Published in final edited form as:

Psychoneuroendocrinology. 2008 August ; 33(7): 1023–1026. doi:10.1016/j.psyneuen.2008.04.011.

Naturalistic Stress and Cortisol Response to Awakening: Adaptation to Seafaring

Jonathan Liberzon, B.Sc.¹, James L. Abelson, M.D., Ph.D., Anthony King, Ph.D., and Israel Liberzon, M.D.

University of Michigan School of Natural Resources, Ann Arbor, MI. University of Michigan Medical School, Department of Psychiatry, Ann Arbor, MI. Ann Arbor VA Medical Center, Ann Arbor, MI

Abstract

Study of the hypothalamic-pituitary adrenal (HPA) axis has been critical to advancing our understanding of human adaptation to stress. The cortisol response to awakening (CRA) is a potentially useful measure for understanding group and individual differences in HPA axis regulation. In this study, the CRA was examined in the context of a naturalistic stressor – a six-week voyage of work and study aboard an oceangoing ship, including both experienced and novice sailors. Thirty-one subjects provided weekday and weekend baseline CRA data onshore prior to boarding, followed by three CRAs at sea and one shore leave CRA. Subjective measures of sleep, stress and control were also collected. Results suggest that novice sailors' cortisol response to awakening was elevated at sea relative to both a shoreside weekend and a shore leave during the voyage, but the most striking elevation was found during a workday onshore. Inexperienced students' profiles changed differently over the course of the voyage from those of professional crew. CRAs were not affected by sleep variables and were not predicted by subjective ratings. These data support the value of the cortisol response to awakening as a neuroendocrine marker of HPA regulatory responses to a naturalistic stressor, influenced by changes in work and living environment, and perhaps prior experience with the stressor.

Keywords

cortisol response to awakening; stress; naturalistic stressors; HPA axis

Introduction

The cortisol response to awakening (CRA), a measure of hypothalamic-pituitary adrenal (HPA) axis activity, is defined as the change in free cortisol from wakeup to sometime afterwards (usually 30-90 minutes). The CRA is part of the cortisol circadian cycle and has been characterized in healthy adults as a 50-160% rise in salivary free cortisol in the first 30 minutes after awakening (Clow et al., 2004). After peaking, cortisol concentrations return to basal levels within 1-2 hours. The CRA is more dynamic than 24 hr basal cortisol secretion and may primarily reflect adrenal cortical sensitivity, which may in turn be shaped by prolonged stressor exposure (Schmidt-Reinwald et al., 1999). The measure shows high intra-subject stability and

¹Corresponding Author: Jonathan Liberzon 1350 Orkney Ann Arbor MI 48103 (734) 546-3556 jonlib@umich.edu

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

is independent of adult age, weight, smoking status and alcohol consumption (Pruessner et al., 1997).

A number of biological, psychological and behavioral factors affect the CRA. It appears altered in some psychopathological states associated with HPA dysregulation, such as post-traumatic stress disorder (Wessa et al., 2006) and major depressive disorder (Huber et al., 2004). In healthy adults, it is affected by workload and perceived work overload, socioeconomic status and gender. For example, higher CRA was found in women with high job strain and low decision latitude at work (Alderling et al. 2006). Self-reported chronic stressors such as worry and social stress predict higher CRAs in healthy subjects (Wüst et al., 2000). Also, CRA was found to be smaller on weekend days than on working days (Kunz-Ebrecht et al., 2004; Schlotz et al., 2004). It has thus been proposed as a useful marker of subtle changes in HPA axis reactivity in response to stressor exposure or psychiatric symptoms (Schmidt-Reinwald et al., 1999).

This study used a six-week voyage onboard an educational sailing vessel as a stress model, measuring the CRA and subjective distress in experienced and novice sailors to verify the sensitivity of this measure to a naturalistic stressor, and to examine the influence of prior experience with the stressor, sleep duration and perceived control. The sea voyage was selected because it exposed both novice students and experienced staff to factors that have been shown to stimulate the HPA axis, such as risk to life, high job strain and low decision latitude (Alderling et al. 2006), examination and performance evaluations (Martinek et al., 2003; Ng et al., 2003; Lindahl et al., 2005) and social stress, while controlling for time of awakening, supervisor expectations, physical load, work type and frequency of social contacts.

