-13.
17. Sorrells SF, Sapolsky RM. An inflammatory review of glucocorticoid actions in the CNS. Brain, behavior, and immunity 2007;21:259-72.
18. Busillo JM, Cidlowski JA. The five Rs of glucocorticoid action during inflammation: ready, reinforce, repress, resolve, and restore. Trends in endocrinology and metabolism: TEM 2013;24:109-19.
19. McEwen BS, Biron CA, Brunson KW, et al. The role of adrenocorticoids as modulators of immune function in health and disease: neural, endocrine and immune interactions. Brain Res Brain Res Rev 1997;23:79-133.
20. Elenkov IJ, Chrousos GP. Stress system--organization, physiology and immunoregulation. Neuroimmunomodulation 2006;13:257-67.
21. Brinkmann V, Kristofic C. Regulation by corticosteroids of Th1 and Th2 cytokine production in human CD4+ effector T cells generated from CD45RO- and CD45RO+ subsets. J Immunol 1995;155:3322-8.
22. Wiegers GJ, Reul JM. Induction of cytokine receptors by glucocorticoids: functional and pathological significance. Trends Pharmacol Sci 1998;19:317-21.

23. Abraham IM, Meerlo P, Luiten PG. Concentration dependent actions of glucocorticoids on neuronal viability and survival. Dose-response : a publication of International Hormesis Society 2006;4:38-54.
24. Plaschke K, Muller D, Hoyer S. Effect of adrenalectomy and corticosterone substitution on glucose and glycogen metabolism in rat brain. Journal of neural transmission 1996;103:89-100.
25. Belanoff JK, Gross K, Yager A, Schatzberg AF. Corticosteroids and cognition. Journal of psychiatric research 2001;35:127-45.
26. Roozendaal B. Stress and memory: opposing effects of glucocorticoids on memory consolidation and memory retrieval. Neurobiology of learning and memory 2002;78:578-95.
27. Brown ES. Effects of glucocorticoids on mood, memory, and the hippocampus. Treatment and preventive therapy. Annals of the New York Academy of Sciences 2009;1179:41-55.
28. de Kloet ER, Oitzl MS, Joels M. Stress and cognition: are corticosteroids good or bad guys? Trends in neurosciences 1999;22:422-6.
29. Decani S, Federighi V, Baruzzi E, Sardella A, Lodi G. Iatrogenic Cushing's syndrome and topical steroid therapy: case series and review of the literature. The Journal of dermatological treatment 2014;25:495-500.
30. Kenna HA, Poon AW, de los Angeles CP, Koran LM. Psychiatric complications of treatment with corticosteroids: review with case report. Psychiatry and clinical neurosciences 2011;65:549-60.
31. Buijs RM, Markman M, Nunes-Cardoso B, Hou YX, Shinn S. Projections of the suprachiasmatic nucleus to stress-related areas in the rat hypothalamus: a light and electron microscopic study. The Journal of comparative neurology 1993;335:42-54.
32. Watts AG, Swanson LW. Efferent projections of the suprachiasmatic nucleus: II. Studies using retrograde transport of fluorescent dyes and simultaneous peptide immunohistochemistry in the rat. The Journal of comparative neurology 1987;258:230-52.
33. Jacobson L. Hypothalamic-pituitary-adrenocortical axis regulation. Endocrinology and metabolism clinics of North America 2005;34:271-92, vii.
34. Herman JP, Ostrander MM, Mueller NK, Figueiredo H. Limbic system mechanisms of stress regulation: hypothalamo-pituitary-adrenocortical axis. Progress in neuro-psychopharmacology & biological psychiatry 2005;29:1201-13.
35. Dallman MF, Akana SF, Levin N, et al. Corticosteroids and the control of function in the hypothalamo-pituitary-adrenal (HPA) axis. Annals of the New York Academy of Sciences 1994;746:22-31; discussion -2, 64-7.
36. Jasper MS, Engeland WC. Splanchnic neural activity modulates ultradian and circadian rhythms in adrenocortical secretion in awake rats. Neuroendocrinology 1994;59:97-109.
37. Buijs RM, Wortel J, Van Heerikhuize JJ, et al. Anatomical and functional demonstration of a multisynaptic suprachiasmatic nucleus adrenal (cortex) pathway. The European journal of neuroscience 1999;11:1535-44.
38. Kiessling S, Sollars PJ, Pickard GE. Light stimulates the mouse adrenal through a retinohypothalamic pathway independent of an effect on the clock in the suprachiasmatic nucleus. PloS one 2014;9:e92959.
39. Husse J, Leliavski A, Tsang AH, Oster H, Eichele G. The light-dark cycle controls peripheral rhythmicity in mice with a genetically ablated suprachiasmatic nucleus clock. FASEB journal : official publication of the Federation of American Societies for Experimental Biology 2014.
40. Ishida A, Mutoh T, Ueyama T, et al. Light activates the adrenal gland: timing of gene expression and glucocorticoid release. Cell metabolism 2005;2:297-307.
41. Oster H, Damerow S, Kiessling S, et al. The circadian rhythm of glucocorticoids is regulated by a gating mechanism residing in the adrenal cortical clock. Cell metabolism 2006;4:163-73.
42. Bailey SL, Heitkemper MM. Circadian rhythmicity of cortisol and body temperature: morningness-eveningness effects. Chronobiology international 2001;18:249-61.

