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# **Assessing the role of anticipation in psychobiological stress responding**

**Olivia Alice Craw**

**PhD**

**2016**



# **Assessing the role of anticipation in psychobiological stress responding**

**Olivia Alice Crow**

**A thesis submitted in partial  
fulfillment of the requirements of  
the University of Northumbria at  
Newcastle for the degree of Doctor  
of Philosophy**

**Faculty of Health and Life Sciences**

**September 2016**



## **Abstract**

The acute stress response is an adaptive and necessary function which, when activated under appropriate conditions, promotes survival. However, studies have demonstrated that chronic over-activation of the systems that regulate the stress response leads to the dysregulation of the hormonal mediators, which can, subsequently, result in deleterious health outcomes.

Whilst the psychobiological response to acute stressors has been explored extensively, literature assessing the anticipatory and recovery windows surrounding stressor exposure is currently in its infancy. It has been observed that the anticipatory period prior to exposure to a stressor can prolong the activation of the stress response; however less is known of the effects of delayed recovery following stress exposure.

The present thesis addressed the question of whether anticipatory patterns differ between a naturalistic and laboratory stressor, by firstly developing an ecological and easily administered socially evaluative stressor paradigm, and using this stressor as a tool for a) assessing the psychobiological response to the novel stressor and b) assessing the anticipatory and recovery period following this stressor, through the collection of psychobiological data over four days (the day prior to exposure, the day of exposure, the day after and on a control day). A similar sampling protocol was applied to assess the anticipation period preceding a naturalistic stressor (skydiving). Individual differences, which may potentially exacerbate or buffer the negative effects of stress, were also explored within the context of these stressors.

In addition to assessing anticipation of forthcoming stressful events, following recent suggestions that forthcoming positive activities may also elicit similar patterns of anticipatory responses, the current thesis also addressed the question of whether these anticipatory responses may represent a reaction to memory recall for an upcoming task, and not exclusively a response experienced prior to a stressful event. This was addressed by assessing the anticipatory period prior to the requirement to remember to complete a simple task in order to receive a reward.

The findings indicate that the developed stressor successfully elicited a stress response, and was anticipated to be a forthcoming demanding situation. State anxiety was greater on the day of planned stressor exposure, as was stress and worry about the event. In the skydiving study, those who knew they would complete a skydive that day secreted greater levels of cortisol across the day compared to those who were unsure whether they would participate in a skydive or not, and those who knew for certain that they would not complete the skydive. The study assessing the psychobiological response to anticipation of a pleasant stimulus, however, revealed no significant effects. The exploration of a range of individual difference factors demonstrates the importance of appraisal of the event. That is, irrespective of the nature of the event, an individual's perception of the event is an important determinant of psychological and biological responding.

On the basis of the empirical findings of this thesis, it is concluded that both the laboratory and naturalistic stressors elicit some form of anticipatory response. This finding is in concordance with previous suggestions that the stress

response can be observed prior to direct stressor exposure. Furthermore, exploration of the role of individual differences in the anticipation of novel events identified a number of characteristics which may serve to buffer or exacerbate the negative effects of prolonged stress on health outcomes.

Finally, the findings from this thesis do not suggest that the anticipatory responses reported for stressors are extended to the anticipation of pleasant events, but that they may exclusively apply to stimuli perceived to be stressful and ultimately, a forthcoming demand. However, when individual differences are taken into account, it appears that the novelty of a forthcoming event in general may be more influential in appraisals of the event, rather than the nature of the task itself.

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*Publications*

**Hare, O. A.,** Wetherell, M. A., & Smith, M. A. (2013). State anxiety and cortisol reactivity to skydiving in novice versus experienced skydivers. *Physiology & behavior, 118*, 40-44.

(published in maiden name)

*Publications and contributions from the present thesis:*

- Excerpts of Chapter 3 are to be published in a forthcoming book, the International Handbook of Psychobiology.
  - A poster presentation of the findings of Study 1 were presented at the Midlands Health Psychology Annual Conference (2014).
  - Study 4 was presented at The Northumbria University Annual Research Conference (2015) and The PsyPAG Annual Conference (2016).
  - The findings of Study 3 were recently reported at The Psychobiology Section of the British Psychological Society Annual Scientific Meeting (2016):
- Craw, O, Smith, M.A, Wetherell, M.A (2016). Weather to Fly: The psychobiological effects of anticipation of a forthcoming skydive. The Psychobiology Section of the British Psychological Society Annual Scientific Meeting, the Lake District.

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## **Declaration**

I declare that the work contained in this thesis has not been submitted for any other award and that it is all my own work. I also confirm that this work fully acknowledges opinions, ideas and contributions from the work of others.

Any ethical clearance for the research presented in this thesis has been approved. Approval has been sought and granted by the Faculty of Health and Life Sciences Ethics Committee.

I declare that the Word Count of this Thesis is 64,499 words

Name: Olivia Alice Crow

Signature:

Date

## **Chapter 1**

A literature review examining the mechanisms underlying the psychobiological response to stressful events

## **Introduction**

Stress can be defined as a threat, real or perceived, to the psychological or physical integrity of an individual (McEwen, 2000). When faced with threat, the body mounts an appropriate stress response; an innate and adaptive preservation mechanism which, during early periods of evolutionary development, allowed humans to withstand challenges to survival (Cannon, 1953).

The stress response involves a complex collection of processes, which require the synchronisation of a variety of physiological systems within an individual, initiated by information acquisition from the external environment, and are subsequently communicated to the body through the nervous system. When an individual experiences stress, the amygdala, which plays a key role in emotion processing, sends a distress signal to the hypothalamus via the nervous system. The nervous system is a complex network of nerves and cells that pass messages to and from the brain and spinal cord to other relevant areas of the body. This system is the coordinator of physiological processes, collecting information not only from the environment, but also from within the body itself, in order to determine appropriate subsequent actions.

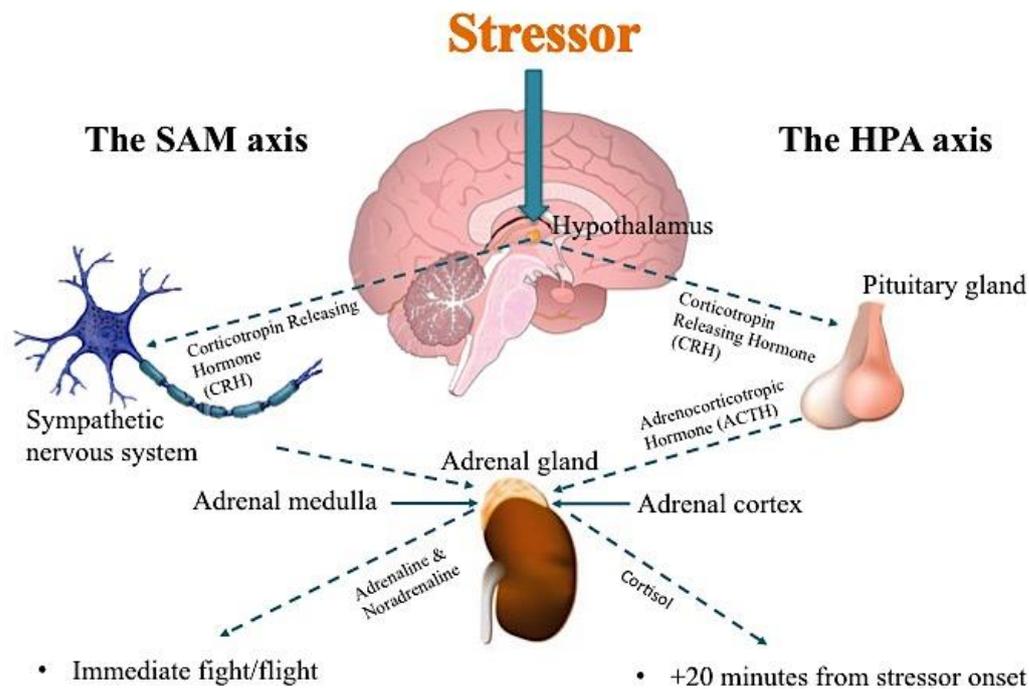
### *The Central Nervous System*

Like many decision-making systems, the nervous system manages input, data processing, and output, and is divided into two parts: The Central Nervous System (CNS) and the Peripheral Nervous System (PNS). The CNS consists of the brain and spinal cord, and is the central command system where executive decisions are made based on information communicated to the CNS via signals.

The PNS, on the other hand, comprises the somatic and automatic nervous system (ANS). The CNS receives transmissions from the sensory nervous system (SNS) and, in turn, provides instructions to the somatic nervous system (muscles). However, within the PNS, the ANS is responsible for involuntary survival functions such as breathing, activity of the heart muscle and general operation of the organs. Under stressful circumstances, the ANS maintains these essential functions without conscious intervention of the individual, allowing them to focus exclusively on coping with the stressor.

When a stress signal is received from the hypothalamus, this activates the ANS, which is divided into two specific branches: the sympathetic and parasympathetic nervous systems. The sympathetic branch is responsible for activating the Sympathetic Adrenal Medullary (SAM) axis (see Figure 1.1), which acts instantaneously to facilitate mobilisation of the body when immediate action is required (i.e. for the flight or fight response); accelerating heart rate, raising blood pressure, increasing oxygen uptake in the lungs (Smeets, 2010), and generally relieving non-essential bodily functions. These actions are supported by adrenaline, which is produced in the adrenal medulla by direct nerve stimulation for the hypothalamus (Bitsika, Sharpley, Sweeney & McFarlane, 2014). The parasympathetic system has the opposite effect, seeking to calm the body, maintaining normal body functions; slowing heart rate, absorbing nutrients, and conserving physical resources. These opposing systems work together to maintain homeostasis in order to ensure the stability of vital functions such as heartbeat. For example, with regard to acute cardiovascular responses, when activity is required, these systems work together simultaneously, with sympathetic activity

increasing heart rate to facilitate the function of fight or flight. Once activity is no longer necessary, the parasympathetic system regains control to lower the heart rate and to maintain or reactivate acute non-adaptive bodily functions, previously shut down by the sympathetic system (such as digestion).



**Figure 1.1** Stress mechanisms: The Sympathetic Adrenal Medullary (SAM) and Hypothalamic Pituitary Adrenal (HPA) axes (created by author).

As with the SAM axis, the Hypothalamic Pituitary Adrenal (HPA) axis (see Figure 1.1) is initiated by the hypothalamus, based upon external environment and internal monitoring inputs (Bitsika et al., 2014). The HPA axis is a slower-responding system than the SAM, as it is activated via hormone release rather than the nervous system. When faced with a stressor, the paraventricular nucleus of the hypothalamus releases corticotropin-releasing hormone (CRH), which leads to the secretion of adrenocorticotrophic hormone (ACTH) from the

pituitary gland, which in turn leads to the release of cortisol into the bloodstream from the adrenal gland (Allen, Kennedy, Cryan, Dinan & Clark, 2014) in a pulsatile fashion (Young, Abelson & Lightman, 2004). Cortisol regulates its own release via the 'negative feedback loop' in the CNS, binding to specific receptors throughout the limbic system, including the hippocampus, amygdala and prefrontal cortex (Herman, Ostrander, Mueller & Figueiredo, 2005). In relation to acute reactivity this part of the stress response is less-easily triggered than the SAM axis and is activated under conditions of prolonged or extreme stress which exceed the individual's perceived ability to cope (Lazarus & Folkman, 1984), such as in response to highly socially evaluative laboratory stressors (e.g., Kirschbaum, Pirke & Hellhammer, 1993) or when participating in high-risk extreme sports (e.g., skydiving: Hare, Wetherell & Smith, 2013; and rock climbing: Hodgson, Draper, McMorris, Jones, Fryer & Coleman, 2009).

Early theories of stress described the stress response as a defence mechanism, operated by physiological systems in place to protect the organism from environmental challenge to bodily processes, the "sum of all nonspecific changes (within an organism) caused by function or damage" (coined the General Adaption Syndrome hypothesis; GAS: Selye, 1946). Selye postulated that stressors of all varieties, regardless of level of threat, elicit a uniform, general reaction to external stressors following a three-stage sequence. During the first stage, the 'alarm reaction', the adrenal medulla secretes adrenaline and the adrenal cortex produces glucocorticoids, both of which are responsible for aiding homeostasis. This restoration of homeostasis leads to the second stage, termed 'resistance'. At this stage defence and adaption are sustained. If the threat persists,

the third stage, 'exhaustion', follows. This prolonged exposure to stress, that is, chronic stress, eventually facilitates the cessation of the adaptive response which, in turn, can result in 'diseases of adaption'.

