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Review

The cortisol awakening response: More than a measure of HPA axis function

Angela Clow [a,*], Frank Hucklebridge [b], Tobias Stalder [a,c], Phil Evans [a], Lisa Thorn [a]

[a] Psychophysiology and Stress Research Group, Department of Psychology, University of Westminster, 309 Regent Street, London W1B 2UW, UK
[b] Psychophysiology and Stress Research Group, Department of Human and Health Sciences, University of Westminster, 115 New Cavendish Street, London W1W 6UW, UK
[c] Department of Biological Psychology, Faculty of Science, Technical University of Dresden, 01062 Dresden, Germany

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ABSTRACT

In most healthy people morning awakening is associated with a burst of cortisol secretion: the cortisol awakening response (CAR). It is argued that the CAR is subject to a range physiological regulatory influences that facilitate this rapid increase in cortisol secretion. Evidence is presented for reduced adrenal sensitivity to rising levels of ACTH in the pre-awakening period, mediated by an extra-pituitary pathway to the adrenal from the suprachiasmatic nucleus (SCN). A role for the hippocampus in this pre-awakening regulation of cortisol secretion is considered. Attainment of consciousness is associated with 'flip-flop' switching of regional brain activation, which, it is argued, initiates a combination of processes: (1) activation of the hypothalamic pituitary adrenal (HPA) axis; (2) release of pre-awakening reduced adrenal sensitivity to ACTH; (3) increased post-awakening adrenal sensitivity to ACTH in response to light, mediated by a SCN extra-pituitary pathway. An association between the CAR and the ending of sleep inertia is discussed.

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1. Introduction

The cortisol awakening response (CAR), the rapid increase in cortisol levels following morning awakening, was first described in the mid 1990s (Pruessner et al., 1997). Since then, research into this aspect of the circadian cortisol cycle in humans has flourished although findings regarding associations with the CAR have frequently been inconsistent (see Clow et al., 2004; Fries et al., 2009, for reviews). A high proportion of research in this area has been approached from a psychological perspective, i.e. exploration of the CAR in relation to a spectrum of individual difference measures. Although there is some level of understanding, there has been less research from a physiological perspective, i.e. systematic exploration into the regulation of the CAR in order to illuminate meaning and functional significance and the impact of dysregulation in terms of pathology. This paper describes some potentially interconnected physiological processes that occur during the immediate pre- and post-awakening period and attempts, sometimes speculatively, to relate these to available evidence about the regulation of the CAR. The overall purpose is to try to make sense of interacting physiological influences at the same time as identifying gaps in understanding that warrant further investigation. It is hoped that this overview of physiological processes will facilitate a fuller understanding of CAR regulation and enlighten its contribution to biopsychosocial research.

2. The transition from sleep to awakening

The cortisol awakening response is what the name implies: a physiological response to awakening (Wilhelm et al., 2007). It is a

* Corresponding author. Fax: +44 0207 7911 5106.
E-mail address: clowa@wmin.ac.uk (A. Clow).

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discreet and distinct component of the cortisol circadian cycle, with characteristics unrelated to those of cortisol secretion throughout the rest of the day (see Clow et al., 2004; Fries et al., 2009). In order to gain insight into potential regulatory influences it seems appropriate to examine the CAR in relation to other physiological processes also initiated by the awakening process.

Awakening from sleep involves a rapid attainment of consciousness followed by the relatively slow re-establishment of full alertness, some 20–30 min later (Balkin et al., 2002). The awakening process is associated with rapid reciprocal switching between activation in cortical and sub-cortical brain regions. This process, is referred to as 'flip-flop' switching, and is similar to the transition between various stages of sleep, i.e. REM/non-REM (Braun et al., 1997; Lu et al., 2006; Saper et al., 2001; Sil'kis, 2009). Such flip-flop switching of brain areas arises from activation of mutually inhibitory neuronal pathways orchestrated by a control system in the pontine brainstem (Lu et al., 2006). Reciprocal interconnections between aminergic inhibitory neurons and cholinergic excitatory neurons form the basis of this flip-flop switching: the activity of aminergic neurons is highest during waking and is lowest during REM sleep, whereas the activity of cholinergic cells shows the reverse pattern (see Hobson, 2009). Although full recovery from sleep takes upwards of 30 min it is clear that the shift from sleep to consciousness involves a rapid switch off of some areas of sub-cortical brain activation. Notably the hippocampus (along with the pons, anterior cingulate, caudate, midbrain, and parahippocampal gyrus) is especially active during REM sleep and becomes inhibited during non-REM sleep and upon awakening (Balkin et al., 2002; Sil'kis, 2009). It is plausible that this switching of brain circuitry associated with the transition between sleep and consciousness may also be associated with initiation of the CAR as it is known to be actively initiated by the process of awakening (Spathschwalbe et al., 1992; Vancauter et al., 1994; Wilhelm et al., 2007). To enlighten linkages between the CAR and awakening requires an understanding of the regulatory processes that influence cortisol secretion both pre- and post-this central 'tipping point'.