43. Donner NC, Montoya CD, Lukkes JL, Lowry CA. Chronic non-invasive corticosterone administration abolishes the diurnal pattern of tph2 expression. Psychoneuroendocrinology 2012;37:645-61.
44. Lightman SL. The neuroendocrinology of stress: a never ending story. J Neuroendocrinol 2008;20:880-4.
45. Nicolaides NC, Kyratzi E, Lamprokostopoulou A, Chrousos GP, Charmandari E. Stress, the stress system and the role of glucocorticoids. Neuroimmunomodulation 2015;22:6-19.
46. Gibbison B, Spiga F, Walker JJ, et al. Dynamic pituitary-adrenal interactions in response to cardiac surgery. Crit Care Med 2015;43:791-800.
47. Spiga F, Zavala E, Walker JJ, Zhao Z, Terry JR, Lightman SL. Dynamic responses of the adrenal steroidogenic regulatory network. Proc Natl Acad Sci U S A 2017;114:E6466-E74.
48. Ma XM, Levy A, Lightman SL. Emergence of an isolated arginine vasopressin (AVP) response to stress after repeated restraint: a study of both AVP and corticotropin-releasing hormone messenger ribonucleic acid (RNA) and heteronuclear RNA. Endocrinology 1997;138:4351-7.
49. Dallman MF. Stress update Adaptation of the hypothalamic-pituitary-adrenal axis to chronic stress. Trends in endocrinology and metabolism: TEM 1993;4:62-9.
50. Henley DE, Russell GM, Douthwaite JA, et al. Hypothalamic-pituitary-adrenal axis activation in obstructive sleep apnea: the effect of continuous positive airway pressure therapy. The Journal of clinical endocrinology and metabolism 2009;94:4234-42.
51. Boonen E, Vervenne H, Meersseman P, et al. Reduced cortisol metabolism during critical illness. The New England journal of medicine 2013;368:1477-88.
52. Peeters B, Meersseman P, Vander Perre S, et al. Adrenocortical function during prolonged critical illness and beyond: a prospective observational study. Intensive Care Med 2018;44:1720-9.
53. De Kloet ER, Vreugdenhil E, Oitzl MS, Joels M. Brain corticosteroid receptor balance in health and disease. Endocr Rev 1998;19:269-301.
54. Reul JM, de Kloet ER. Two receptor systems for corticosterone in rat brain: microdistribution and differential occupation. Endocrinology 1985;117:2505-11.
55. Dallman MF. Fast glucocorticoid actions on brain: back to the future. Frontiers in neuroendocrinology 2005;26:103-8.
56. Russell GM, Henley DE, Leendertz J, et al. Rapid glucocorticoid receptor-mediated inhibition of hypothalamic-pituitary-adrenal ultradian activity in healthy males. The Journal of neuroscience : the official journal of the Society for Neuroscience 2010;30:6106-15.
57. Lowenberg M, Verhaar AP, van den Brink GR, Hommes DW. Glucocorticoid signaling: a nongenomic mechanism for T-cell immunosuppression. Trends in molecular medicine 2007;13:158-63.
58. Orchinik M, Murray TF, Franklin PH, Moore FL. Guanyl nucleotides modulate binding to steroid receptors in neuronal membranes. Proc Natl Acad Sci U S A 1992;89:3830-4.
59. Orchinik M, Murray TF, Moore FL. A corticosteroid receptor in neuronal membranes. Science 1991;252:1848-51.
60. Joels M, Pasricha N, Karst H. The interplay between rapid and slow corticosteroid actions in brain. European journal of pharmacology 2013;719:44-52.
61. Walker JJ, Spiga F, Waite E, et al. The origin of glucocorticoid hormone oscillations. PLoS biology 2012;10:e1001341.
62. Patel PD, Lopez JF, Lyons DM, Burke S, Wallace M, Schatzberg AF. Glucocorticoid and mineralocorticoid receptor mRNA expression in squirrel monkey brain. Journal of psychiatric research 2000;34:383-92.
63. Groeneweg FL, Karst H, de Kloet ER, Joels M. Rapid non-genomic effects of corticosteroids and their role in the central stress response. The Journal of endocrinology 2011;209:153-67.
64. de Kloet ER, Fitzsimons CP, Datson NA, Meijer OC, Vreugdenhil E. Glucocorticoid signaling and stress-related limbic susceptibility pathway: about receptors, transcription machinery and microRNA. Brain research 2009;1293:129-41.