Whilst it is acknowledged that the GAS hypothesis highlighted the negative impact that persistent stress can impose on the immune system, since its inception, approaches to stress have progressed to become more sophisticated, and the complexity of the response with regards to the impact of social and individual contexts is more widely acknowledged. A plethora of studies have since demonstrated the adaptive abilities of the stress response, as well as the different patterns of response that are related to specific types of stressor (for review, see Dickerson & Kemeny, 2004). An additional shortfall of Selye's GAS hypothesis is that the fight or flight response has since been suggested not to apply equally to all individuals: for example, sex differences in stress responses have been demonstrated, with suggestions that that the fight or flight response, as it has typically been described, applies specifically to male animals only (McEwen, 2005).

#### *The role of cortisol in healthy physiological functioning*

One of the key mediators involved in the stress response is cortisol, the hormonal end-product of the HPA axis. Cortisol is a glucocorticoid, and as such it has a major role in the rapid mobilisation of amino acids and fat from cells to be used as energy (Levine, Zagoory-Sharon, Feldman, Lewis & Weller, 2007). Whilst playing a fundamental role in the acute stress response, cortisol also has a pivotal role in healthy physiological functioning, supporting emotion-appropriate behaviour through the regulation of metabolic processes, where it regulates

glucose storage (Sapolsky, Romero & Munck, 2000). It assists in the regulation of immune functioning (Sheridan, Dobbs, Brown & Zwillig, 1994), controlling the magnitude and duration of inflammatory responses to stressors (Sapolsky et al., 2000).

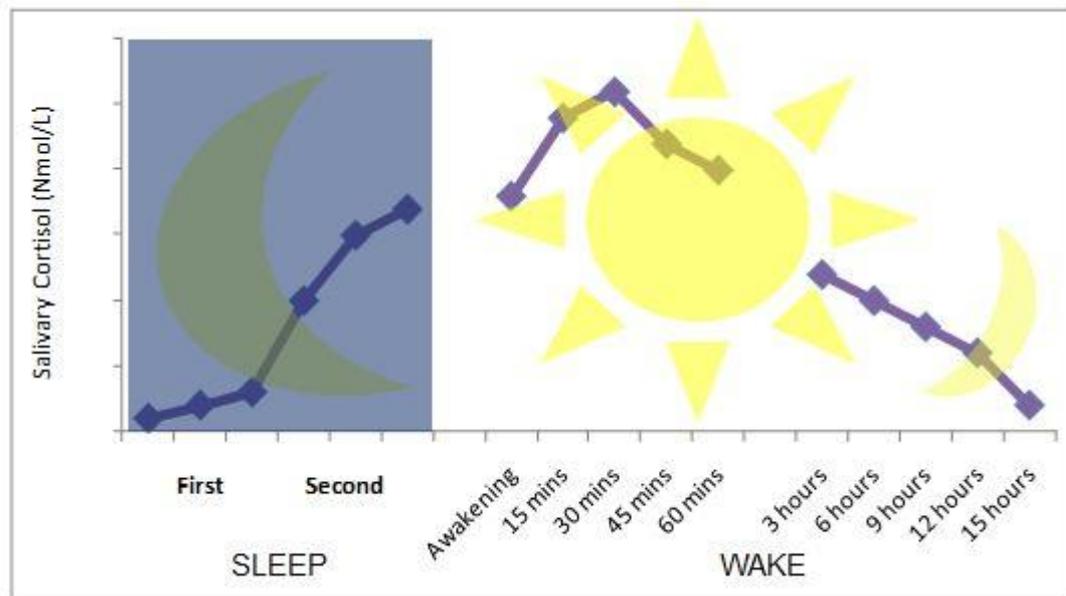
In healthy individuals cortisol levels also follow a well-documented circadian rhythm, which develops during the first year of life (de Weerth, Zijl & Buitelaar, 2003). This circadian rhythm is adaptive, demonstrated through findings of disrupted profiles in response to particular environmental influences, such as exposure to acute stressors and chronic stress (McEwen, 1998).

The highest levels of cortisol secretion are observed following awakening (see Figure 1.2), followed by a decline throughout the day, and levels reach a nadir in the late evening (Gunnar & Cheatham, 2003; Saxbe, 2008). The secretion of cortisol is at its lowest at sleep onset, and throughout the slow wave sleep period. Cortisol concentrations continue to decline slowly until a few hours before awakening, when they gradually begin to rise (Steiger, 2002; Wilhelm, Born, Kudielka, Schlotz & Wüst, 2007). Immediately following waking, cortisol typically increases between 50% and 160%, with the highest peak of secretion between 30-45 minutes post-awakening (Clow et al., 2004; Pruessner et al., 1997). This phenomenon is a distinct facet of the circadian cortisol rhythm, and is referred to as the cortisol awakening response (CAR: Pruessner et al., 1997).

The CAR maintains relative stability across days and weeks, suggesting that the response is a trait characteristic (Pruessner et al., 1997). Sleep duration and quality have, however, been found to have an impact on observed dynamics of the CAR. For example, some studies have suggested an influence of waking

time and sleep duration on morning peak levels of cortisol; earlier awakening has been associated with a greater increase in cortisol (Edwards et al., 2001; Kudielka & Kirschbaum, 2003). However, these findings lack consistency, with contradictory reports that later awakening is linked with higher waking levels of cortisol (Stalder, Hucklebridge, Evans & Clow, 2009).

The associations between longer sleep duration, lower evening levels, and steeper diurnal decline of cortisol could suggest that longer sleep duration is related to more dynamic cortisol secretion. However, other studies have found associations between longer sleep duration and a reduced CAR, which suggests a relationship between longer sleep duration and a less dynamic response. Shift-work studies have provided further support for the notion that the CAR adapts to changes in awakening times (e.g., Kudielka, Buchtal, Uhde & Wüst, 2007). However, the findings in the literature are mixed (Kunz-Ebrecht, Kirschbaum & Steptoe, 2004), with other studies demonstrating no effect of time of waking or sleep duration on the CAR (Wüst, Wolf, Hellhammer, Federenko, Schommer & Kirschbaum, 2000). Associations between sleep quality in relation to anticipation of a stressor and the CAR will be considered further in Chapter 5 and 6.



**Figure 1.2** The circadian (24 hour) cortisol profile (supplied with permission from Dr Mark A. Wetherell).

*The measurement of cortisol and the predicament of adherence*

To assess the circadian rhythm of cortisol in humans, the main methods adopted in physiological studies are through peripheral blood and saliva. Whilst blood sampling has, by some, been a recommended method in terms of integrity (e.g., Hellhammer, Wüst & Kudielka, 2009) the method encompasses a number of disadvantages: firstly, there have been suggestions that the method of venipuncture itself may elicit activation of the HPA axis (Kirschbaum & Hellhammer, 2000); it is also labour-intensive, expensive and is considerably more invasive than saliva sampling, and therefore fewer samples can be collected during a short space of time. Recently, however, a growing number of studies have observed a linear relationship between blood and salivary cortisol concentrations (e.g., Hellhammer et al., 2009; Nater et al., 2008).

The use of salivary cortisol as a biomarker of HPA axis activation is therefore advantageous in that the collection of the samples is non-invasive, meaning samples can be collected repeatedly throughout the day, and under a number of different conditions (Miller et al., 2013). In addition, saliva collection does not require a technician to withdraw the samples, as is the case when collecting blood samples. Furthermore, cortisol assessed through saliva sampling is unbound (biologically active) whereas only 2-15% of cortisol released into the bloodstream is unbound or free, with the majority binding rapidly to carriers such as corticosteroid-binding globin (CBG) and albumin (Kirschbaum & Hellhammer, 2000). Therefore saliva sampling provides a more reliable indication of biologically active cortisol. Due to the advantageous non-invasiveness, and the ability to collect samples at an almost unlimited frequency (Kirschbaum & Hellhammer, 1994), saliva sampling is the most practical, thus, perhaps popular, method of cortisol measurement.

Although the measurement of cortisol through saliva is advantageous, perhaps unsurprisingly, the assessment of the CAR in particular has, due to the nature of the response (i.e. the sensitivity of the observed cortisol levels to strict timings of samples), been a longstanding challenge for those wishing to accurately measure the phenomenon using saliva. The CAR has been statistically assessed using a variety of methods, including repeated measures analysis of samples taken at several consecutive time points between waking and 60 minutes later, and area under the curve (AUC) indices (Clow, Thorn, Evans & Hucklebridge, 2004). Two key parameters have been assessed: overall concentration of cortisol released over

the waking period, and the change (typically, increase) of cortisol from the level recorded upon awakening (Kudielka & Kirschbaum, 2003).

Studies have demonstrated that when the self-provided 'waking' sample is delayed by >15 minutes, the CAR appears reduced (or blunted) and this is due to the CAR commencing prior to the collection of the waking sample (Stalder et al., 2016). This is just one example demonstrating the importance of strict adherence to sampling protocol, and as such, highlights an issue that must be carefully considered in studies involving the assessment of the CAR (Stalder et al., 2016).

The accurate adherence to timings of saliva samples when being provided in domestic settings is a recurrent issue with this method of cortisol assessment as it relies on participants' adherence to strict protocol, providing samples at specific intervals following awakening in order to provide an accurate diurnal profile (particularly when measuring the CAR). In an attempt to monitor sampling times, saliva sampling diaries are typically requested to be completed by participants, whereby they report the times at which they provide each sample. However when these sampling times are studied objectively, the results can be a cause for concern (e.g., Kudielka et al., 2003). Saliva sample collection times have been studied using electronically tagged containers which record the times at which saliva collecting devices are opened (Kudielka et al., 2003), demonstrating that not only do a concerning number of participants not provide samples when instructed, but that individuals whose subjective, self-reported time and objective time were discrepant showed only a minimal increase in cortisol between waking and 30 minutes later (consistent with findings from Okun et al., 2010), instead of the large increase observed and expected in typically functioning individuals (e.g.,

Fries, Dettenborn, & Kirschbaum, 2009). In a separate study, non-compliant participants were reported to have a lower CAR than those who were compliant (Kunz-Ebrecht, Kirschbaum, Marmot & Steptoe, 2004). This further demonstrates the importance of adherence to sampling times, and of utilising objective measures, when practically possible, to check for discrepancies in reported collection times, as a delay in taking the first (waking) sample could mean that cortisol levels have already begun to rise prior to the 'waking' sample. As the aforementioned studies demonstrate, failure to provide the waking sample upon awakening can lead to a reduced CAR being observed (Kunz-Ebrecht et al., 2004), rendering the results misleading. However, more recent studies have revealed that self reported waking sampling times are consistent with objective waking times (Kraemer et al., 2006; Dockray Bhattacharyya, Molloy & Steptoe, 2008) and that this method is a suitable method of adherence monitoring, in cases where objective assessments are not possible. This method is especially successful when accompanied by training in the technique and emphasis of the importance of adherence has been sufficiently expressed to participants.

#### *The impact of psychosocial factors on cortisol secretion*

The relatively recent introduction of practical sampling methods for the measurement of cortisol has led to an increased interest in the assessment of psychosocial factors which may influence diurnal secretion. Greater cortisol output across the day has been associated with a number of psychosocial and environmental factors: a number of internalising disorders including anxiety and depressive symptoms have been associated with higher evening levels of cortisol, when levels are typically at their nadir in healthy individuals (Young, Carlson &

Brown, 2001). In children in foster care placements for a duration of 2 to 45 months, an environment which is typically characterised by uncontrollability and extended separation from parents, greater cortisol output has been observed across the day, compared with children continuously living with their parents (Dozier et al., 2006). Greater diurnal cortisol secretion has been observed on ‘bad’ compared to ‘good’ days in assembly line workers, and has also been associated with greater mental strain (Lundberg, 1989). Lower socioeconomic status has also been associated with greater cortisol secretion across the day; with lower grade male workers demonstrating greater cortisol compared with higher grade workers (Steptoe et al., 2003).

Despite findings demonstrating altered diurnal profiles of cortisol across the day, dependent on psychosocial factors, the CAR has also been studied in relation to specific events, and the assessment of the CAR is widely used as a key biomarker of integrity and function of the HPA axis. However, as the circadian rhythm of cortisol secretion and the CAR are two distinct processes, associations with psychosocial factors are often observed in one of these indices, but not the other (e.g., enhanced CARs but not greater diurnal secretion when comparing high and low job strain; Steptoe, Cropley, Griffith & Kirschbaum, 2000).