3. Pre-awakening influences on cortisol secretion

Circadian influences on physiological systems are mainly transmitted via the body's endogenous central pacemaker: the suprachiasmatic nucleus (SCN). The circadian rhythm of the HPA axis is largely controlled by the SCN, which influences adrenocortical activity via input to the paraventricular nuclei of the hypothalamus (Buijs et al., 2003; Dickmeis, 2009; Kalsbeek et al., 2006; Krout et al., 2002). These patterns of cortisol secretion result from changes in an underlying much faster ultradian rhythm: pulses of cortisol secretion that occur approximately once an hour (Lightman, 2008; Windle et al., 1998). Differences in amplitude (not frequency) of these pulses predominantly account for differing cortisol concentrations across the day/night cycle (Windle et al., 1998). Under the influence of the SCN HPA axis activity gradually increases towards the end of nighttime sleep. In particular the HPA axis secretes ACTH and cortisol during episodes of non-REM sleep when the hippocampus is inhibited (Buijs et al., 2003; Wilhelm et al., 2007).

However, activation of the HPA axis is not the sole regulatory pathway for cortisol secretion. Evidence for this lies in the fact that levels of pituitary-derived ACTH can be dissociated from levels of cortisol. For example in non-stressed rodents the diurnal rhythm of corticosterone is of high amplitude, typically of 5- to 10-fold from the acrophase to the nadir, whereas the rhythm of plasma ACTH is of low amplitude (up to 2-fold) (Kalsbeek et al., 2006; Ulrich-Lai et al., 2006). This observation has been accounted for by a coincident rhythm in adrenal sensitivity to ACTH, with peak sensitivity occurring at the onset of the active phase and a trough in ACTH sensitivity during the inactive phase (Kaneko et al., 1981). Changes in adrenal sensitivity to ACTH (both increases and decreases) have been linked with time of day and the SCN (Bornstein et al., 2008; Buijs et al., 1997, 2003) and are mediated via sympathetic innervation of the adrenal gland by the splanchnic nerve (Edwards and Jones, 1993; Ehrhart-Bornstein et al., 1998; Engeland and Arnhold, 2005; Sage et al., 2002; Ulrich-Lai et al., 2006). SCN-mediated extra-pituitary regulation of cortisol secretion is reported to be particularly important with regards to the fine-tuning of circadian influences (Buijs et al., 2003; Dickmeis, 2009; Ishida et al., 2005; Ulrich-Lai et al., 2006) and is additional to its influence on HPA axis-dependent cortisol secretion via circadian regulation of levels of ACTH.

In order to fully understand the interaction between HPA axis and extra-pituitary regulatory input to the CAR, it is necessary to examine relationships between cortisol and its pituitary-derived secretagogue ACTH. Unfortunately most CAR studies in humans do not measure ACTH levels as it cannot be measured in saliva samples. Furthermore, most studies of the CAR are undertaken in the domestic setting and do not capture the period immediately prior to awakening. However, a recent well-controlled study in humans measured ACTH and cortisol both before and after awakening (Wilhelm et al., 2007). This study documents the gradual rise in both hormones prior to awakening and a sharp response to awakening for both hormones. However, the pre-awakening rise in ACTH was steeper than the respective rise in cortisol (reported in Hellhammer et al., 2009) suggesting that there may be a pre-awakening dissociation between the secretion of these two hormones, as previously described in rodent studies.