Methods

Data were collected by an investigator from 31 subjects (18 females) enrolled in or instructing class 195A of the Sea Education Association semester program, which consisted of onshore study followed by 6 weeks of work and study aboard the SSV Corwith Cramer. Subjects included 23 students, four professional science staff and four professional ship's crewmembers, ranging in age from 18 to 38 years. All subjects signed informed consent after full description of the study, which was approved by the University of Michigan Institutional Review Board.

Subjects provided saliva samples on six separate mornings, at zero, 30 and 45 minutes after awakening. They were instructed not to eat, drink, smoke, brush teeth or rinse their mouths until after the 45 minute sample. Two mornings were sampled prior to boarding the ship. One shore sample was taken on a working day (OSWD) during a normal week at the campus of the Sea Education Association. The second shore sample was taken the morning of a break day prior to departure. This sample (OSWE) was intended to simulate an average weekend morning, since subjects did not have to work or prepare for exams. Subjects slept off-campus the night before the OSWE sample, and a few slept at home. Only two scientific staff members provided OSWD and OSWE samples, and no ship's crewmembers were sampled onshore.

Prior to boarding, subjects were divided into three groups or "watches", assigned to three rotating, offset shifts (morning, mid-day and night) to provide round-the-clock coverage of sailing duties. Each "watch" awoke at a different time, and each watch rotated through all three shifts in each 72 hour period. In order to sample all subjects at the same waking time, the "watches" were sampled on consecutive days, as they rotated through the morning shift (5:30 a.m. wakeup). Sampling began the morning of day 4, because a high rate of seasickness posed a substantial confound during the first few days at sea. The three groups provided their initial samples (Sea-1) on days 4, 5 and 6 of the voyage, respectively. A second set of samples was similarly obtained on days 10, 11 and 12 (Sea-2); a third set was obtained on days 19, 20 and

21 (Sea-3). Samples were also obtained from all groups upon awakening onboard ship for the second of two shore leaves on day 31 (M-2). The second shore leave was chosen to reduce the potential impact of shore leave novelty on the CRA. OSWE samples were taken 2-8 days after OSWD samples, most within 3-4 days. Sea-1 samples were taken an average of 9 days after OSWE samples.

Subjects completed one seven-point Lykert scale of perceived stress and one of perceived control concomitant with their first saliva sample on each sampling morning. Stress was reported based on the prompt "How stressed do I feel?" and control was reported based on the prompt "How in-control do I feel?" Subjects also recorded sleep duration and time of awakening before each sample. Saliva samples were frozen immediately and kept frozen at $-12.5^{\circ} \pm 5^{\circ}\text{C}$ until assay. Cortisol was assayed using the Coat-A-Count assay from Diagnostic Products Corporation (Los Angeles, Calif). The primary cortisol outcome measure utilized was area under the curve for awakening response (CRA AUC, calculated by trapezoidal approximation after subtracting the waking value from the +30 and +45 minute values). We also examined total area under the curve (AUC_{tot}, using all 3 samples without subtraction) and maximum awakening response (larger of the +30 and +45 minute values minus the waking value).

Results

A clear increase in salivary cortisol was detected after awakening in the majority of samples. CRAs were characterized by an average salivary cortisol increase of 31%. Analysis of CRA AUC over the course of the study revealed a main effect of time, indicating that subjects' CRAs changed as a function of time or changes in working/living conditions (repeated measures ANOVA, $p=.0009$, Figure 1). The CRA AUC was smaller on the weekend shore morning compared to all other sample points except shore leave (Post-hoc Fisher tests: workday onshore, $p<.0001$; Sea-1, $p=.012$; Sea-2, $p=.056$; Sea-3, $p=.0005$; shore leave, $p=.140$), indicating that the weekend day elicited smaller CRAs than working days onshore or at sea.

Interestingly, the CRA AUC was greater on the workday shore sample compared to Sea-2 ($p=0.023$) and shore leave (M-2) ($p=0.007$), indicating that CRAs were smaller on some sea mornings (of both working and break days) than on the morning of a workday onshore. Total cortisol secretion at awakening time (AUC_{tot}) was also lower at sea than during the onshore workday (significant for Sea-1, $p=0.049$ and Sea-2, $p=0.045$). The CRA AUC was greater for the Sea-3 sample compared to M-2 ($p=0.037$), indicating that CRAs decreased from the third at-sea working day sampled to the shore leave day, on which subjects were not scheduled to work. The basic pattern of results was almost identical when using maximum awakening response instead of AUC response. The pattern was similar but with less overall significance using AUC_{tot}.