In healthy populations increased CARs have been observed on workdays, characterised by increased demands, compared with weekends, that are associated with fewer demands, in civil servants (Kunz-Ebrecht, et al., 2004) as well as in school teachers experiencing job-strain (Steptoe et al., 2000) and newly qualified doctors at the beginning of a clinical placement (Brant, Wetherell, Lightman, Crown & Vedhara, 2010). Greater CARs are also observed in individuals

reporting high chronic stress compared with low stress (Schulz, Kirschbaum, Pruessner & Hellhammer, 1998) and those experiencing chronic work overload (Schlotz, Hellhammer, Schulz & Stone, 2004) social stress, and worry (Wüst et al., 2000). In another study examining cortisol secretion in civil servants, those high in work over-commitment also demonstrated greater CARs and cortisol output over the day compared to their low over-commitment counterparts (Steptoe, Siegrist, Kirschbaum & Marmot, 2004). In a single case study, assessing diurnal cortisol secretion over 48 days in a healthy, 27 year old male, days which included a greater number of planned commitments, and therefore anticipated as busier, had greater CARs than comparably less busy days (Stalder, Evans, Hucklebridge & Clow, 2010).

Caregivers, a group who typically experience greater burden, anxiety, and depressive symptoms compared with the general population (Argüelles, Loewenstein, Eisdorfer & Argüelles, 2001) have demonstrated greater CARs; in caregivers of Alzheimer's Disease patients compared with healthy senior non-caregivers (Wahbeh, Kishiyama, Zajdel & Oken, 2008), and Dementia patient caregivers compared with age-matched non-caregiver controls (de Vugt et al., 2005). The level of demand also appears to affect the CAR, with higher waking levels of cortisol in caregivers of patients with high behavioural and psychological symptoms of Dementia (BPSD) compared with carers of patients with low BPSD (de Vugt et al., 2005), indicative of a potential dose-response relationship.

Furthermore, naturalistic studies measuring cortisol concentrations from awakening on the day of specific, challenging events have also observed greater concentrations on these days compared with non-event days. In dancers

participating in competitive ballroom competitions, where participants are assessed based on their performance and critically compared with other competitors, greater waking cortisol levels were observed on the day of the competition compared with a typical control day (Rohleder, Beulen, Chen, Wolf & Kirschbaum, 2007). In college students preparing for examinations, CARs were assessed over 7 low examination days (either the term before or after examinations) and 2 high examination days (during the examination period), which were counterbalanced, as well as providing self-reported mood assessments. CARs were greater during the high examination period compared with the low examination period, as were feelings of anxiety and stress, and students also reported lower feelings of happiness (Weekes et al., 2008). On the day of graded demonstration lessons, characterised as a formal and strenuous teacher training assessment, higher waking levels of cortisol were observed in student teachers on the day of the observation compared with a typical day (Wolfram, Bellingrath, Feuerhahn & Kudielka, 2013). Furthermore, in a sample of newly qualified doctors who were preparing for the national professional examination (an assessment involving their results being ranked and their rank order determining whether or not they would be given the opportunity to carry out specialist medical training) cortisol was significantly higher on the days leading up to the examination, compared with those after (Gonzalez-Cabrera, Fernandez-Prada, Iribar-Ibabe & Peinado, 2014). Similar responses have been observed on the morning of a socially salient laboratory stressor, whereby greater CARs were observed on the day participants attended the laboratory for a pre-planned,

socially evaluative stressor, compared with the day prior to the stressor (Wetherell, Lovell & Smith, 2014).

Although the distinct physiological role of the CAR remains relatively unknown, the observed associations between psychosocial factors and the CAR demonstrate that the CAR can be altered by proximate characteristics and that higher demand is associated with greater levels of cortisol upon awakening. Some studies have further suggested a possible dose-response relationship in this association, with differing levels of challenge producing greater morning cortisol secretion (e.g., greater CARs in caregivers of patients with high versus low BPSD; de Vugt et al., 2005; greater perceived social-evaluative threat is associated with greater cortisol; Rohleder et al., 2007). These associations withstand controlling for sleep-related factors such as waking time (Chida & Steptoe, 2009) and therefore there is general consensus that the CAR is not merely a response to waking, but that other underlying pathways must be involved in the function of the response. It has therefore been proposed that the CAR is an adaptive phenomenon, assisting with the process of preparing an individual for coping with forthcoming demands of the day, with greater CAR responses reflecting the need for more energy to meet demands (Schulz, Kirschbaum, Pruessner & Hellhammer, 1998). More specifically, the CAR seems to be linked with the anticipation of forthcoming demands of the day, and is associated with the reactivation of memory representation via activation of neocortical networks, thereby stimulating HPA axis activity (Powell & Schlotz, 2012).

Greater CARs have consistently been reported on days characterised by higher demand or challenge, however, the specific pathways through which the

CAR prepares an individual for forthcoming demand is unclear. However, increased CARs have been associated with lower levels of fatigue, increased alertness, and energy (Adam et al., 2006; Law et al., 2013) all of which could have beneficial effects in some of the circumstances discussed thus far; when sitting examinations (Weekes et al., 2008); participating in elite competitions (Rohleder et al., 2007); and completing a cognitively demanding multitasking stressor (Wetherell et al., 2014). Anticipation of challenging events and forthcoming demand will be discussed in more depth in a later chapter (see Chapter 5).

#### *Dysregulation of the HPA axis*

Whilst the anticipatory response is useful in the short term and promotes survival, chronic overestimation of forthcoming demand can lead to inappropriate and excessive activation of the HPA axis, which, if extended over a long period, can become chronic, and subsequently have negative long-term effects on health (Schulkin, 2011) due to exhaustion of stress mediators (McEwen, 2005). Blunted CARs, characterised by flattened or absent responses following awakening, have been observed in individuals suffering from post-traumatic stress disorder (PTSD) compared with both trauma exposed non-PTSD individuals and healthy controls (Wessa, Rohleder, Kirschbaum & Flor, 2006), and chronic fatigue syndrome patients compared with healthy controls (Roberts, Wessely, Chalder, Papadopoulos & Cleare, 2004). Attenuated CARs have also been observed in teachers experiencing high symptomology of burnout compared with low burnout (Pruessner et al., 1999), depression (Stetler & Miller, 2005; Huber, Issa, Schik & Wolf, 2006) and chronic pain (Geiss, Varadi, Steinbach, Bauer & Anton, 1997).

Furthermore, in a military sample participating in an intense training programme over several weeks, attenuated CARs were also observed (Clow et al., 2006), indicating a possible response to exhaustion (Law, Hucklebridge, Thorn, Evans & Clow, 2013).

Chronic stress is associated with overproduction of stress responsive adrenal glands (cortisol and catecholamines) and the above-mentioned dysregulation of the CAR and diurnal cortisol profile (McEwen, 1998). Excessive or long-term exposure to glucocorticoids (particularly cortisol) is linked to neuronal cell death (Sapolsky, 1986), cancer and depression, as well as a plethora of mental health problems (Shonkoff & Bales, 2011; Bauer, Jeckel & Luz, 2009). Chronic psychological stress is also a major risk factor for cardiovascular disease (e.g., Yusef et al., 2004).

Cortisol is essential and adaptive in order to facilitate the (acute) response to a stressor, however, as suggested by the previous examples, it can also lead to the dysregulation of healthy physiological functioning, if secreted for inappropriately long periods of time (for example, in the case of chronic stress). The protective and damaging effects of the physiological response to stress have become known as “allostasis” and “allostatic load” (McEwen, 1998), thus morphing the GAS hypothesis of stress into the Allostasis-Allostatic Load Model (Sterling & Eyer, 1988). This model postulates that every living organism strives to maintain a complex and dynamic equilibrium or homeostasis, for survival. When homeostasis is threatened by a stressor in the environment, to the point that the stressor exceeds the temporal threshold, the body enters what is referred to as an allostatic state in order to cope with the demands being imposed on the system.

### *The adaptation of the stress response*

Allostasis refers to the adaptive processes by which “stability is achieved through change”, and occurs in order to maintain (or return to) homeostasis. During this process, an imbalance of the primary mediators occurs, providing an indication of the excessive production of some, and an inadequate production of others. These key changes in the central nervous system (CNS) lead to the facilitation of neural pathways which serve adaptive functions such as arousal and focused attention, whilst inhibiting neural pathways which serve acute non-adaptive (non essential) functions such as digestion, immune function and reproduction. It is this function which is commonly known as the fight or flight response (Cannon, 1932).

As an illustrative example, an antelope, with no warning, is attacked by a lion; the antelope is injured but manages to escape, and continues to be stalked and chased by its predator over the next hour, until the lion finally becomes exhausted and withdraws. During this event, the antelope’s life is threatened, so the organism enters an allostatic state, whereby acute non-essential systems cease to function, saving energy for increased function and efficiency of acute essential processes (focus and vigilance, as well as adrenaline release and cardiovascular arousal for ‘flight’, in this situation). Although the antelope is injured, the priority for survival is to escape, so the immune system/healing process does not activate until it has escaped the lion (threat). As the threat of the stressor remains for an hour or so, the HPA system remains activated in order to maintain homeostasis (survival). This process is referred to as allostasis. As the lion becomes exhausted

before the antelope expends all its energy resources, the prey is able to stop and recover from the event, so homeostasis can be restored.

This response is specifically designed for dealing with acute stress (i.e. for immediate and limited periods) and in these situations the stress response is both essential and adaptive (McEwen, 2000). When the body is under threat and in an allostatic state, the allostatic load of the organism increases. Allostatic load refers to the price the body pays for being forced to constantly adapt to adverse stressful events (Hackney, 2006). More specifically, it refers to either the presence of too much stress (challenge), or inefficient performance of the stress response. However, the allostatic state (allostasis) can only be sustained as long as there is sufficient fuel for the homeostatic mechanisms required. If the stressor exceeds the capabilities of allostasis (i.e. is activated for long periods and the stress becomes chronic), and allostatic load increases in intensity and frequency, this results in allostatic overload. Allostatic overload provides no benefit to the organism, only negative health outcomes, such increased risk for the development of chronic illnesses and disease (McEwen, 1998; 2000; 2003).

It should be considered that the challenges faced by hunter-gatherers to whom the fight or flight response originally applied are very different to those encountered by individuals on a daily basis in contemporary Western environments. Although it could be argued that there are, in some areas of modern society, individuals who may well face considerable struggle or demand on resources, generally, the challenges met by those today are largely subjective, and seldom circumstances requiring immediate physical threat to survival. Despite this consideration, the pressures experienced today appear to provoke the same

sequence of preparatory modifications that are observed when under evolutionary physical threat. This highlights why it is as important as ever to understand the consequences of aberrant and chronic responses to stress.

As mentioned above, it is now widely accepted that individuals may be exposed to a variety of different stressors, differing in levels of threat, intensity and duration. Although there are a number of different ways in which stressors may be categorised, in basic terms they can be classified as either psychosocial or biogenic. Psychosocial stressors refer to non-metabolically demanding stimuli which become stressors by virtue of the cognitive interpretation of the event (i.e. the meanings to which they are assigned: Lazarus & Folkman, 1984). For example, waiting in a queue is a neutral event, but can become a stressor depending on how the individual interprets the situation (i.e. as a threat or undesirable). In this situation, if the individual perceives the event as positive or neutral, no stress response occurs. However, if a negative appraisal is made, a stress response will follow. Some stressors are, however, inherently more stressful than others: for example, public speaking has been found to elicit a significantly greater magnitude of stress response than completing cognitive tasks in a wide range of studies (for review, see Dickerson & Kemeny, 2004). With such stressors there is less variation in the interpretation of the situation, though there are still individual differences with regards to how an individual adapts to the stressor.

#### *The assessment of acute stress: in natural settings and in the laboratory*

As the purpose of the fight or flight response is to prepare an organism for immediate physical action (to fight or flee), studies aiming to investigate real-life acute stress response have done so by conducting field studies, assessing

responses in those taking part in high-risk extreme sports (e.g., skydiving: Hare, Wetherell & Smith, 2013; and rock climbing: Hodgson, Draper, McMorris, Jones, Fryer & Coleman, 2009). Skydiving is perceived as a high-risk sport, but more specifically it is also characterised by high socio-evaluative threat and low perceived control. Despite laboratory studies demonstrating habituation to stressors in a relatively short period of time (Kirschbaum, 1995), this is not the case in individuals taking part in a skydive (i.e. whether taking part in their first or 1000<sup>th</sup> parachute descent, the physiological stress response is activated: Hare, Wetherell & Smith, 2013). This demonstrates that skydiving is a useful tool for assessing a real-life stress response, where the individual is responding to a potentially (perceived) life-threatening event. These studies have demonstrated robust stress responses in real-world settings, however, naturalistic studies lack experimenter control and standardisation of protocol, and therefore, in order to study the physiological stress response in a controlled environment, there is a need to create situations that will be perceived as stressful to individuals in the laboratory.