An additional study in humans, which sheds light on the immediate pre-awakening period, comes from the earlier work of Born et al. (1999). In their paper these authors demonstrate an anticipatory rise in ACTH to forewarned early awakening (short sleep as opposed to surprise early awakening). However, this anticipatory rise in ACTH was not accompanied by pre-awakening stimulation of cortisol secretion, which is consistent with the findings of Wilhelm and colleagues and evidence from the animal studies, reported above. Due to space limitations at time of publication it was not possible to publish the data supporting this statement; however, the authors have kindly agreed to allow publication here (see Fig. 1). This study demonstrates a pre-awakening dissociation between ACTH and cortisol in healthy people (see Born et al., 1999 for full details of the study design). In other words, immediately pre anticipated awakening the adrenal glands appear relatively desensitised to the stimulatory effects of rising levels of the secretagogue ACTH. It is speculated here that pre-awakening decreases in sensitivity to ACTH provides steroidogenic capacity with which to initiate a marked increase in cortisol secretion in the immediate post-awakening period.

As a similar reduction in adrenal sensitivity to ACTH has been documented prior to the onset of the active phase in rodents (see Bornstein et al., 2008) it is tempting to speculate that they may share a common extra-pituitary SCN-mediated mechanism. Certainly SCN related neural pathways described in rodents have been shown to be similarly organised in humans (Buijs et al., 2003) so it seems credible that in healthy people a SCN extra-pituitary pathway may provide the basis of the pre-awakening reduced adrenal sensitivity to ACTH.

One of the most consistent findings from the literature is that the hippocampus appears to play a central role in the regulation of the CAR (see Fries et al., 2009). This conclusion has been drawn from studies of clinical populations in which the hippocampus is impaired (e.g. Buchanan et al., 2004; Wolf et al., 2005) and following brain imaging studies which have revealed positive

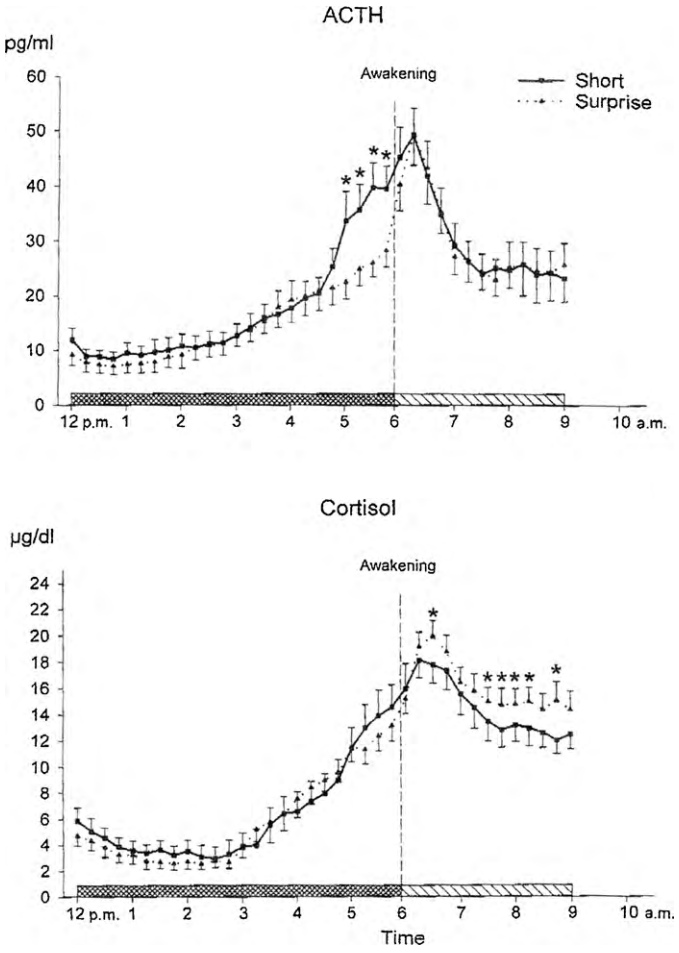

Fig. 1. Mean ± SE plasma concentrations of ACTH and cortisol for subjects under 'short sleep', i.e. planned awakening at the actual awakening time and for 'surprise' awakening, when the participants were expected to be woken later than the actual awakening time. The broken vertical line indicates waking. Subjects stayed in bed for 3 h after waking. Asterisks indicate statistically significant pre-awakening differences between ACTH levels for 'short sleep' and 'surprise' conditions. This anticipatory rise in ACTH was not accompanied by an associated rise in plasma cortisol (with permission of the authors; for full details see Born et al., 1999).

associations between hippocampal volume and the CAR (Bruehl et al., 2009; Pruessner et al., 2007). This body of evidence indicates that functional integrity of the hippocampus is associated with the CAR and suggests a causal linkage. Involvement of the hippocampus in the regulation of the CAR is further suggested by the existence of anatomical and functional pathways linking the hippocampus to the SCN (Krout et al., 2002; Pace-Schott and Hobson, 2002; Stranahan et al., 2008).