65. Russell GM, Kalafatakis K, Lightman SL. The importance of biological oscillators for HPA activity and tissue glucocorticoid response: Coordinating stress and neurobehavioural adaptation. J Neuroendocrinol 2014.
66. Lewis JG, Mopert B, Shand BI, et al. Plasma variation of corticosteroid-binding globulin and sex hormone-binding globulin. Hormone and metabolic research = Hormon- und Stoffwechselforschung = Hormones et metabolisme 2006;38:241-5.
67. Lewis JG, Bagley CJ, Elder PA, Bachmann AW, Torpy DJ. Plasma free cortisol fraction reflects levels of functioning corticosteroid-binding globulin. Clin Chim Acta 2005;359:189-94.
68. Hammond GL, Smith CL, Underhill DA. Molecular studies of corticosteroid binding globulin structure, biosynthesis and function. J Steroid Biochem Mol Biol 1991;40:755-62.
69. Frairia R, Agrimonti F, Fortunati N, Fazzari A, Gennari P, Berta L. Influence of naturally occurring and synthetic glucocorticoids on corticosteroid-binding globulin-steroid interaction in human peripheral plasma. Annals of the New York Academy of Sciences 1988;538:287-303.
70. Cameron A, Henley D, Carrell R, Zhou A, Clarke A, Lightman S. Temperature-responsive release of cortisol from its binding globulin: a protein thermocouple. The Journal of clinical endocrinology and metabolism 2010;95:4689-95.
71. Kyrou I, Chrousos GP, Tsigos C. Stress, visceral obesity, and metabolic complications. Annals of the New York Academy of Sciences 2006;1083:77-110.
72. Chapman K, Holmes M, Seckl J. 11beta-hydroxysteroid dehydrogenases: intracellular gate-keepers of tissue glucocorticoid action. Physiol Rev 2013;93:1139-206.
73. Seckl JR. 11beta-hydroxysteroid dehydrogenases: changing glucocorticoid action. Curr Opin Pharmacol 2004;4:597-602.
74. Verma M, Kipari TMJ, Zhang Z, et al. 11beta-hydroxysteroid dehydrogenase-1 deficiency alters brain energy metabolism in acute systemic inflammation. Brain, behavior, and immunity 2018;69:223-34.
75. Follenius M, Simon C, Brandenberger G, Lenzi P. Ultradian plasma corticotropin and cortisol rhythms: time-series analyses. Journal of endocrinological investigation 1987;10:261-6.
76. Hartmann A, Veldhuis JD, Deuschle M, Standhardt H, Heuser I. Twenty-four hour cortisol release profiles in patients with Alzheimer's and Parkinson's disease compared to normal controls: ultradian secretory pulsatility and diurnal variation. Neurobiology of aging 1997;18:285-9.
77. Rivest RW, Schulz P, Lustenberger S, Sizonenko PC. Differences between circadian and ultradian organization of cortisol and melatonin rhythms during activity and rest. The Journal of clinical endocrinology and metabolism 1989;68:721-9.
78. Waite EJ, McKenna M, Kershaw Y, et al. Ultradian corticosterone secretion is maintained in the absence of circadian cues. The European journal of neuroscience 2012;36:3142-50.
79. Ixart G, Barbanel G, Nouguier-Soule J, Assenmacher I. A quantitative study of the pulsatile parameters of CRH-41 secretion in unanesthetized free-moving rats. Experimental brain research 1991;87:153-8.
80. Spiga F, Waite EJ, Liu Y, Kershaw YM, Aguilera G, Lightman SL. ACTH-dependent ultradian rhythm of corticosterone secretion. Endocrinology 2011;152:1448-57.
81. Spiga F, Liu Y, Aguilera G, Lightman SL. Temporal effect of adrenocorticotrophic hormone on adrenal glucocorticoid steroidogenesis: involvement of the transducer of regulated cyclic AMP-response element-binding protein activity. J Neuroendocrinol 2011;23:136-42.
82. Lim C, Allada R. Emerging roles for post-transcriptional regulation in circadian clocks. Nat Neurosci 2013;16:1544-50.
83. Liston C, Cichon JM, Jeanneteau F, Jia Z, Chao MV, Gan WB. Circadian glucocorticoid oscillations promote learning-dependent synapse formation and maintenance. Nat Neurosci 2013;16:698-705.
84. Lightman SL, Conway-Campbell BL. The crucial role of pulsatile activity of the HPA axis for continuous dynamic equilibration. Nature reviews Neuroscience 2010;11:710-8.