A variety of different psychologically challenging stressors have been developed as tools for studying stress in the laboratory (for a full discussion of stressors, see Chapter 3). Although they vary both with regards to the methods through which they elicit arousal (such as through social evaluation, cognitive demand, and so on) and the magnitude of stress response elicited, generally, physical components are kept to a minimum.

### *Conclusions*

At this point, both the adaptive and pathophysiological effects of physiological mediators involved in the stress response have been discussed. The study of individuals when they are experiencing stress is essential in order to broaden knowledge about the stress mechanisms which are activated during stress, not only because responses to modern stressors appear to elicit patterns of reactivity similar to those of risk to survival, but also because excessive stress has become widely recognised as a significant challenge to public health (Everly & Lating, 2012). Acquiring knowledge of the protective factors which could buffer the negative effects of stress on well-being could help increase resilience in those recognised as at greater risk of stress-related ill-health.

This research programme will evaluate and contribute to the existing literature relating to the study of the stress response through the utilisation of both a laboratory and naturalistic paradigm. These models of stress will be employed to assess the anticipatory stress responses preceding forthcoming demand, and the subsequent recovery period following the event. Furthermore, in light of the purported influence of psychosocial and individual difference factors on stress reactivity, the final empirical chapter of this thesis will assess these responses in the context of individual differences, in order to examine and identify characteristics which may predispose individuals to greater risk of poor health outcomes, or serve to buffer these negative effects.

## **Chapter 2**

### General methodology

This chapter will detail the general methods used in the programme. Subsequent empirical chapters will describe methods specific to each study exclusively.

### *Salivary cortisol*

#### *Sample collection and storage*

Samples were collected using Salivette saliva collection tubes (Sarstedt, Germany). These tubes comprise a cotton roll within a plastic tube; this cotton swab is inserted to the mouth, moved to the inside of the cheek, and participants are instructed to engage in a chewing motion for 3 minutes in order to obtain the sample (see Figure 2.1 for an illustration of this process). Saliva samples were stored in domestic fridges until they were returned to the researcher at the participants' earliest convenience, where they were subsequently frozen immediately at  $-20^{\circ}\text{C}$  and remained frozen until they were removed for assay. To avoid contamination of samples with interfering substances, participants were requested to refrain from eating, drinking and smoking within 30 minutes prior to providing a sample (Salimetrics Europe, Newmarket, United Kingdom).



**Figure 2.1** Collection of saliva using Salivettes (Sarstedt, Germany).

### *Sample handling and processing*

On the day of assay, samples were thawed, vortexed and centrifuged at 3000 rpm for 15 minutes to extract the saliva from the cotton swab. This process removes mucins and particulate matter which could otherwise interfere with antibody binding, which could lead to falsely elevated results (Schwartz, Granger, Susman, Gunnar & Laird, 1998). All assays were conducted in house using the enzyme-linked immunosorbent assay method (Salimetrics Europe, Cambridge UK, intra and inter assay coefficients <10%), in accordance with the manufacturer's instructions. Cortisol values were converted to nmol (ug/dl x 27.6) prior to statistical analysis.

### *Treatment of data*

Cortisol data was treated in two ways to provide different markers of HPA axis activity; firstly, CAR magnitude was calculated as the difference between cortisol levels at waking and peak cortisol during the waking period (i.e. within the first 45 minutes post awakening). In addition, total cortisol secretion for each day was assessed by area under the curve ( $AUC_G$ ).  $AUC_G$  was calculated by using the cortisol value (nmol) at each sampling point and the time (minutes) between each sample (Pruessner et al., 2003). CAR magnitude and  $AUC_G$  were not calculated for participants who failed to provide data for the required time points. To correct skew, raw cortisol values were  $\log_{10}$  transformed as necessary.

### *Adherence to protocol*

The Cortisol Awakening Response (CAR) is characterised by a rapid increase, and consequent decrease in cortisol secretion following awakening (Wilhelm et al., 2007). The measurement of cortisol in saliva therefore requires

strict adherence to sampling protocol, and this is particularly important when measuring the CAR. When the first sample is delayed in relation to awakening, the cortisol concentration can be considerably higher, thus a smaller increase to the maximum concentration is observed (typically observed +30: Fries, Dettenborn, & Kirschbaum, 2009). Discrepancies such as this can be detrimental to research findings, as this can reduce the apparent size of the response, with blunted and even negative CARs being reported as a function of measurement rather than for clinical reasons. As such, it is crucial that the first waking sample is collected immediately upon awakening, and that the timing of the following samples (+30, +45, and +60) are also strictly adhered to.

A number of methods have been utilised to investigate the relationship between subjective and objective sampling times, with varying levels of reliability (Stalder et al., 2016). However, whilst the use of objective measures is considered optimal, these methods are often costly, and the practical implication of obtaining such measures can be prohibitive. Therefore the utilisation of objective monitoring in a random subsample can provide an indication of the degree of non-compliance that may be occurring in the sample as a whole, although this method does not allow for identification of non-adherent participants across the full sample. However, despite some concerns reported in the literature with regard to the accuracy of self-reported saliva samples (Kudielka, Broderick, & Kirschbaum, 2003) evidence suggests that subjective assessments can accurately represent adherence to waking samples (Kraemer et al., 2006; Dockray et al., 2008), especially when accompanied by training and when the importance of adherence is thoroughly expressed to the participant. Furthermore, participant adherence to

saliva sampling procedures is higher when participants are aware that their compliance is being monitored, compared to those who are unaware (Broderick et al., 2004). In a sample of patients and healthy adults, objective compliance among participants who were unaware of monitoring was 71%, while self-reported compliance was 93%. However, those who were aware that the assessments were monitored had compliance rates of 90%, which was consistent with their self-reported compliance (93%). These findings indicate that self-reports can provide a reliable assessment of adherence, when participants are aware that compliance will be determined.

In addition to subjective measures of adherence, the more recent use of actigraphy (see below) as an objective measure of waking time has been demonstrated to be an effective objective method, which can be cross referenced with self-reported sampling times, enabling data to be removed if the times are not consistent and non-adherence is suspected (see following sub-section for further details).

The present research programme encouraged adherence by including the following methods: providing a thorough demonstration and briefing of saliva sampling to participants prior to the study, whereby the importance of accurate timings of samples was expressed clearly; notifying participants that their data would be assessed for adherence to protocol; sending text reminders to participants the night before sampling days, reminding them to prepare their salivettes for the following morning; requesting that participants complete a saliva sampling diary and a sleep diary; providing activity monitors to a random subsample of participants.

Collectively, this information allowed cross-examination of the data to ensure that the time of waking reported by participants matched the time at which the first saliva sample (for the time of waking) was provided. Based on previous findings that waking samples delayed by more than 15 minutes can significantly impact upon the observed cortisol awakening response (Dockray et al., 2008; Okun et al., 2010) this was used as the cut-off, and therefore discrepancies >15 minutes between time of waking and waking samples were considered non-adherent to the protocol, and were thus excluded from cortisol data analysis.

### *Actigraphy*

Actigraphy is a technique which allows the objective assessment of sleep and waking through the use of actiwatches. Actigraphs are small motion detectors capable of distinguishing wakefulness from sleep based on algorithms representing reduced movement characterising the sleep state. The devices therefore provide an objective measure of sleep and awakening, which can be cross-referenced with subjective sleep diary entries to ensure adherence to protocol has been maintained.

Participants were asked to wear a tri-axial accelerometer (GENEActiv, Activinsights Ltd, Kimbolton, UK) on their non-dominant wrist for the duration of Studies 2 and 3. These devices are effective in measuring sleep duration (van Hees et al., 2015), the sleep-wake cycle (Anderson et al., 2013), number of awakenings, sleep onset latency, and activity levels. This information was derived from data collected over the course of one week to measure both the anticipatory period leading up to the stressor, and the recovery period following the 'stressful' event.

The current programme used actigraphy to objectively report timings of samples as a method to monitor adherence to protocol.

### *Questionnaires*

Participants across all studies in the present research programme were required to provide demographic information, in addition to the following questionnaires:

#### *Morning (state) mood and sleep quality*

In order to assess morning levels of mood and sleep quality, and to measure potential anticipation of the day ahead, a questionnaire was devised combining the State anxiety scale (see below) and two additional Visual Analogue Scale (VAS) items, scored from 0-100: happiness and stress. In relation to sleep, the scale included items asking when the participant went to bed the night before, when they woke up the present morning, how mentally alert and physically tense they were in bed the night before, and how well they felt on that morning. With regards to direct anticipation items, VAS items were included asking ‘to what extent have you been thinking about the [stressor session/skydive/task]’, and ‘to what extent have you been worrying about the [stressor session/skydive/task]’. In order to ensure any observed responses were not due to confounding variables relating to understanding about the protocol, or other events of the sampling days, the following items were included: ‘To what extent do you understand about what you are going to do?’ and ‘Other than taking part in the study, is this a typical day for you? If no, briefly describe why’. Females were also required to provide menstrual cycle information (Almeida, McGonagle & King, 2009).

### *Physical symptoms*

*Cohen-Hoberman Inventory of Physical Symptoms (CHIPS: Cohen & Hoberman, 1983)*

Physical symptoms were measured using the CHIPS, a 33-item list of physical symptoms including items such as ‘headache’, ‘nausea’, and ‘muscle cramps’. Items are rated for the degree to which the health complaint affected the individual during the previous 2 weeks. Response choices range on a five-point Likert scale, from 0 (‘not at all’) to 4 (‘extremely’). Higher total scores represent high levels of reported physical symptoms. The scale has been utilised in studies investigating associations of stress and sleep on health (Benham, 2010) and has demonstrated good psychometric qualities in both clinical and community samples (e.g., Benham, 2010; Rutter, Weatheril, Krill, Orazem & Taft, 2013), with reported internal reliabilities (Cronbach’s alpha) of .88 (Lench, 2011) and .93 (Benham, 2010) respectively.

### *Anxiety*

#### *Trait anxiety*

*The State-Trait Anxiety Inventory (STAI: Spielberger, Gorsuch & Lushene, 1970)*

The trait anxiety subscale of the STAI measures one’s general levels of anxiety, assessing what is deemed a stable individual difference in proneness to experience anxiety, and tendencies to perceive stressful situations as threatening (Spielberger & Reheiser, 2009). Participants are required to report how they ‘generally feel’ in response to a number of statements, including: ‘I am happy’, and ‘I feel nervous and restless’. Responses are marked on a 5-point Likert scale, with anchors ranging from 0 (‘not at all’) to 3 (‘most of the time’). Participants’

level of trait anxiety is calculated by positively scoring emotions suggesting anxiety (e.g., ‘I feel like a failure’) and reverse scoring those that do not (e.g., ‘I feel secure’). Higher total scores indicate considerable proneness to experience anxiety. The scale has been used in a number of samples, in healthy adult populations, for university students (e.g., Mehroof & Griffiths, 2010), and clinical populations (e.g., Perpiñá-Galvañ, Richart-Martínez & Cabañero-Martínez, 2011). Psychometric properties of the scale are acceptable, with internal reliabilities (Cronbach’s alpha) reported by Spielberger, Gorsuch, and Lushene (1970) at .86 for the trait anxiety subscale. Test-retest reliability coefficients are also acceptable, with a range between .73 to .86 over intervals of 20 to 104 days in university students (Spielberger, 2009).

#### *State anxiety*

#### *Short-form of the Spielberger State-Trait Anxiety Inventory (STAI: Marteau, & Bekker, 1992)*

The 6-item short form of the original STAI measures anxiety intensity at a particular time of subjective feelings of tension, apprehension, and worry. Participants are required to report their current levels of anxiety, on items such as: ‘I feel tense’ and ‘I feel calm’. An additional two items were added for individual assessment of stress and happiness (‘I feel happy’; ‘I feel stressed’). Response choices were modified from the original Likert scale to 100mm Visual Analogue Scale (VAS), still ranging from ‘not at all’ to ‘very much so’. Respondents mark on the line what they rate their current state to be in relation to each item. Internal consistencies of the scale provide a meaningful index of reliability, and these are

high, with coefficient alphas of .82 observed in samples of medical students and student nurses (Martineau & Bekker, 1992).