It is, however, paradoxical that the CAR, a dynamic activation of adrenocortical activity, is dependent upon the hippocampus, a region of the brain better known for its inhibitory effect on HPA axis activity (Herman and Cullinan, 1997; Herman et al., 2005). This ambiguity has yet to be adequately explained. It is conceivable that the role of the hippocampus in the regulation of the CAR occurs *prior to awakening*. This possibility is consistent with the fact that REM sleep (typically dominant in the later stages of sleep and immediately pre-awakening) is associated with marked hippocampal activation which provides inhibitory tone on cortisol secretion, whereas awakening is associated with switching off of hippocampal activation (Balkin et al., 2002; Braun et al., 1997). In other words, pre-awakening activation of the hippocampus could be implicated in regulation of pre-awakening cortisol secretion. It is possible, and speculated here, that this regulation may be related to the SCN-mediated extra-pituitary fine tuning of adrenal sensitivity to ACTH in the pre-awakening period, as described above. Although speculative there is sufficient circumstantial evidence to merit further investigation of these relationships in their role in the determination of the CAR.

Of course SCN input is not the only regulator of pre-awakening cortisol secretion. Cortisol levels are subject to a range of inputs, including negative feedback inhibition of the HPA axis via glucocorticoid receptors which operate at the level of the hypothalamus and pituitary as well as the hippocampus (De Kloet, 2004). Low doses of dexamethasone administration prior to sleep dramatically reduce cortisol concentrations during sleep and in the first waking sample (and virtually eliminates the CAR) (Galli et al., 2009; Pruessner et al., 1999; Vreeburg et al., 2009; Wust et al., 2009). This negative feedback is modulated by an interaction between different types of glucocorticoid receptors (GR and MR), gender and genotype and ultimately informs the availability of pituitary-derived ACTH (van Leeuwen et al., in press). Thus pre-awakening cortisol secretion is affected by a combination of inputs informing the activity of the HPA axis as well as the sensitivity of the adrenal to ACTH.

4. Post-awakening influences on cortisol secretion: the CAR

In one of the few human studies that measured patterns of neuroendocrine function pre- and post-awakening, ACTH and cortisol both exhibited a response to awakening (Wilhelm et al., 2007). Furthermore the same study demonstrated that the maximum concentration of cortisol shortly after waking was positively related to levels of ACTH, confirming the importance ACTH signalling (and hence the HPA axis) for the CAR. However, there is evidence for additional extra-pituitary input to the CAR, again mediated through the SCN.

Awakening after nighttime sleep is often coincidental with dawn and the increased availability of light. In mammals, light activates retinal ganglion cells to communicate with the SCN in order to entrain the circadian clock to environmental light-dark cycles (Ishida et al., 2005; Moore, 1997). The SCN regulates the circadian pattern of cortisol secretion, and is sensitive to time of day as light exposure has been shown to increase cortisol secretion in the morning but not in the evening (Leproult et al., 2001). A role for the SCN in the regulation of the CAR in healthy humans is indicated by its sensitivity to light: morning awakening in total darkness reduces the dynamic of the CAR relative to morning awakening in light (Scheer and Buijs, 1999). Furthermore, consistent with these findings, it has been shown that waking up using a dawn simulator (a device which gradually increases light levels before awakening) is associated with an increased CAR (Thorn et al., 2004). In neither of these studies was the level of the first waking sample affected by the light manipulation. In rodents light-induced effects on glucocorticoid secretion were absent following lesion of the SCN or in SCN-intact mice with denervation of adrenal sympathetic nerves (Buijs et al., 2003). Thus the SCN-mediated extra-pituitary regulation of cortisol secretion is indicated.