Professional crew did not provide on-shore samples, so analysis of group differences included only the four shipboard samples (Sea-1, Sea-2, Sea-3 and M-2). Ship's crew ($n=4$) and students ($n=20$) were compared on CRA AUC in a repeated measures ANOVA, revealing a main effect of time ($p=.0008$) and a time-by-group interaction ($p=.003$), due to rising responses over time at sea in crew, with relatively stable responses over time in students (see Figure 2). Analyzed separately, the CRA AUC for the ship's crew was significantly greater at Sea-3 than at Sea-1 ($p=.009$) or Sea-2 ($p=.017$), whereas the students analyzed separately showed no differences across time. No significant differences were found between students and crew in subjective stress or sense of control.

No differences were found between men and women for the variables investigated. Subjective ratings of stress and control showed a strong and significant negative correlation, such that less

perceived control was associated with higher perceived stress. Students reported lower perceived stress during onshore weekend morning than during onshore workday morning or any at-sea mornings ($p < .001$), but no significant difference was found in perceived stress between onshore workday and at-sea workday mornings. No significant correlations were found between CRAs and subjective ratings of stress or control. Time of awakening and total sleep time did not correlate with cortisol variables, except for waking cortisol (0 min) values, which correlated with time of awakening in the M-2 sample. Time of awakening and stress did not together predict cortisol response to awakening on any sampling day (using multiple regressions with correction for multiple comparisons).

Discussion

This study successfully sampled the CRA during a standard working and weekend day, and while working at sea. CRAs were not correlated with sex, sleep parameters or subjective stress and control.

CRA was greater during onshore workdays compared with weekend days, replicating the findings of Kunz-Ebrecht et al. (2004) and Schlotz et al. (2004) within the novel context of a student cohort removed from the routines of normal home life. These findings support the hypothesis that the CRA provides a useful marker of naturalistic challenge stress and is elevated in anticipation of a more challenging day. Further research is needed to determine exactly what about a working day boosts the CRA.

Awakening time varied between OSWD and OSWE samples, but we found no correlation between awakening time or sleep duration and CRA magnitude, suggesting that differences in sleep were likely not the cause of the workday/weekend CRA difference. Williams et al. (2004) suggest that interactions between early waking time and post-wakeup stress may affect the CRA, but we were unable to predict CRA on any sampling day in this study using awakening time, stress, or their combination (in multiple regression analyses). It does not appear that either sleep variables or our subjective stress measure, or their interaction, can explain the cortisol response to awakening in this data set.

Contrary to our hypothesis, workdays at sea did not elicit greater CRAs than workdays onshore among students. Our hypothesis had been based on the greater novelty, more intense performance evaluation (crew evaluating ship duty performance, staff and peers evaluating oral presentations), reduced decision latitude and control (rigid schedules, need to “follow orders”), longer work hours, more physical labor, and heightened level of danger present on board ship relative to classroom activities. However, CRAs of students were smaller during at-sea workdays than during onshore workdays. There is evidence that CRA is lower in individuals with higher positive affect and optimism (Lai et al., 2005). It also is lower the morning before a test challenge in students who react to test-induced stress with confidence (endorsing the belief that “I can solve this task”) (Lindhal et al., 2005). It is possible that students who elect to spend a semester at sea perceive seafaring challenges more positively than the stressors associated with on-shore classroom activity. A challenge-seeking attitude, self confidence, and positive excitement for seafaring may have attenuated the CRA, serving as antidotes to novelty, evaluative stress and a sense of lack of control that are known to activate the HPA axis (Dickerson and Kemeny, 2004). Perhaps these action-oriented, adventure-seeking students reacted more strongly to evaluative stress in a classroom onshore than at sea. Social bonding onboard or confounding environmental factors may also have played a role in altering the CRA at sea.

We predicted that the crew, who had more experience and control, and were subjected to fewer evaluative tests, would demonstrate stable CRAs consistently smaller than those of students.

Surprisingly, CRAs increased during the voyage in ship's crew, while student CRAs remained constant. Interpretation here must be cautious, due to the small sample, but it may be that the crew's sense of control diminished as they turned over operation of the ship to the students, though they remained responsible for its safe passage. The crew's subjective ratings of control did not decline over time, but a response bias is likely as crew members were required to project an "in-control" attitude at all times.

The shore leave sample, intended to approximate a "weekend at sea", was characterized by smaller student CRAs than both at-sea and onshore workdays. Despite our prediction that OSWE and M-2 CRAs would be analogous, students showed greater CRAs during the shore leave compared to onshore weekends. Though there were minimal duties and no evaluations on this day, students spent part of their shore leave on an unfamiliar foreign island. Anticipation of this novel environment may have kept CRAs above onshore weekend levels.