85. Stavreva DA, Wiench M, John S, et al. Ultradian hormone stimulation induces glucocorticoid receptor-mediated pulses of gene transcription. Nature cell biology 2009;11:1093-102.
86. Conway-Campbell BL, Pooley JR, Hager GL, Lightman SL. Molecular dynamics of ultradian glucocorticoid receptor action. Molecular and cellular endocrinology 2012;348:383-93.
87. George CL, Lightman SL, Biddie SC. Transcription factor interactions in genomic nuclear receptor function. Epigenomics 2011;3:471-85.
88. So AY, Chaivorapol C, Bolton EC, Li H, Yamamoto KR. Determinants of cell- and gene-specific transcriptional regulation by the glucocorticoid receptor. PLoS genetics 2007;3:e94.
89. Zalachoras I, Houtman R, Meijer OC. Understanding stress-effects in the brain via transcriptional signal transduction pathways. Neuroscience 2013;242:97-109.
90. Sarabdjitsingh RA, Conway-Campbell BL, Leggett JD, et al. Stress responsiveness varies over the ultradian glucocorticoid cycle in a brain-region-specific manner. Endocrinology 2010;151:5369-79.
91. Sarabdjitsingh RA, Jezequel J, Pasricha N, et al. Ultradian corticosterone pulses balance glutaminergic transmission and synaptic plasticity. Proc Natl Acad Sci U S A 2014.
92. Kalafatakis K, Russell GM, Harmer CJ, et al. Ultradian rhythmicity of plasma cortisol is necessary for normal emotional and cognitive responses in man. Proc Natl Acad Sci U S A 2018;115:E4091-E100.
93. Gjerstad JK, Lightman SL, Spiga F. Role of glucocorticoid negative feedback in the regulation of HPA axis pulsatility. Stress 2018:1-14.
94. Bornstein SR, Engeland WC, Ehrhart-Bornstein M, Herman JP. Dissociation of ACTH and glucocorticoids. Trends in endocrinology and metabolism: TEM 2008;19:175-80.
95. Silverman MN, Miller AH, Biron CA, Pearce BD. Characterization of an interleukin-6- and adrenocorticotropin-dependent, immune-to-adrenal pathway during viral infection. Endocrinology 2004;145:3580-9.
96. Franchimont D, Bouma G, Galon J, et al. Adrenal cortical activation in murine colitis. Gastroenterology 2000;119:1560-8.
97. Viblanc VA, Schull Q, Cornioley T, et al. An integrative appraisal of the hormonal and metabolic changes induced by acute stress using king penguins as a model. Gen Comp Endocrinol 2018;269:1-10.
98. Cruz-Topete D, Cidlowski JA. One hormone, two actions: anti- and pro-inflammatory effects of glucocorticoids. Neuroimmunomodulation 2015;22:20-32.
99. Biddie SC, Conway-Campbell BL, Lightman SL. Dynamic regulation of glucocorticoid signalling in health and disease. Rheumatology 2012;51:403-12.
100. Miller GE, Cohen S, Ritchey AK. Chronic psychological stress and the regulation of pro-inflammatory cytokines: a glucocorticoid-resistance model. Health psychology : official journal of the Division of Health Psychology, American Psychological Association 2002;21:531-41.
101. Oster H, Challet E, Ott V, et al. The Functional and Clinical Significance of the 24-Hour Rhythm of Circulating Glucocorticoids. Endocr Rev 2017;38:3-45.
102. Keller M, Mazuch J, Abraham U, et al. A circadian clock in macrophages controls inflammatory immune responses. Proc Natl Acad Sci U S A 2009;106:21407-12.
103. Boivin DB, James FO, Wu A, Cho-Park PF, Xiong H, Sun ZS. Circadian clock genes oscillate in human peripheral blood mononuclear cells. Blood 2003;102:4143-5.
104. Koo JW, Russo SJ, Ferguson D, Nestler EJ, Duman RS. Nuclear factor-kappaB is a critical mediator of stress-impaired neurogenesis and depressive behavior. Proc Natl Acad Sci U S A 2010;107:2669-74.
105. Pace TW, Hu F, Miller AH. Cytokine-effects on glucocorticoid receptor function: relevance to glucocorticoid resistance and the pathophysiology and treatment of major depression. Brain, behavior, and immunity 2007;21:9-19.