Dissociation between ACTH and cortisol secretion in the post-awakening period in humans has been documented; one early laboratory study (Fehm et al., 1984) highlighted this finding: . . . '*in the case of the physiological morning peak there was no adequate rise in mean ACTH levels*'. Evidence also comes from the observation that the adrenal androgen dehydroepiandrosterone (DHEA) does not mount a dynamic 'response to awakening', despite ACTH also being its secretagogue (Hucklebridge et al., 2005). This could be accounted for by functional zonation of the adrenal cortex. Cortisol is secreted from the zona fasciculate, a region abundant in sympathetic innervations. DHEA is secreted from the zona

reticularis, for which evidence of such sympathetic innervations is lacking (Charlton et al., 1992). These finding are consistent with what has been found in rodents (Buijs et al., 1997, 2003; Ulrich-Lai et al., 2006) and demonstrates light-sensitive enhanced adrenal sensitivity to ACTH (specifically in the zona fasciculate) in the immediate post-awakening period, which contrasts with the reduced adrenal sensitivity to ACTH in the pre-awakening period. Thus, it seems that in healthy individuals the dynamic of the morning awakening cortisol elevation is modulated by a light-sensitive extra-pituitary pathway to the adrenal.

It is noteworthy that the proposed SCN related influences described above are opposite in nature: pre-awakening *reduced* adrenal sensitivity to ACTH and post-awakening *increased* adrenal sensitivity to ACTH. Certainly this is plausible as it is known that the SCN has the potential to exert oscillating inhibitory and excitatory influences on cortisol secretion (Buijs et al., 2003; Ulrich-Lai et al., 2006). Thus it is proposed here that the CAR arises as a consequence of interacting regulatory influences from the HPA axis and extra-pituitary mechanisms, activation of which is initiated by the process of awakening and associated switching in regional brain activity. It has been suggested by others that extra-pituitary mechanisms work in harmony with the HPA axis to 'fine tune' hormone activity appropriate for the time of day (Buijs et al., 2003). To summarise, evidence suggests that in healthy people the SCN is involved in regulation of the CAR in at least three different ways (see Fig. 2): (1) pre-awakening input to the paraventricular nuclei of the HPA axis which synchronises neuroendocrine function (and hence ACTH signalling) with the time of day; (2) extra-pituitary neural pathways to reduce adrenocortical sensitivity to ACTH in the pre-awakening period and hence restrain levels of the first waking cortisol sample; (3) via light-sensitive extra-pituitary neural pathways to increase post-awakening adrenal sensitivity to ACTH and consequently enhance the dynamic of the CAR.

Of course the legitimacy these proposed interconnected physiological pathways needs to be directly tested in carefully controlled laboratory conditions. Furthermore it is not suggested that these connections are the sole driver determining the CAR, rather that they may be amongst a number of influences that affect this discreet aspect of the circadian cortisol cycle including glucocorticoid receptor sensitivity, gender and genotype (see Wust et al., 2009).

5. Relationship between patterns of awakening and post-awakening cortisol secretion

As most studies of the CAR are undertaken in the domestic setting there is limited direct evidence of the relationship between patterns of pre- and post-awakening cortisol secretion. A notable exception is the Wilhelm study of 2007, which has already been described in some detail. This study reported that high nocturnal levels of cortisol secretion were significantly associated with an attenuated incremental post-awakening increase. Furthermore, and in line with these effects, a significant negative relation was also detected between the post-awakening serum cortisol increase and the level immediately upon awakening (Wilhelm et al., 2007). Thus it seems that the first waking sample (S1), if taken at the correct time, i.e. immediately post-awakening represents a measure of the end of the pre-awakening period and can be analysed in relation to the dynamic of the CAR, which is the measure of the post-awakening cortisol secretion.

It is a frequent (although not ubiquitous) observation that high levels of salivary cortisol in S1 are associated with an attenuated CAR (Adam et al., 2006; Dahlgren et al., 2009; Stalder et al., 2009; Vreeburg et al., 2009; Wilhelm et al., 2007). Although not directly referred to in the papers it is also noteworthy that visual inspection of the data from the two main studies of the CAR in clinical populations with impaired hippocampal function suggests that