### *Personality*

#### *The Big Five Mini-Markers Personality Test (Saucier, 1994)*

The Big Five Mini-Markers is a shortened version of Goldberg's Big-Five Markers (Goldberg, 1992), developed by Saucier (1994). The questionnaire comprises 40 items (e.g., 'bashful', 'moody', 'kind' etc.) which measure five personality factors; extraversion, agreeableness, conscientiousness, emotional stability, and openness to experience (or intellect). Participants are required to rate themselves for each item on a 9-point Likert scale, with anchors ranging from 'extremely inaccurate' to 'extremely accurate'. Each trait is attributed to one of the five personality factors (if the trait has a loading of .30 or above, this trait is attributed to it); for example, 'unenvious', 'relaxed', 'moody', 'jealous', 'temperamental', 'envious', and 'touchy' are used to measure 'Emotional Stability'. Traits with a negative loading are reverse-scored. The sum of the scores of the attributed traits is divided by 8 (the number of traits per personality factor) and this gives the final score for the individual personality factor. Saucier and Ostendorf (1999) reported the following coefficient alpha reliabilities for the mini-markers questionnaire: .83 for extraversion, .81 for agreeableness, .83 for conscientiousness, .78 for emotional stability, and .78 for openness to experience, demonstrating acceptable internal reliability of the scale. Consistent with psychometric properties reported by Saucier and Ostendorf (1999), subsequent studies have reported internal reliability scores (Cronbach's alphas) between .75 and .82 (Lawson, Bundy & Harvey, 2007), further demonstrating good internal

reliability of the scale. The scale has been used in a range of clinical and community samples, maintaining good psychometric properties across these populations.

### *Type D personality*

#### *The DS14 (Denollet, 2005)*

The DS14 comprises 14 items with two 7-item subscales: negative affectivity (NA: e.g., “I am often in a bad mood”) and social inhibition (SI: e.g., “I find it hard to start a conversation”). Response choices range on a Likert scale from 0 (false) to 4 (true), with the maximum score on each being 28. The NA and SI subscales can be scored as individual continuous variables (range from 0-28). With regard to Type D assessment, two methods of calculation for this variable were analysed in the present research programme: as a dichotomous variable (whereby a score of  $\geq 10$  on both the NA and SI subscales results in classification as Type D, and scores  $< 10$  were classified as non-Type D) and as a continuous variable whereby the NA and SI scores were multiplied to produce a Type D score. The scale has been applied to both clinical and community samples (e.g., Barnett, Ledoux, Garcini & Baker, 2009; Denollet, 2005; Spindler, Kruse, Zwisler & Pedersen, 2009) and yields good psychometric properties with internal reliability scores (Cronbach’s alphas) at .88 and 3-month test-retest reliability scores ranging between .72 and .82 for both the NA and SI subscales (Denollet, 2005).

### *Perceived stress*

#### *Perceived Stress Scale (PSS-10: Cohen & Williamson, 1988)*

The PSS-10 is a 10-item scale measuring the degree to which one perceives aspects of one's life as uncontrollable and unpredictable. Sample items include: "In the last month..." 'How often have you felt confident about your ability to handle your personal problems?' and 'How often have you been upset because of something that happened unexpectedly?' Possible responses to each question range on a 5-point Likert scale ranging from 0 (never) to 4 (very often), indicating how often they have felt a certain way in the past month. Scores range from 0-40, with higher scores indicating greater perceived stress. The PSS-10 is a revised version of the originally published 14-item scale, and demonstrates marginally better psychometric properties than its predecessor. Cohen & Williamson (1988) reported adequate internal reliability for the PSS-10 at .85, and more recent studies have further supported its psychometric properties, reporting Cronbach's alphas of .89 (Benham, 2010) and .85 (Schiffirin & Nelson, 2010). A two-day test-retest reliability of .85 has also been reported (Cohen & Williamson, 1988). The scale has been used in non-clinical studies of perceived stress (e.g., Pizzagalli, Bogden, Ratner & Jahn, 2007), and with student populations, and is among the most widely used self-report assessment of perceived stress in studies of stress and health.

## *Coping style*

### *Brief COPE (Carver, 1997)*

Coping style was assessed using the Brief COPE, a shortened version of the COPE (Carver, Scheier & Weintraub, 1989). The scale contains 28 items measuring strategies adopted by individuals to cope with problems and stress. 14 conceptually different coping reactions are measured in the scale, some of which are seen to be adaptive, whilst others are considered problematic (Carver, 1997). Participants are instructed to read each statement and think about what they usually do when they are 'under a lot of stress'. Sample items include: 'I've been turning to work or other activities to take my mind off things' and 'I've been giving up trying to deal with it'. Response choices range on a Likert scale from 1 ('I haven't been doing this at all') to 4 ('I've been doing this a lot'), with high total scores being indicative of an 'avoidant' coping style, and lower scores demonstrating an 'approach' coping style. Items are assigned to a specific type of coping style, providing individual scores for the following: self-distraction; active coping; denial; substance use; use of emotional support; use of instrumental support; behavioural disengagement; venting; positive reframing; planning; humour; acceptance; religion; and self-blame. The Brief COPE has demonstrated adequate internal consistency in non-clinical populations, with coefficients for the 14 primary scales ranging between .50 and .66 (Berrocal, Pennato & Bernini, 2009; Carver, 1997).

### *Self-esteem*

#### *Rosenberg's Self Esteem Scale (RSE: Rosenberg, 1965)*

The RSE comprises 10 items assessing global self-esteem (GSE). Sample items include: 'I feel I do not have much to be proud of' and 'On the whole, I am satisfied with myself', with response choices ranging from 0 (strongly disagree) to 4 (strongly agree). Higher total scores are indicative of high GSE, whilst lower scores represent low GSE. The RSE is the most widely used scale to measure global self-esteem (Blascovich & Tomaka, 1991; Marsh, Scalas & Nagengast, 2010). Studies demonstrate good psychometric properties of the scale, with internal reliability scores ranging between .88 and .90 (Robins, Hendin & Trzesniewski, 2001).

### *Prospective memory*

#### *The Prospective and Retrospective Memory Questionnaire (PRMQ: Smith, Della Sala, Logie & Maylor, 2000)*

The PRMQ is a 16-item self-report scale measure of prospective and retrospective memory failure in daily life. Each item can be categorised across a total of three dimensions: eight items assess prospective memory (e.g., "Do you decide to do something in a few minutes time and then forget to do it?") and eight measure retrospective memory (e.g., "Do you fail to recognise a place you have visited before?"); self-cued memory (e.g., "Do you fail to recall things that have happened to you in the last few days?"); 'environmentally-cued memory (e.g., "Do you forget to buy something you planned to buy, like a birthday card, even when you see the shop?"); and finally short-term (e.g., "Do you forget something that you were told a few minutes before?") and long-term memory (e.g., "Do you

forget appointments if you are not prompted by someone else or by a reminder such as a calendar or diary?”). Responses are marked on a 5-point scale: Very often, Quite often, Sometimes, Rarely, and Never, with numerical values of 5 (very often) to 1 (never). Thus the larger the score, the greater the number of self-reported memory errors.

The psychometric properties of the scale are good, with the internal consistency (Cronbach’s alpha) of the scale reported at .89 (95% CI = .88 to .90: Crawford, Smith, Maylor, Della Sala & Logie, 2010). For the individual subscales internal consistencies were reported as .84 for the Prospective scale and .80 for Retrospective scale respectively.

### *Perseverative thinking*

#### *Perseverative Thinking Questionnaire (PTQ: Zetsche, Ehring & Ehlers, 2009)*

The Perseverative Thinking Questionnaire (PTQ) is a 15-item scale assessing repetitive negative thinking (RNT). The PTQ includes 3 items for each of the following thought characteristics: repetitive (e.g., ‘The same thoughts keep going through my mind again and again’); intrusive (e.g., ‘Thoughts intrude into my mind’); and difficult to disengage from (e.g., ‘I feel driven to continue dwelling on the same issue’). Responses are made on a Likert scale ranging from 0 (never) to 4 (almost always), thus the higher the score, the greater the individuals’ self-reported repetitive negative thinking. The PTQ has been utilised in research with both clinical and non-clinical populations and has been shown to demonstrate excellent psychometric properties; internal consistencies (Cronbach’s alpha) reported between .94 and .95, and test re-test reliabilities from .66 to .69 ( $p < .001$ : Ehring, Zetsche, Weidacker, Wahl, Schonfeld & Ehlers, 2011).

*Stressor manipulation (Studies 1 and 2 only)*

*The Multi-tasking Framework (MTF: Purple Research Solutions, UK)*

The MTF is a platform for the presentation of performance-driven and cognitively demanding tasks. The framework comprises eight tasks, each of which can be presented singularly or in combination, up to a maximum of four tasks, where each task occupies a quadrant of the screen. Levels of workload stress are manipulated either by increasing the number of tasks the participant must attend to, or by altering the difficulty of the tasks. This research programme used the maximum of four tasks: auditory monitoring, number tap, visual monitoring and a Stroop task, presented at medium workload intensity.

*Auditory monitoring*

The auditory monitoring element of the task requires participants to listen for a high pitch tone among low tones. Low tones are to be ignored, but when a high tone sounds, the participant must immediately click on the '@' button (see top left quadrant, Figure 2.2) to register their response. 10 points are awarded for correct responses, and deducted for missed responses.

*Number tap*

In the number tap quadrant of the framework (top right, Figure 2.2), there is a number keyboard to the right, and a presentation box to the left. At set intervals, a 10-digit number appears in the box and the participant is required to use the keyboard to the right to input the number into the box underneath. Once they have inputted the number, they are required to click on the green phone button (to the right of the type-in box) to submit their response. As with the other

tasks, 10 points are awarded for correct responses, and deducted for incorrect or missed responses.

### *Visual monitoring*

Visual monitoring is assessed with six ‘warning’ bars, presented side-by-side (see bottom left quadrant, Figure 2.2). The bars travel upwards at different speeds towards the warning line. When the 6<sup>th</sup> bar has reached the top, the warning sign flashes along the top of the task quadrant and the participant is required to click on them in the order in which they are numbered (the order in which they reached the top). 10 points are awarded for correct responses, 10 are deducted for incorrect or missed responses.

### *Stroop*

The Stroop quadrant of the task follows the original Stroop test of selective attention and response inhibition. Four coloured rectangles (blue, green, red and yellow) appear on the right hand side of the task (see bottom right quadrant, Figure 2.2). At set intervals a colour name appears to the left of the coloured rectangles. The aim of the task is to click the rectangle related to the font colour, regardless of the colour described (e.g., if the word ‘red’ appears in blue ink, the correct response is the blue rectangle). The task is time-pressured, with time-outs of 20 seconds on the medium intensity setting, and 10 points subtracted for every missed or incorrect response.

These tasks were chosen as they require a number of cognitive processes including perceptual, attentional, psychomotor and memory abilities, which are typically employed in everyday functioning. All tasks are performance related and

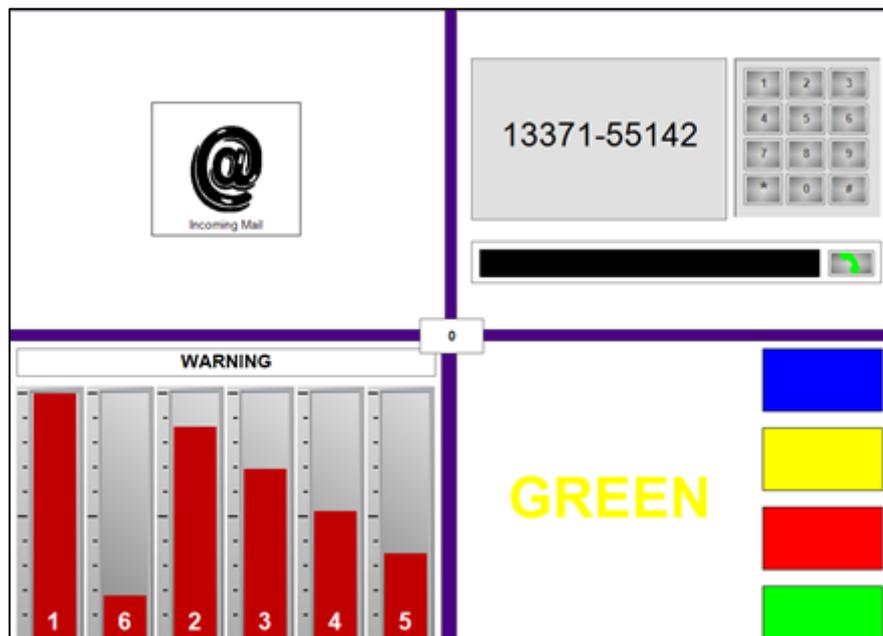
points are awarded for correct responses, and deducted for incorrect or missed responses. A running total score is presented in the middle of the screen and respondents are instructed to achieve as high a score as they can by being fast and accurate on all the tasks. The framework forms both the cognitive demand, and uncontrollability element of the stressor paradigm. The social evaluative threat element of the paradigm is incorporated by instructing participants to stand behind a podium, completing the task on the screen in front of them, whilst facing the experimenter. The task screen is projected onto the wall behind them, enabling the researcher to observe participants' performance throughout the study. Prior to the stressor commencement, participants are instructed to work as fast as they can to obtain the highest score possible, attending to all tasks. They are also notified that if later examination of their data reveals that they have not attended to all four tasks equally, that their data will not be able to be included in the final analysis. At set intervals during the stressor, participants are reminded to work as hard as they can to achieve their best score. The Multitasking Framework has demonstrated robust stress-inducing capabilities, evidenced through increases in heart rate, blood pressure, as well as negative effects on mood (Scholey et al., 2009; Wetherell & Carter, 2014) following exposure to the stressor.