106. Pace TW, Mletzko TC, Alagbe O, et al. Increased stress-induced inflammatory responses in male patients with major depression and increased early life stress. The American journal of psychiatry 2006;163:1630-3.
107. Cohen S, Janicki-Deverts D, Doyle WJ, et al. Chronic stress, glucocorticoid receptor resistance, inflammation, and disease risk. Proc Natl Acad Sci U S A 2012;109:5995-9.
108. Spiegel K, Leproult R, Van Cauter E. Impact of sleep debt on metabolic and endocrine function. Lancet 1999;354:1435-9.
109. Hauner H, Schmid P, Pfeiffer EF. Glucocorticoids and insulin promote the differentiation of human adipocyte precursor cells into fat cells. The Journal of clinical endocrinology and metabolism 1987;64:832-5.
110. Dallman MF, Pecoraro NC, La Fleur SE, et al. Glucocorticoids, chronic stress, and obesity. Prog Brain Res 2006;153:75-105.
111. Tsigos C, Papanicolaou DA, Kyrou I, Defensor R, Mitsiadis CS, Chrousos GP. Dose-dependent effects of recombinant human interleukin-6 on glucose regulation. The Journal of clinical endocrinology and metabolism 1997;82:4167-70.
112. McEwen BS. Sleep deprivation as a neurobiologic and physiologic stressor: Allostasis and allostatic load. Metabolism 2006;55:S20-3.
113. Zhu B, Shi C, Park CG, Zhao X, Reutrakul S. Effects of sleep restriction on metabolism-related parameters in healthy adults: A comprehensive review and meta-analysis of randomized controlled trials. Sleep Med Rev 2019;45:18-30.
114. Gavrila A, Peng CK, Chan JL, Mietus JE, Goldberger AL, Mantzoros CS. Diurnal and ultradian dynamics of serum adiponectin in healthy men: comparison with leptin, circulating soluble leptin receptor, and cortisol patterns. The Journal of clinical endocrinology and metabolism 2003;88:2838-43.
115. Knutson KL, Van Cauter E. Associations between sleep loss and increased risk of obesity and diabetes. Annals of the New York Academy of Sciences 2008;1129:287-304.
116. Adam TC, Epel ES. Stress, eating and the reward system. Physiol Behav 2007;91:449-58.
117. Young EA, Carlson NE, Brown MB. Twenty-four-hour ACTH and cortisol pulsatility in depressed women. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 2001;25:267-76.
118. Heuser I, Yassouridis A, Holsboer F. The combined dexamethasone/CRH test: a refined laboratory test for psychiatric disorders. Journal of psychiatric research 1994;28:341-56.
119. Ising M, Kunzel HE, Binder EB, Nickel T, Modell S, Holsboer F. The combined dexamethasone/CRH test as a potential surrogate marker in depression. Progress in neuro-psychopharmacology & biological psychiatry 2005;29:1085-93.
120. Krishnan V, Nestler EJ. The molecular neurobiology of depression. Nature 2008;455:894-902.
121. Miller AH. Depression and immunity: a role for T cells? Brain, behavior, and immunity 2010;24:1-8.
122. Koo JW, Duman RS. IL-1beta is an essential mediator of the antineurogenic and anhedonic effects of stress. Proc Natl Acad Sci U S A 2008;105:751-6.
123. Horowitz MA, Zunszain PA, Anacker C, Musaelyan K, Pariante CM. Glucocorticoids and inflammation: a double-headed sword in depression? How do neuroendocrine and inflammatory pathways interact during stress to contribute to the pathogenesis of depression? Mod Trends Pharmacopsychiatry 2013;28:127-43.
124. Munhoz CD, Lepsch LB, Kawamoto EM, et al. Chronic unpredictable stress exacerbates lipopolysaccharide-induced activation of nuclear factor-kappaB in the frontal cortex and hippocampus via glucocorticoid secretion. The Journal of neuroscience : the official journal of the Society for Neuroscience 2006;26:3813-20.
125. Pariante CM. Glucocorticoid receptor function in vitro in patients with major depression. Stress 2004;7:209-19.