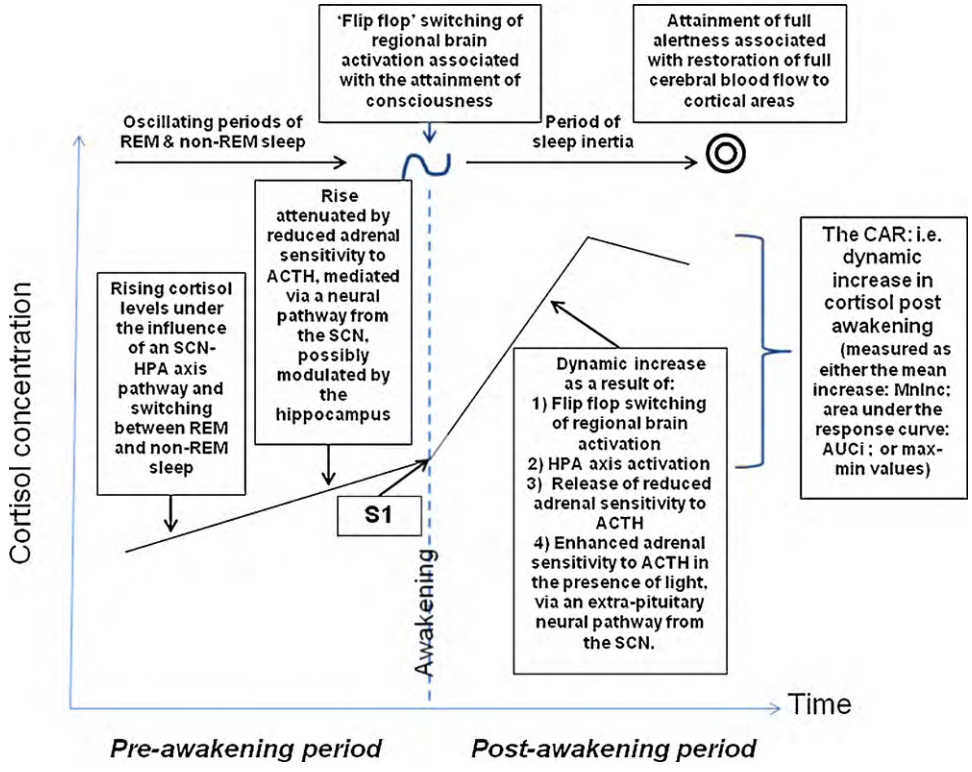

Fig. 2. Proposed influences on pre- and post-awakening cortisol secretion. The processes highlighted are also modulated by glucocorticoid receptor inhibitory feedback, gender and genotype (see text for a full discussion). CAR = cortisol awakening response; S1 = the level of cortisol in the first sample immediately after awakening; REM = rapid eye movement.

although there is a severely attenuated dynamic of the CAR the S1 is elevated (Buchanan et al., 2004; Wolf et al., 2005). Although results of associations between psychosocial and health variables and the CAR are notoriously inconsistent, it is reported that increasing age (Kudielka and Kirschbaum, 2003) as well as a range of conditions, e.g. cardiovascular disease; autoimmune conditions; slow wound healing; clinical depression; mild cognitive impairment; Alzheimer's disease and attachment anxiety, are associated with a high first waking sample and an attenuated dynamic increase following awakening (e.g. Arsenault-Lapierre et al., in press; Buske-Kirschbaum et al., 2007; Ebrecht et al., 2004; Huber et al., 2006; Kudielka and Kirschbaum, 2003; Quirin et al., 2008). A notable and consistent exception to this pattern is post-traumatic stress disorder, which is discussed below. Although a high S1 could possibly be a result of non-adherence to protocol or indeed dysregulation of an as yet unknown regulatory mechanism these findings are consistent with a reduction in SCN-mediated 'fine-tuning' of adrenal sensitivity to ACTH, i.e. less pre-awakening reduced adrenal sensitivity to ACTH (high S1). The sometimes related phenomenon of an attenuated CAR could possibly be associated with less cortisol secretory 'impetus' if the switch from *reduced* to *increased* adrenal sensitivity, thought to occur upon awakening as a result of flip-flop switching of regional brain activation, is inefficient. If this is proven to be the case it would indicate roles for the SCN, and possibly the hippocampus, as well as the HPA axis in a range of conditions associated with this pattern of CAR.