In the present programme the MTF was set to medium intensity, for a 20 minute duration.

#### *Relaxation stimuli*

A 20-minute relaxation period followed stressor exposure. During this period, the researcher vacated the room, re-entering only to collect samples after 10 minutes. Participants watched a nature documentary, as utilised in similar

studies (Evans, Greaves-Lord, Euser, Franken & Huizink, 2012). The clip presented in the present paradigm was taken from Frozen Planet (2011: BBC documentary).



**Figure 2.2** The Multitasking Framework, including tasks presented in the present research programme (top left: auditory monitoring, top right: number tap, bottom left: visual monitoring, bottom right: Stroop).

*Cardiovascular reactivity assessment (Studies 1 and 2 only)*

*DINAMAP 200 V2*

The DINAMAP 200 V2 (1997) was used to record heart rate (HR), diastolic blood pressure (DBP), and systolic blood pressure (SBP) in Studies 1 and 2. The cuff was attached to the non-dominant arm.

In addition to their use in physiological research, the DINAMAP monitors are predominantly used in clinical settings, for which studies have demonstrated

they consistently meet their requirements for accuracy and reliability (e.g., de Greef, Reggiori & Shennan, 2007).

*Ethical statement*

All studies in the research programme were granted ethical approval by Northumbria University's School of Health and Life Sciences Ethics committee, acting in accordance with the Northumbria University Research Ethics and Governance Handbook.

## **Chapter 3**

Stressor paradigms and their application in stress research:

A literature review

This literature review has contributed to a chapter in the forthcoming International Handbook of Psychobiology.

## Introduction

Exposure to a situation perceived as challenging or threatening, which exceeds an individual's ability to cope, leads to a set of specific physiological stress responses which assist in managing the demand (e.g., Tomaka, Blascovich, Kelsey & Leitten, 1993). As described in detail previously (see Chapter 1) the physiological stress response to challenges in daily life mediates adaptation when activated under short-term conditions (McEwen, 2003). However, repeated and unnecessary activation of these processes is associated with adverse effects on cardiovascular, immune, metabolic and psychological health (McEwen, 1998).

In order to fully understand the pathways by which exposure to stress leads to deleterious health outcomes, it is necessary to develop methods which allow the observation of individuals while they are experiencing stress. Previous studies have employed a variety of different physiological and psychological stressors to assess responses of the HPA and SAM axes, and provide an insight into their effects on health, cognition and emotion (Schwabe, Hadda & Schachinger, 2008). These stressor paradigms vary considerably in terms of the mechanisms by which they elicit a stress response, and demonstrate varying levels of external and ecological validity (Chida & Hamer, 2008) as well as issues of ease of administration (e.g., Trier Social Stress Test: Kirschbaum, Pirke & Hell, 1993), proneness to habituation (e.g., Kirschbaum, 1995), and safety (e.g., Insulin Tolerance Test: Fraser, Albright & Smith, 1941).

Naturalistic stressor studies offer an ecologically valid method for assessing the processes involved in the stress response in real-world settings. Naturally-occurring paradigms which have been assessed for stress reactivity

include those involving assessments (college examinations: Weekes et al., 2008; trainee teacher class observations: Wolfram et al., 2013; and medical graduate examinations: Gonzalez-Cabrera, Fernandez-Prada, Iribar-Ibabe & Peinado, 2014); competitions (ballroom competitions: Rohleder et al., 2007; Judo competitions: Salvador, Suay, Gonzalez-Bono & Serrano, 2003; and powerlifting competitions: Le Panse et al., 2010); and high-risk sports (skydiving: Hare et al., 2013; rock-climbing: Hodgson et al., 2009; and paragliding: Filaire, Rouveix, Alix & Le Sanff, 2007). Whilst naturalistic studies are useful in illustrating responses to real-world stressors, the advantages of paradigms of this nature come at the cost of reduced control and standardisation, and the protocols are often relatively laborious (Reinhart, Schmahl, Wüst & Bohus, 2012).

Laboratory stressors on the other hand, provide an environment in which stimuli can be manipulated and any confounding factors can be robustly controlled (Kudielka, Hellhammer & Wüst, 2009). These benefits allow for more specific assessment of the causal factors involved in responses that may lead to such deleterious health outcomes (Wetherell & Carter, 2014). Furthermore, in order to retain the advantages of naturalistic stressors, efficient laboratory stressors should provide a 'snapshot' of how an individual would respond to a stressor encountered in a real-life setting (Wetherell et al., 2006). It is, therefore, of crucial importance that individuals being observed in the laboratory are engaging with a stressor that is representative of their experiences in natural settings.

Early theory defined stress as a non-specific response of the body, characterised by the secretion of glucocorticoids to any demand (Selye, 1946).

However, this ideology has since been challenged by theories that acknowledge the importance of emotional reactions and appraisals, which determine a specific stress response. An abundance of studies adopting a wide range of stressor paradigms have reported varying degrees to which these stressor tools stimulate the SAM and HPA axes, demonstrating a high level of variability in responses to specific types of stressor (Dickerson & Kemeny, 2004). Here we discuss a number of tools that have been used to assess the stress response under laboratory conditions, as well as the response patterns associated with each of them.

### *Physical stressors*

Stress responses have been observed following a number of physical stressor techniques. For example, the Insulin Tolerance Test (ITT: Fraser, Albright & Smith, 1941), a technique originally designed to evaluate suspected growth hormone deficiency in adults, has been used to test the integrity of the HPA axis in the laboratory (Lange et al., 2002). The ITT involves the administration of intravenous insulin to participants, a protocol which has been found to reliably trigger a cortisol response (e.g., Abdu, Elhadd, Neary & Clayton, 1998). However, this technique lacks the benefit of convenience for a number of reasons: firstly, and most crucially, the ITT is dangerous, with reports of deaths and serious adverse effects being reported in previous literature (e.g., Burke, 1992). Secondly, the protocol requires a specially trained technician to administer the ITT (Lange et al., 2002), which makes the technique limited in terms of ease of administration.

The heel prick technique is another physical stressor which has also been found (predominantly in new-born infants) to reliably provoke a stress response

(Buske-Kirschbaum et al., 2004) and which is routinely used in order to obtain a capillary blood sample to test for metabolic disease. This has been found to induce elevation of cortisol in new-borns (Buske-Kirschbaum et al., 2004; Mantagos et al., 1991) and provides an example of the use of stressors to demonstrate the stress response in infants. The heel-prick technique is so robust in its abilities to elicit a cortisol response, that it has also been used to test the integrity of HPA axis responsiveness to acute stress in new-borns with atopic disposition (Buske-Kirschbaum et al., 2004). However, this is a method which has only been investigated in new-born infants and, whilst it remains effective within this sample, there are many other stressors which are known to hold similar properties, in adult populations (and as such, are more widely utilised).

With regard to physical techniques more commonly used in stress research, one such method is the controlled inhalation of carbon dioxide (CO<sub>2</sub>). The inhalation of CO<sub>2</sub> is a technique, which, although originally implemented for assessment of the biological mechanisms in the aetiology of panic disorder (e.g., Papp et al., 1997), has also demonstrated promising physiological stress-inducing capabilities (Wetherell et al., 2006). The application of this stressor involves participants inhaling a mixture of CO<sub>2</sub> and oxygen, which has been found to demonstrate dose-response increases in both psychological (e.g., anxiety and fear) and somatic (e.g., dizziness) outcomes (Kaye et al., 2004). Physiological responses to CO<sub>2</sub> inhalation demonstrate the capability of the stressor to activate the SAM axis, with observed increases in systolic blood pressure, and decreases in heart rate (Wetherell et al., 2006). Increases in cortisol post CO<sub>2</sub> inhalation have also been observed, demonstrating activation of the HPA axis. Psychological

responses to CO<sub>2</sub> inhalation further demonstrate stress-inducing properties of the stressor with significant effects on mood: increases in levels of anxiety and fear were observed post CO<sub>2</sub> inhalation, as well as decreases in reported happiness (Wetherell et al., 2006). In addition to these findings, the inhalation of CO<sub>2</sub> has also proved a useful tool in distinguishing cortisol responders and non-responders (Wetherell et al., 2006).

The CO<sub>2</sub> inhalation technique provides a unique physical laboratory stressor that is capable of inducing a stress response across both the SAM and HPA axes and which also benefits from ease of administration. The stressor is useful in the examination of the pathways that can lead to deleterious outcomes, and defining and investigating individual differences in stress responding. However, despite these uses the stressor does not possess a great deal of ecological validity, and is therefore, limited in its use.

An additional physical stressor which has been widely used in the laboratory is the Cold Pressor Test (CPT: e.g., Gluck, Geliebter, Hung & Yahav, 2004; Lovallo, 1975). The CPT consists of a baseline period (usually 30 minutes) followed by submerging the hand up to the wrist in ice water (0-4°C) for 1 or 2 minutes. Physiological responses are assessed during the baseline period, during the CPT, and measured until responses return to baseline. The CPT was originally used as a means of experimentally increasing blood pressure in studies of hypertension (Hines & Brown, 1932) but its physiological stress-response-inducing properties have led to its use in the area of stress research. Previous studies have demonstrated the profound SAM axis activation capabilities of the CPT, evidenced by increased skin conductance (Buchanan et al., 2006), increased

heart rate, and elevated blood pressure (al'Absi, Petersen & Wittmers, 2002). However, the stressor has limited use with regards to the observation of the HPA axis activation: previous studies have found moderate increases in cortisol in response to the CPT (Gluck et al., 2004; al'Absi et al., 2002), but others have reported no cortisol elevation (Duncko, Johnson, Merikangas & Grillon, 2007), calling into question the CPT's HPA axis activation capabilities. More recently, however, the combination of the CPT and a socially evaluative component, whereby a researcher watches as the participant immerses their hand in cold water, has demonstrated a significant cortisol response, compared with immersion in warm water while being observed (SECPT: Schwabe, Haddad & Schachinger, 2008).

#### *Combined physical and psychosocial stressors*

Following the inception of the SECPT (Schwabe et al., 2008), other studies have developed stressor protocols combining both physical and psychosocial components. The Maastricht Acute Stress Test (MAST: Smeets et al., 2012) is a relatively novel stressor technique whereby participants are exposed to physical and psychological challenge. The stressor protocol involves a 5 minute preparation phase before participants are instructed to complete 5 socially evaluated cold pressor trials over 10 minutes, whereby during intervals they are required to perform a mental arithmetic task (counting backwards in steps of 17) as fast and accurately as possible. To form the socially evaluative element of the task subjects receive negative feedback when they make a mistake, and are instructed to start at the beginning. The MAST matches the CPT and SECPT with regards to observed patterns of subjective stress responses (stressfulness,

painfulness & unpleasantness) and cardiovascular responses, yet elicits a significantly greater magnitude of cortisol response (Smeets et al., 2012). These findings support suggestions of Dickerson and Kemeny's (2004) extensive review, whereby they propose a dose-response relationship with the inclusion of stressor domains reported to elicit a profound stress response: social evaluation elicits HPA axis reactivity, whilst physical stressors instantaneously trigger the activation of the ANS and SAM axis, leading to an enhanced response when these components are combined (e.g., Ulrich-Lai & Herman, 2009). The MAST has been successfully adjusted for use in the fMRI environment, named the imaging Maastricht Acute Stress Test (iMAST; Quaedflieg, Meyer & Smeets, 2013). As the name suggests, the protocol is almost identical to that of the original MAST, however, the impracticality of submerging participants' hands in ice-cold water whilst in the scanner led to modification of the procedure. In place of the cold pressor component of the original MAST, an fMRI compatible thermode set to 2°C is placed on participants' left forearm, and they receive 5 sets of exposure for a duration of 45, 60, or 90 seconds. As reported in the original protocol, the iMAST is effective in eliciting subjective stress, as well as cortisol reactivity (Quaedflieg et al., 2013).