126. Kenis G, Maes M. Effects of antidepressants on the production of cytokines. Int J Neuropsychopharmacol 2002;5:401-12.
127. Bjornsdottir S, Sundstrom A, Ludvigsson JF, Blomqvist P, Kampe O, Bensing S. Drug prescription patterns in patients with Addison's disease: a Swedish population-based cohort study. The Journal of clinical endocrinology and metabolism 2013;98:2009-18.
128. Dunlop D. Eighty-Six Cases of Addison's Disease. British medical journal 1963;2:887-91.
129. Giordano R, Marzotti S, Balbo M, et al. Metabolic and cardiovascular profile in patients with Addison's disease under conventional glucocorticoid replacement. Journal of endocrinological investigation 2009;32:917-23.
130. Johannsson G, Falorni A, Skrtic S, et al. Adrenal insufficiency: review of clinical outcomes with current glucocorticoid replacement therapy. Clinical endocrinology 2015;82:2-11.
131. Lovas K, Loge JH, Husebye ES. Subjective health status in Norwegian patients with Addison's disease. Clinical endocrinology 2002;56:581-8.
132. Feek CM, Ratcliffe JG, Seth J, Gray CE, Toft AD, Irvine WJ. Patterns of plasma cortisol and ACTH concentrations in patients with Addison's disease treated with conventional corticosteroid replacement. Clinical endocrinology 1981;14:451-8.
133. Isidori AM, Venneri MA, Graziadio C, et al. Effect of once-daily, modified-release hydrocortisone versus standard glucocorticoid therapy on metabolism and innate immunity in patients with adrenal insufficiency (DREAM): a single-blind, randomised controlled trial. The lancet Diabetes & endocrinology 2018;6:173-85.
134. Bancos I, Hazeldine J, Chortis V, et al. Primary adrenal insufficiency is associated with impaired natural killer cell function: a potential link to increased mortality. European journal of endocrinology / European Federation of Endocrine Societies 2017;176:471-80.
135. Bjanesoy TE, Andreassen BK, Bratland E, et al. Altered DNA methylation profile in Norwegian patients with Autoimmune Addison's Disease. Molecular immunology 2014;59:208-16.
136. Langenheim J, Ventz M, Hinz A, Quinkler M. Modified-release prednisone decreases complaints and fatigue compared to standard prednisolone in patients with adrenal insufficiency. Hormone and metabolic research = Hormon- und Stoffwechselforschung = Hormones et metabolisme 2013;45:96-101.
137. Mallappa A, Sinaii N, Kumar P, et al. A phase 2 study of Chronocort, a modified-release formulation of hydrocortisone, in the treatment of adults with classic congenital adrenal hyperplasia. The Journal of clinical endocrinology and metabolism 2015;100:1137-45.
138. Lovas K, Husebye ES. Continuous subcutaneous hydrocortisone infusion in Addison's disease. European journal of endocrinology / European Federation of Endocrine Societies 2007;157:109-12.
139. Venneri MA, Hasenmajer V, Fiore D, et al. Circadian Rhythm of Glucocorticoid Administration Entrains Clock Genes in Immune Cells: A DREAM Trial Ancillary Study. The Journal of clinical endocrinology and metabolism 2018;103:2998-3009.
140. Oksnes M, Bjornsdottir S, Isaksson M, et al. Continuous subcutaneous hydrocortisone infusion versus oral hydrocortisone replacement for treatment of addison's disease: a randomized clinical trial. The Journal of clinical endocrinology and metabolism 2014;99:1665-74.
141. Riedel M, Wiese A, Schurmeyer TH, Brabant G. Quality of life in patients with Addison's disease: effects of different cortisol replacement modes. Experimental and clinical endocrinology 1993;101:106-11.
142. van Staa TP, Leufkens HG, Abenhaim L, Begaud B, Zhang B, Cooper C. Use of oral corticosteroids in the United Kingdom. QJM : monthly journal of the Association of Physicians 2000;93:105-11.
143. Overman RA, Yeh JY, Deal CL. Prevalence of oral glucocorticoid usage in the United States: a general population perspective. Arthritis care & research 2013;65:294-8.
144. Curtis JR, Westfall AO, Allison J, et al. Population-based assessment of adverse events associated with long-term glucocorticoid use. Arthritis and rheumatism 2006;55:420-6.