The importance of the sensitivity of negative feedback inhibition of the HPA axis for the CAR is illustrated by contrasting the CAR profiles observed in populations suffering from post-traumatic stress disorder (PTSD) and clinical depression; conditions associated with hyper- and hypo-sensitivity of feedback inhibition respectively. Patients with PTSD characteristically show an attenuated CAR with a low first waking sample, whilst results for associations with clinical depression although less consistent clearly do not show this profile (Fries et al., 2009). What is clear from these studies is that in PTSD there is no 'impetus' for the CAR, as hypothesised above. Enhanced negative feedback, associated with PTSD, results in reduced ACTH signalling (Yehuda et al., 1996), which may be the reason for the characteristic low and flat CAR typical of this population (rather than a SCN related mechanism). It is clear that negative feedback of the HPA axis, which consistently and severely reduces ACTH activity, tends to wipe out the CAR (Jacobson and Sapolsky, 1991) indicating its permissive role for activation of this waking response. This observation is consistent with the suggestion that HPA axis derived ACTH signalling is a key aspect of the CAR, affecting the first waking sample and the potential for a dynamic increase in cortisol secretion following awakening.

The tendency for an inverse relationship between time of awakening and the CAR (e.g. Dahlgren et al., 2009; Edwards et al., 2001; Kudielka and Kirschbaum, 2003; Vreeburg et al., 2009) mirrors the finding of an inverse relationship between levels of S1 and the CAR reported above. Indeed a recent case study exploring within-participant state variability of the CAR over 50 sampling days demonstrated that the waking time-CAR association was driven by the first waking sample: for this individual later waking time was associated with a higher waking cortisol sample which in turn was associated with a reduced dynamic of the CAR (Stalder et al., 2009). This sensitivity to the stage of the circadian rhythm, rather than sleep characteristics (see Fries et al., 2009), is indicative of SCN–HPA axis input to S1.

6. Sleep inertia and the CAR

The period between regaining consciousness (i.e. awakening) but before attainment of full alertness is described as 'sleep inertia': a transitory period of impaired arousal and behavioral performance lasting between 15 and 60 min (Ferrara et al., 2006; Ikeda and Hayashi, 2008). Sleep inertia is associated with measurable physiological processes. For example, following morning awakening it takes about 30 min to reach global cerebral blood flow velocity values corresponding to the waking state of the previous evening (Hajak et al., 1994). Furthermore, dissociation between regional brain activation associated with consciousness and alertness has been mapped: reactivation in the thalamus, caudate, brainstem and anterior cingulate cortex occurs almost immediately upon awakening whereas the time course of reactivation of the prefrontal cortex corresponds to recuperation from sleep inertia (Balkin et al., 2002). The lag in reactivation of cortical brain areas after awakening has also been demonstrated using EEG spectral power analysis in which post-awakening EEG hypoarousal (a decrease of delta and theta accompanied by an increase in the alpha frequency) was shown to last at least 10 min (Ferrara et al., 2006).

Thus, it seems that the initiation of the CAR is temporally associated with the attainment of consciousness and that the dynamic of the CAR closely parallels that of reactivation of the prefrontal cortex and attainment of full alertness. Whilst this might be viewed as coincidental, there is some evidence indicating that the CAR may indeed be associated with the regaining of arousal following awakening. Indirect support for this is provided by the relatively consistent finding that acute bursts of cortisol have a stimulatory influence on psychological arousal and lead to a reduction of fatigue. This effect was confirmed using self-report measures (Tops et al., 2006), arousal ratings in response to non-arousing stimuli (Abercrombie et al., 2005) as well as electroencephalographic (EEG) indicators of central alertness (Chapotot et al., 1998).

Only a few studies have examined the CAR in relation to state arousal or levels of physiological activation. However, the results available to date have been supportive of a role of the CAR in the regaining of arousal, suggesting a positive association between state arousal at 45 min post-awakening and post-awakening cortisol levels (Thorn et al., 2004) as well as the dynamic of the CAR (Thorn et al., 2009). This finding is also in general agreement with results of Adam et al. (2006) showing an association between a larger mean CAR and lower average fatigue levels over a 3-day period. State arousal/anticipations of a busy day ahead at 45 min post-awakening have also been shown to relate positively with the CAR (Stalder et al., 2009). In addition high levels of sleepiness were associated with lower levels of cortisol 15 min after awakening in healthy office workers (Dahlgren et al., 2009). In summary the evidence of causal linkages between the dynamic of the CAR and recovery from sleep inertia are currently speculative, but deserving of further investigation.