These data indicate that the CRA may be a useful neuroendocrine marker of day-to-day fluctuations in stress levels, influenced by changes in work and living environment. We have shown it to be sensitive to contextual factors and to group differences in previous experience with a naturalistic stressor. Further work is needed to identify more precisely those aspects of context and experience that shape the CRA. This work will help us understand more precisely what is "stressful" to humans and may facilitate our search for ways to reduce stress and its harmful effects on health.

References

- Alderling M, Theorell T, de la Torre B, Lundberg I. The demand control model and circadian saliva cortisol variations in a Swedish population based sample (The PART study). *BMC Public Health* 2006;6:288. [PubMed: 17129377]
- Clow A, Thorn L, Evans P, Hucklebridge F. The Awakening Cortisol Response: Methodological Issues and Significance. *Stress* 2004;7(1):29–37. [PubMed: 15204030]
- Dickerson S, Kemeny M. Acute Stressors and Cortisol Responses: A Theoretical Integration Synthesis of Laboratory Research. *Psychol. Bull* 2004;130(3):355–391. [PubMed: 15122924]
- Huber T, Issa K, Schik G, Wolf T. The cortisol awakening response is blunted in psychotherapy inpatients suffering from depression. *Psychoneuroendocrinology* 2006;31(7):900–904. [PubMed: 16707227]
- Kunz-Ebrecht SR, Kirschbaum C, Marmot M, Steptoe A. Differences in cortisol awakening response on work days and weekends in women and men from the Whitehall II cohort. *Psychoneuroendocrinology* 2004;29:516–528. [PubMed: 14749096]
- Lai JCL, Evans PD, Ng SH, Chong AML, Siu OT, Chan CLW, Ho SMY, Ho RTH, Chan P, Chan CC. Optimism, positive affectivity, and salivary cortisol. *Br. J. Health Psychol* 2005;10:467–484. [PubMed: 16238860]
- Lindahl M, Theorell T, Lindblad F. Test performance and self-esteem in relation to experienced stress in Swedish sixth and ninth graders - saliva cortisol levels and psychological reactions to demands. *Acta Paediatr* 2005;94(4):489–495. [PubMed: 16092465]
- Martinek L, Oberascher-Holzinger K, Weishuhn S, Klimesch W, Kerschbaum HH. Anticipated academic examinations induce distinct cortisol responses in adolescent pupils. *Neuro Endocrinol. Lett* 2003;24(6):449–453. [PubMed: 15073574]
- Ng V, Koh D, Chia SE. Examination stress, salivary cortisol, and academic performance. *Psychol. Rep* 2003;93(2):1133–1134. [PubMed: 14765582]
- Pruessner JC, Wolf OT, Hellhammer DH, Buske-Kirschbaum A, von Auer K, Jobst S, Kaspers K, Kirschbaum C. Free cortisol levels after awakening: A reliable biological marker for the assessment of adrenocortical activity. *Life Sci* 1997;61(26):2539–2549. [PubMed: 9416776]
- Schmidt-Reinwald A, Pruessner JC, Hellhammer DH, Federenko I, Rohleder N, Schürmeyer TH, Kirschbaum C. The cortisol response to awakening in relation to different challenge tests and a 12-hour cortisol rhythm. *Life Sci* 1999;64(18):1653–1660. [PubMed: 10328525]

- Schlotz W, Hellhammer J, Schultz P, Stone AA. Perceived Work Overload and Chronic Worrying Predict Weekend–Weekday Differences in the Cortisol Awakening Response. *Psychosom. Med* 2004;66:207–214. [PubMed: 15039505]
- Wessa M, Rohleder N, Kirschbaum C, Flor H. Altered cortisol awakening response in posttraumatic stress disorder. *Psychoneuroendocrinology* 2006;31:209–215. [PubMed: 16154709]
- Williams E, Magid K, Steptoe A. The impact of time of awakening and concurrent subjective stress on the cortisol response to awakening. *Psychoneuroendocrinology* 2005;30:139–148. [PubMed: 15471612]
- Wüst S, Federenko I, Hellhammer D, Kirschbaum C. Genetic Factors, perceived chronic stress, and the free cortisol response to awakening. *Psychoneuroendocrinology* 2000;25:707–720. [PubMed: 10938450]

1. .

2. .