### *Cognitive stressors*

A relatively novel stressor, The Mannheim Multicomponent Stress Test (MMST; Reinhardt, Schmahl, Wüst & Bohus, 2012), incorporates a cognitive domain, emotional domain (presentation of disgust and fear-related images), acoustic domain (continuous white noise) and motivational domain (threat of reducing financial reimbursement if performance is poor). Participants complete a

serial addition task while being shown negative-emotion inducing images and are exposed to increasing decibels of white noise. Although little research has been conducted using this stressor, the developers of the paradigm report significant increases in heart rate, self-reported anxiety and cortisol activity in response to the MMST (Reinhardt et al., 2012).

#### *Mental arithmetic stressors*

A further stressor, the Paced Auditory Serial Addition Test (PASAT: Gronwall, 1977), a paradigm originally developed as an experimental tool to examine the role of immediate memory and attention, has also been reported to elicit a stress response (Mathias, Stanford & Houston, 2004). The task requires participants to attend to the auditory presentation of a series of single digit numbers, where the last two digits must be summed. Participants are required to respond with the answer quickly, prior to the presentation of the next two digits, in order for a response to be scored as correct (and are thus under time-pressure). The PASAT assesses attention and information capacity (Cohen, 1993), as well as mathematical ability (Chronicle & MacGregor, 1998) and has been used to assess attentional deficits among patients with traumatic brain injury (Brooks, Fos, Greve & Hammond, 1999). However, despite its original, intended use, complaints that the PASAT was unpleasant for participants prompted studies investigating the effect of the task on mood and autonomic arousal (Mathias et al., 2004; Holdwick & Wingenfeld, 1999). With regard to stress-inducing properties, significant increases in heart rate and blood pressure have been observed in individuals exposed to the PASAT (Mathias et al., 2004). These effects have been noted specifically during the stressor, with higher heart rate and blood pressure

during the stressor, compared with baseline and recovery periods. In terms of subjective reports of psychological responses to the PASAT, a study focusing specifically on the task's effect on mood responses revealed a significant increase in negative mood during the faster trials of the task (Holdwick & Wingenfeld, 1999), suggesting that the task is perceived as negative or unpleasant. These findings indicate that the PASAT is both physiologically and psychologically taxing, evidenced through observations of activation of the SAM axis, and increases in negative mood. However, the PASAT's HPA axis activation capabilities have not yet been explored. Despite this, if the soon-to-be discussed Dickerson and Kemeny's (2004) guidelines are applied, HPA axis activation would not be expected in response to this stressor as the paradigm lacks key components required to reliably elicit such a response.

Similarly to the PASAT, other mental arithmetic tasks have also been implemented to induce a stress response in the laboratory, in a number of studies (Noto, Sato, Kudo, Kurata & Hirota, 2005). In a study conducted by Noto and colleagues (2005) participants were exposed to 15 minutes of mental arithmetic tasks, involving serial subtraction of 13 from a randomly selected 4-digit number presented on a computer. Responses were given verbally, and once an answer was confirmed to be correct, participants were required to continue subtracting 13 from the resulting number. With regard to stress-inducing capabilities of mental arithmetic stressors, subjective assessments of state anxiety taken throughout the task revealed significant increases in anxiety over the course of the stressor period, with a return to baseline occurring within 10 minutes following cessation. In terms of physiology, significant increases in heart rate have been observed

(e.g., Langewitz & Rüdell, 1989), demonstrating activation of the SAM axis. HPA axis activation has also been assessed in response to mental arithmetic, however, no changes in cortisol levels have, as yet, been reported in response to this task (Noto et al., 2005).

Users of mental arithmetic stressors consider the task to be a typical day-to-day environmental stressor due to its cognitive demand, time-pressured requirements, and the fact that high scores are rewarded (Sharpley et al., 2000). However, this is not a view shared across stress research, with others arguing that tasks of this nature are not ecologically valid (e.g., Chida & Hamer, 2008; Wetherell et al., 2006). Despite this disadvantage, both the PASAT and general mental arithmetic stressors have demonstrated capabilities in SAM axis activation, as well as effects on mood (Holdwick & Wingenfeld, 1999; Mathias et al., 2004; Noto et al., 2005).

#### *The role of specific stressor domains in physiological stress responding*

As the variety of responses to the discussed stressor paradigms demonstrate, there is considerable heterogeneity in the literature in support of the concept of an adaptive stress response, as not all types of negative situations/stressors trigger the same magnitude of psychological or physiological alteration (Dickerson & Kemeny, 2004; Mason, 1968). As the stress response is characterised by the reliable activation of both the HPA and SAM axes, along with associated changes in psychological responses, it could be argued that laboratory stressors with the ability to measure only one axis hold reduced value as a tool in stress research (e.g., Dienstbier, 1989).

In a meta analysis of acute laboratory stressors and cortisol responses, Dickerson and Kemeny (2004) identified three key stressor components which play an important role in the elicitation of the cortisol response, which can explain why not all acute stressors observe activation of both the SAM and HPA axes. These domains are described as social evaluative threat, uncontrollability, and motivated performance.

Social evaluative threat refers to sources or environments which threaten a valued aspect of self-identity, or the social self, and usually applies to situations where these valued resources are perceived as being, or are deemed to be at risk of being, negatively judged by others (Dickerson & Kemeny, 2004). As social organisms, humans possess a fundamental motivation to be accepted, liked and included by others, and social evaluation poses a threat to the primary human goal of achieving and maintaining a positive “social self” (Kenrick, Griskevicius, Neuberg & Shaller, 2010). In support of this theory, social stressors are amongst the most reliable forms of stress in humans and other species, reflected both via SAM and HPA axis activation (Dickerson & Kemeny, 2004; Tamashiro et al., 2005).

Another key stressor component, identified by Dickerson and Kemeny (2004), to play a role in the engagement of the HPA axis, is uncontrollability. In the context of a stressor paradigm, uncontrollability refers to the inability of the individual to affect an outcome by engaging in a behavioural response (Thompson, 1981) and usually applies specifically to the avoidance of negative consequences (Reinhardt, Schmahl, Wüst & Bohus, 2012). This domain is often linked with the social evaluation component of a stressor as participants may feel

unsure about how they will be judged by their assessors, thus often providing a feeling of uncontrollability.

Finally, motivated performance refers to tasks, like the majority of those previously discussed, which are performance-based, and which involve positive performance (e.g., high scores in a task) being rewarded. For example, receiving positive feedback/not receiving negative feedback or being ranked highly compared with peers. However, whilst all three of these stressor components form a successful formula for observations of stress in the laboratory, social evaluation is considered the most powerful in eliciting such a response (Dickerson & Kemeny, 2004), and stressors including this component are discussed in the following section.

#### *Psychosocial stressors*

As indicated previously, social stressors are amongst the most reliable in eliciting stress responses in humans and other species (Dickerson & Kemeny, 2004) and therefore a number of tools encompassing social evaluation have been used in stress research. Religious identity threat, for example, has been used as a stressor in Catholic, Protestant and Muslim populations (Ysseldyk, Matheson & Anisman, 2011). The study specifically focused attention to religious orientation: intrinsic (whereby religion facilitates spiritual development, guidance and meaning); and extrinsic (whereby religion is used primarily for personal or social benefits: Pargament & Park, 1997). The procedure required participants to read a fictitious article including survey results which indicated that the majority of Canadians report feelings of hostility towards [Catholics/Protestants/Muslims], and included a number of offensive quotes such as “I can’t stand

[Catholics/Protestants/Muslims]. It seems they have to involve religion in every single thing they do”. Following completion of reading the article, participants were notified that the regional government was considering including data from the survey in its funding discussions, with a view to potentially reduce financial support available to those affiliated with the particular religious faith, in order to heighten perceived religious threat. Religious identity threat was perceived as threatening towards both extrinsic and intrinsic religious motivations, and the findings indicate that when one’s valued religious group membership is threatened; personal perceptions of threat are increased. Whilst this paradigm is not a widely used stressor protocol, it demonstrates the range of stressors previously used to elicit and observe different forms of stress.

A number of stressor paradigms have demonstrated promising methods of assessing psychobiological stress under controlled conditions, however, the Trier Social Stress Test (TSST: Kirschbaum et al., 1993), a psychosocial stressor which combines all three of the earlier identified stressor domains, and is associated with consistent, high, levels of cortisol responding (Dickerson & Kemeny, 2004) is perhaps the best-known, and is considered the ‘gold standard’ laboratory stressor paradigm (Smeets et al., 2012). The standard TSST protocol involves participants standing in front of a panel of 3 assessors, wearing lab coats, who are seated at a table in front of them, and both a video camera and tape recorder documenting the session. Prior to facing the panel, participants are briefed and instructed to prepare to deliver 5 minutes of free speech to convince the panel that they are the perfect applicant for a fictional job vacancy. Following a 10-minute preparation period (which also acts as an anticipatory period) participants return to the panel to

deliver their 5-minute speech. If the speech ends before 5 minutes has passed, a member of the panel tells the participant to continue. Once the speech is over, the participant is asked to serially subtract 13 from 1,022 as fast and accurately as possible. Each time a mistake is made, they are told to stop and start again.

As the TSST comprises the three key domains, specified by Dickerson and Kemeny (2004), the paradigm is associated with the largest effect sizes for cortisol responses observed under laboratory conditions (Dickerson & Kemeny, 2004). Social evaluation is embedded in each stage of the protocol: participants are notified that the panel members are specially trained to monitor non-verbal behaviour and that both the voice and video recordings of their performance will also be analysed. Additionally, participants are prompted to continue talking if their speech finishes before the 5-minute time limit and on each failure in the mental arithmetic task, a member of the panel says “Stop. Mistake, start over at 1,022, please” to instruct them to start again. The task is uncontrollable by nature as the participant is unfamiliar with the task and what will be asked of them. Additionally, participants are unaware of whether the assessing committee will be hostile or friendly and this in itself can remove perceptions of control of the situation.

A ‘friendly’ version of the TSST offers support for Dickerson and Kemeny’s suggestion that social evaluation plays an important role in the activation of the HPA axis. This variation of the original task maintains all the components of the TSST, but removes the social evaluative threat from the paradigm, replacing it with the presence of a friendly and supportive committee (Wiemers, Schoofs & Wolf, 2013). No significant cortisol responses were

observed when exposed to the friendly version, demonstrating that the missing critical social evaluation domain of the paradigm is the key trigger for eliciting a cortisol response. Similarly, a study examining the possibility that mere social presence of another may suffice in eliciting a cortisol response found no significant cortisol responses (Dickerson, Mycek & Zaldivar, 2008). These findings suggest that it is when faced with a negative social evaluative panel specifically, and not the mere presence of another, or of a friendly panel, that a cortisol response may be observed.

Since its inception, variations of the TSST have been trialled in the laboratory: for example, a group version of the stressor (The TSST-G; Von Dawans, Kirschbaum & Heinrichs, 2011). As with the original TSST, the TSST-G involves participants undergoing a preparation phase, whereby they are notified of the subsequent challenging task (delivering a free speech for 2 minutes to a selection panel for a job vacancy) and are informed of the presence of a video camera, and again, that the panel are experts in assessing non-verbal behaviour. The variation, however, lies in the panel session itself, whereby instead of standing in front of the panel alone, 6 subjects face the assessors at once, and deliver their speech and mental arithmetic tasks in turn. The TSST-G produced more than a 3-fold rise in cortisol and a significant increase in heart rate compared with the control condition, whereby the social evaluation and uncontrollability elements of the paradigm were absent from the protocol. As with the original TSST, subjective measures of psychological stress and anxiety revealed significantly more challenge, stress, and strain following cessation of the task, when compared to the control condition. However, a further study, also utilising

the TSST-G found that when shared social identity was encouraged among the participants (by delivering instructions to the group as a whole; providing group name tags; identical group t-shirts as well as other methods of inducing shared social identity), attenuated cortisol reactivity was observed, when compared with participants assigned to a personal identity group (provided with individual name tags and t-shirts etc.). This suggests that the perception that “we are in this together” increases positive appraisals and feelings of social support, compared with mere presence of one’s peers (Häusser, Kattenstroth, van Dick & Mojzisch, 2012). This indicates that the TSST-G only elicits a stress response when no group social identity is formed among participants.