145. McDonough AK, Curtis JR, Saag KG. The epidemiology of glucocorticoid-associated adverse events. Current opinion in rheumatology 2008;20:131-7.
146. Leung DY, Bloom JW. Update on glucocorticoid action and resistance. The Journal of allergy and clinical immunology 2003;111:3-22; quiz 3.

Author contributions

The authors contributed equally to all aspects of the article.

Competing interests

The authors declare no competing interests.

Publisher's note

Figure 1 | Co-ordination of central and peripheral clocks by glucocorticoids. The SCN central clock receives light dark signals which in turn influences HPA and SAM activity leading to circadian CORT production. CORT activates glucocorticoid receptors in peripheral tissues which synchronises peripheral clocks and downstream metabolic, cardiovascular, neuronal and immune pathways. Other zeitgebers such as food, temperature and social cues can also entrain or influence the entrainment of clocks and can alter the output of these downstream pathways.

[Au: Please note that we need you to fill out a third party rights table for all display items that have been published elsewhere so that we can obtain the appropriate permissions. A link to the form is included in my covering email, but please let me know if you have any problems accessing or completing the form. Note that we cannot include unpublished findings in display items.]

Figure 2 | Human cortisol ultradian rhythmicity under basal and stressful conditions. Cortisol is well known to be secreted in a circadian rhythm characterised by a nadir or quiescent phase during the inactive period (overnight whilst sleeping in humans). An anticipatory CORT rise occurs before waking followed by the circadian peak of secretion at the start of the active period. Underlying this rhythm, however, is a complex and dynamic ultradian rhythm consisting of pulses approximately every 3 hours. (A) A schematic representation of a circadian and ultradian cortisol rhythm from a healthy male volunteer. (B) The effect of acute stress of cardiac surgery (shaded area shows when coronary artery bypass grafting occurred– taken from Gibbison et al Critical Care 2015). Cortisol levels remain elevated throughout the sampling period. This occurs via an initial surge in ACTH, followed post operatively by altered adrenal sensitivity to ACTH. **[Au: Are parts a and b the wrong way around in the figure? That is, the original figure's part c (a here) shows cardiac surgery and**

part d (b here) shows the healthy volunteer? The grey area would then indicate the cardiac surgery – YES grey area is surgery in the original pptx slide C is the cardiac surgery (b) and D is the healthy volunteer (a)?] (C) The effect of chronic stress on HPA axis activity in the form of obstructive sleep apnoea, taken from Henley et al JCEM 2009. Here, disordered cortisol pulsatility is seen in the form of an increase in the number of and amount of cortisol secreted per pulse at baseline (before CPAP therapy), especially over the quiescent period when endogenous cortisol levels naturally fall. The profiles improve after 3 months of continuous positive airways pressure therapy. **[Au: As per your figure sketch, I have asked our Art Editor to just include the top 2 panels from the original figure 1b – correct? yes]**