A clear understanding about the direction of causality in such relationships is not yet possible, but there is evidence that the CAR is affected by prior day experiences (Adam et al., 2006; Dahlgren et al., 2009; Doane and Adam, in press; Stalder et al., 2009). It has been suggested that in healthy individuals an increase in the CAR in response to adverse prior day social experiences may provide an extra 'energetic boost' to help the individual meet the demands of the next day (Adam et al., 2006; Doane and Adam, in press). If this is true then it would seem that anticipatory processes can drive the CAR and that the resultant increased arousal (reported in some studies—see above) is secondary to that process. In this interpretation the CAR, known to have substantial intra-individual state variation within healthy people (Hellhammer et al., 2007; Stalder et al., 2009) may be seen to be an adaptive response to restore maximal functioning from day-to-day. Certainly this hypothesis deserves further investigation.

Just as there appears to be multiple regulatory influences on the CAR it may well be that its function is multipurpose; cortisol is one

of the most potent hormones of human physiology, with a wide range of effects on target tissues that are adaptive in normal human functioning. It is not surprising therefore that in addition to the proposed role of the CAR in relation to recovery from sleep inertia and the provision of an 'energetic boost' it has been also been implicated in cognitive function and regulation of the immune system (see Fries et al., 2009). However, it is beyond the scope of this paper to explore these functions more fully. What is clear is that establishment of the role or roles of the CAR should be a high priority in this area of research. Without this basic knowledge it is difficult to interpret the many and varied reports of 'dysfunction' in this aspect of the cortisol circadian cycle.

7. Measurement of the CAR

It is hoped that the evidence presented here for a potential combination of regulatory mechanisms will encourage publication of the key determinants of the CAR: the first waking sample (an indicator of pre-awakening cortisol secretion) as well as the dynamic of the increase. Ideally, each of these component parts could be subject to statistical investigation. Without this information the task of unravelling associations with the host of physiological and psychosocial variables of interest will be difficult, if not impossible: aspects of the regulatory process may be differentially associated. In particular, use of the composite measure AUC_G (i.e. area under the post-awakening cortisol curve with reference to ground/zero) or mean levels of cortisol secretion in the post-awakening period, whilst providing some data of interest will, if presented without information about its component parts (i.e. sample 1 and the dynamic of the increase) be somewhat limited.

Clearly, consistency in the way the CAR is measured and presented is important for the development of this area of research. The strategy suggested here, and which has already been used by a number of researchers (e.g. Adam et al., 2006; Dockray et al., 2008; Steptoe et al., 2005; Whitehead et al., 2007), is to substitute measures of overall hormone levels with a measure of the first waking sample, whilst maintaining a measure of the awakening response (e.g. AUC_I or MnInc). This approach has a number of advantages. Firstly, it provides two clearly distinguishable measures, making it easy to interpret whether a particular association or group difference is with the end state of the pre-awakening cortisol secretion (i.e. S1) or the post-awakening response (i.e. dynamic increase). Secondly, using this method of quantification it becomes unequivocal what is meant by 'cortisol awakening response (CAR)', as this term can then be exclusively reserved for measures of the post-awakening dynamic, e.g. AUC_I, MnInc.

8. Conclusion

It is hoped that this paper has highlighted that the CAR is subject to a range of influences, both pre- and post-awakening. It seems that extra-pituitary input from the SCN serves to 'fine tune' HPA axis functioning in the pre- and post-awakening period. It may not be surprising that the CAR literature is so inconsistent with regard to associations with trait psychosocial and health measures. Putting aside issues related to participant non-adherence to protocol (which undoubtedly severely affects the quality of the evidence base) and marked state-related variability in the CAR (which may cloud associations with more stable trait characteristics) it is likely that different trait factors may be associated with different aspects of the regulatory puzzle, making it very difficult to tease apart. Notwithstanding this complexity it is apparent that the CAR remains a promising pre-clinical biomarker of psychosocial status and health, potentially providing insight into a range of clinically relevant physiological systems. However, the current limited level of understanding hampers interpretation of results and therefore the usefulness of this measure. Without a clearer basic understanding of the regulation and functional significance of the CAR in healthy people, the impact of dysregulation in terms of physical pathology cannot be deduced.

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