Figure 3 | Schematic representation of the HPA axis showing natural inbuilt adrenal delays. After release from the pituitary gland ACTH binds to adrenal Melanocortin 2 receptors. Here natural inbuilt delays occur; MC2R binding leads to increased uptake of cholesterol, the precursor for all steroid hormones. Cholesterol then undergoes a series of biosynthetic steps before being transported into mitochondria, undergoing hydroxylation and the subsequent release of cortisol into the circulation. **[Au: Please include a full figure legend, giving more detail on what is depicted in the legend.] [Au: I am unsure that is meant in the delay section of the figure – please could you clarify what you would like us to redraw here? E.g, by labelling the different parts. – new figure supplied]**

Figure 4 | Importance of gene pulsing. (A) In adrenalectomized rats, hourly pulses of corticosterone result in a similar pattern of association–dissociation kinetics in GR–DNA binding and downstream cyclic recruitment and pulsatile release of the clock gene *Per1* nascent RNA. This effect is not seen with synthetic glucocorticoid treatment (taken from Stavreva et al NCB 2009) B) Ultradian and constant corticosterone treatment have different effects on transcription of Tsc22d3, a GR-regulated gene. Here, pulsatile presentation shows a pulsatile pattern of gene fold induction, whilst constant infusion induces constant RNA release (taken from Stavreva et al NCB 2009). C, control; P, induction; W, withdrawal. (C) Adrenalectomised rats received either pulsatile corticosterone (50 ng/ml or 100ng/ml peaks) or constant corticosterone (12-h constant infusion clamped at 50 ng/ml). They then underwent a 10-minute noise stress. C-fos mRNA expression was analysed in different brain regions. A differential response was found in the pituitary, amygdala, hippocampus and PVN. The effect shown was dependent on both phase and amplitude of the glucocorticoid pulse, which suggests that a differential response is seen in stress circuitry depending on the pattern of exposure (taken from Sarabdjitsingh et al Endocrinology 2010). **[Au: As per your sketch, I have asked our Art Editor to just**

include part c from the original figure, OK? - YES] (D) Healthy male volunteers underwent a chemical adrenalectomy with metyrapone. In a cross over trial design they then received hydrocortisone replacement therapy (total daily dose 20mg) as either standard oral (three times a day), constant circadian subcutaneous infusion or constant pulsatile subcutaneous infusion for 5 days. On day 5, participants underwent functional MRI whilst performing a facial recognition task (FERT). Blood Oxygen level Dependent (BOLD) imaging showed a differential response in attentional bias and recognition of emotional faces depending on whether a pulsatile or a non-pulsatile pattern of glucocorticoid presentation was used. Communication networks within the insula, striatum and amygdala also varied depending on whether the presentation was pulsatile or non-pulsatile. (taken from Kalafatakis et al PNAS 2018)