A further variation of the TSST has been devised specifically for child subjects (TSST-C: Buske-Kirschbaum et al., 1997). The procedure is similar in terms of the number of assessors present, and the equipment involved in the protocol. However, instead of preparing a speech during the preparation period, child participants are asked to think of an exciting ending to a story, to present to the panel. The children are told to try and perform better than all the other children, encouraging competitive (thus, motivated) performance. In keeping with the social evaluative threat presented in the original TSST, child participants are asked to continue, should they stop talking before their 5 minutes is over. Following their story-telling, they are required to undertake a mental arithmetic task as fast as accurately as possible, being told to stop and start again if they make a mistake. The assessors in the TSST-C provide either positive/supportive or negative feedback to the participants, depending on the research question being investigated. The TSST-C, as with the TSST-G, elicits significant increases in

cortisol (Buske-Kirschbaum et al., 2007) though, interestingly, considerably smaller responses have been observed in children who received more positive and supportive feedback, compared with those who received negative feedback (Buske-Kirschbaum, 1997), further highlighting the effectiveness of social evaluation as a stressor component.

An additional variation of the TSST and TSST-C, coined the TSST-M (Yim, Quas, Cahill & Hayakawa, 2010) is a stressor protocol appropriate for both adults and children, devised in order to allow for more effective comparisons to be made across adult's and children's stress responses. The TSST-M includes a speech task, as with the TSST (speech) and the TSST-C (storytelling), however, the content is adjusted to make it identical for both adults and children. Participants are asked to talk about themselves, their personality and what makes them likeable, as well as describing a good and bad thing about themselves. After a 3-minute preparation period participants present their speech to the panel, and complete the mathematical task at the end. This modification also includes a slightly longer speech (6 minutes) and shorter mathematical task (4 minutes) in order to retain children's attention during the math task. In a study assessing responses in 9-12 year old children and 18-25 year old adults significant cortisol responses were observed following exposure to the TSST-M, demonstrating the potential of the paradigm to facilitate identification of individual difference factors across age groups which may pose as risk factors for stress-related illness at a later stage.

A similar, very novel stressor, The Bath Experimental Stress Test for Children (The BEST-C: Cheetham, & Turner-Cobb, 2016) encompasses similar

components to the TSST-C in order to evaluate stress responses in children. However, the uniqueness of this stressor lies in its use of age-matched children to form the panel using peer assessment, rather than using a panel of adult assessors, as has been the case in previous studies with children (e.g., The TSST-C: Buske & Kirschbaum, 1997). The authors suggest that this modification removes the ‘power dynamic’ which may be present in studies where children are being assessed by adults, a dynamic which is absent in adult testing (Cheetham & Turner-Cobb, 2016). Additional adjustments to the TSST protocol include parents assisting with part of the preparation period, a pre-recorded on-screen panel (rather than a live, present one), and following the 6 minute presentation and 4 minute mathematics task, participants were interviewed about the event. As observed in the TSST-C (and TSST-M), the BEST-C (Cheetham & Turner-Cobb, 2016) significantly increases levels of cortisol in participants, again, supporting the efficiency of social evaluation in eliciting a stress response, whilst considering ecological validity for child participants.

Although the TSST is renowned for its efficiency in eliciting stress responses greater than others, it is a relatively laborious and man-power intensive protocol compared to other stressors, which can make administration problematic when resources are sparse. In support of this, the greatest cortisol reactivity is typically observed in studies that adhere fully to the TSST protocol, demonstrating considerable sensitivity of the protocol, and the importance of strict adherence, in order to replicate these findings. The group version of the TSST (The TSST-G: Von Dawans et al., 2011) does reduce both the financial and labour

expenses of the original paradigm, although the resources required for this stressor are still more demanding than some of the other existing techniques.

An additional issue for consideration is the habituation properties of the TSST. A rapid habituation of HPA axis reactivity has been consistently observed for the TSST (Wüst et al., 2005), although uniform activation patterns of the sympathetic nervous system have been reported in response to the stressor (Mischler et al., 2005). These findings suggest that habituation to the TSST may apply only to HPA axis responses.

The stressors included in this review have been discussed in the context of their strengths and weaknesses; all have demonstrated reliable activation of at least one of the stress axes; however, perhaps the most common drawback shared by the majority of the aforementioned stressors relates to poor ecological validity. As stated earlier in the chapter, in order to truly assess an individual's stress response in the controlled conditions of the laboratory, the stressor needs to be as close to a real-life situation as possible (Wetherell et al., 2006). Stressors which are only capable of activating one system of the stress response (i.e. the SAM axis) are therefore of limited use to research investigating the stress response in its entirety.

Many of the stressors discussed to this point are limited because not only are the tasks far removed from day-to-day experiences, but the majority include undertaking just one challenging task at any one time. In everyday life, people are rarely faced with a single stressor and usually face a number of stressors at a time, and from various sources (Chida & Hamer, 2008; Wetherell et al., 2006).

Laboratory stressors that reflect these multiple outputs are, therefore, more representative of the challenges faced in everyday life (Wetherell & Carter, 2014).

As discussed in relation to the TSST, habituation of the psychobiological response is another issue found with many laboratory stressors, which is a consequence of the stressor becoming familiar and, therefore, less challenging (Kudielka & Wüst, 2010). This reduces the use of the stressor, particularly for protocols that require repeated administration with the same individual. However, the Multitasking Framework (MTF: Purple Research Solutions) provides a tool which addresses these issues.

As discussed in Chapter 2, the MTF is a platform for the presentation of performance-driven and cognitively demanding tasks. The framework comprises eight tasks, each of which can be presented singularly or in combination, dependent on the research question. A maximum of four tasks can be displayed at one time and each task occupies a quadrant of the screen. Levels of workload stress are manipulated either by increasing the number of tasks the participant must attend to (1-4), or by altering the difficulty of the tasks (low, medium or high intensity).

The tasks require a number of cognitive processes including perceptual, attentional, psychomotor, and memory abilities, which are typically employed in everyday functioning. All tasks are performance related, with points awarded for correct responses, and deducted for incorrect or missed responses, and a running total score is presented in the middle of the screen. The framework therefore includes two of the three domains recommended by Dickerson and Kemeny's

(2004) review: motivated performance, and uncontrollability, as well as cognitive demand.

The MTF has consistently demonstrated reliable stress-inducing capabilities, with significant increases in heart rate, systolic and diastolic blood pressure being observed following exposure to the stressor (Wetherell & Carter, 2014), demonstrating activation of the SAM axis. With regard to HPA axis activation, one study reported a significant increase in salivary cortisol in response to the MTF (Scholey et al., 2009), which, although suggests activation of the HPA axis, is a finding which should be interpreted with caution due to possible methodological issues related to time of sampling. Thus, to date, evidence of the HPA axis activating capabilities of the MTF is inconclusive. In terms of psychological responses, the MTF has been found to increase alertness, and decrease feelings of calmness, in addition to increasing reports of perceived workload (Wetherell & Carter, 2014). Participants exposed to the MTF also report more stress, state anxiety and lower contentment following stressor cessation (Scholey et al., 2009).

In addition to clear demonstration of the MTF's capabilities in triggering SAM axis activation, the stressor does not suffer the disadvantages that many of the aforementioned stressors often face. For example, the MTF possesses greater ecological validity than most laboratory stressors, as it includes multiple stressors from different sources (i.e. sounds and visual sources). The framework is additionally not subject to rapid habituation, a common limitation found in many other paradigms. This is for two reasons: firstly, as the stressor draws from 8 tasks, there are a number of possible variations to present to participants, which

therefore means that in stress research, not concerned with specific effects of cognitive tasks, the framework can be administered repeatedly for the same individual (Scholey et al., 2009); secondly, as the Framework is performance driven, participants create their own level of workload stress by striving to achieve as high a score as they can.

The present literature review has provided an introduction and discussion of stressor paradigms developed for the investigation of the human stress response under controlled, laboratory conditions. Whilst a number of these tools have successfully elicited stress responses, the response patterns in which these responses present themselves vary considerably, depending on the stressor components employed in each paradigm. Physical stressors, although often successful in eliciting cortisol responses, can present unnecessary risk to participants (e.g., ITT: Fraser, Albright & Smith, 1941), and often lack ecological validity (e.g., CO<sup>2</sup> inhalation: Wetherell et al., 2006); mental arithmetic stressors are easily administered, and elicit SAM axis reactivity, yet when presented in isolation (i.e. not as part of a paradigm including other components), lack the properties to trigger activation of the HPA axis (e.g., Noto et al., 2005). However, psychosocial stressors encompassing social evaluation, uncontrollability, and cognitive demand consistently evoke robust psychobiological responses (e.g., The TSST: Kirschbaum et al., 1993), and are considered the most successful stressor paradigms (Dickerson & Kemeny, 2004). These stressors are, however, often more laborious and thus less economical than many of the alternatives discussed in this review, and this is an issue which is addressed in the following chapter.

## **Chapter 4**

Study 1: The development of a tool for examining the acute psychobiological stress response in a laboratory setting

The findings of the present study were disseminated at the Midlands Health Psychology Annual Conference 2014.

## Introduction

Chapter 3 provided a review of stressor paradigms which have thus far been utilised as tools for the assessment of the stress response under laboratory conditions. As highlighted previously, the patterns of reactivity observed in response to the varying paradigms are dependent on the nature of the stressor itself (Miller, Chen & Zhou, 2007), with some stressors eliciting only SAM axis activation (e.g., mental arithmetic: Noto et al., 2005) whilst others reliably elicit both SAM and HPA axis reactivity (e.g., TSST: Kirschbaum et al., 1993). Psychosocial stressors have consistently elicited the most robust responses (Dickerson & Kemeny, 2004), yet existing paradigms are often also prone to disadvantages such as being labour-intensive, and therefore expensive, making them less accessible than many other paradigms. Another disadvantage of existing stressors such as the TSST is that they often lack ecological validity, making the global interpretation and generalisability of responses observed under these conditions, limited. Moreover, despite strong reliability in triggering HPA axis reactivity, the TSST does not withstand habituation effects, and this, again, limits the use of the paradigm. Therefore, there is a requirement for the development of a novel paradigm, which not only includes components of social evaluation, uncontrollability and cognitive demand, but one which is also ecologically valid, and economical to deliver. Furthermore, as stressors which only activate one stress system are of limited use in stress research, the developed paradigm should elicit both SAM and HPA axes reactivity, and should be resistant to habituation.

As discussed in the previous chapter, the MTF's stress inducing capabilities, ease of administration, ecological validity, and apparent resilience to

habituation make it a strong candidate for further investigation. For the purposes of the first study of this research programme, the Multitasking Framework will therefore act as a platform, expanded upon by the inclusion of critical social evaluation to form a novel, acute laboratory stressor paradigm. The proposed stressor therefore encompasses all the recommended components outlined by Dickerson and Kemeny (2004), whilst increasing ecological validity, thus creating a new tool for the investigation of acute stress in the laboratory.

## **Aims**

One aim of the present study was to develop an acute laboratory stressor, which would address the methodological shortcomings associated with a number of existing paradigms, namely those of ecological validity, ease of administration (including financial and labour-related resources), and habituation. An additional aim was to develop a stressor paradigm which would reliably elicit activation of the HPA axis, demonstrated through cortisol secretion in response to exposure to the proposed model. Finally, the developed stressor is required to be sufficiently salient that participants are thinking about the requirements of the upcoming task in the period preceding the event.

## **Hypotheses**

It was hypothesised that combining a novel, direct critical social evaluation component to the cognitively demanding multitasking framework would result in a stressor paradigm capable of eliciting robust stress responses, including activation of the SAM and HPA axes and psychological stress reactivity. It was hypothesised that individuals exposed to the direct critical social evaluation condition would elicit a stronger response to the stressor than those in the indirect critical social evaluation condition.

## Method

This section provides methods specific to the present study only. For details regarding the self-report measures, the stressor paradigm, and saliva sample collection please refer to Chapter 2.

### *Participants*

The sample comprised 39 healthy adults (5 males and 34 females), ranging between 18 and 37 years of age ( $M_{\text{age}} = 22.0$ ,  $SD_{\text{age}} = 4.62$ ). Participants were recruited from the undergraduate student population at a university in the North East of England and were randomly allocated to either the direct critical social evaluation condition (20 in total: 3 males, 17 females,  $M_{\text{age}} = 21.9$ ,  $SD_{\text{age}} = 4.36$ ) or the indirect critical social evaluation condition (19 in total: 2 males, 17 females,  $M_{\text{age}} = 22.1$ ,  $SD_{\text{age}} = 5.02$ ). There were no significant differences between the two groups in terms of age, perceived stress or trait anxiety (see Table 4.1).

Volunteers were screened for the eligibility criteria, including being aged between 18-40 years, and confirmation of the following: resting blood pressure which did not exceed 140/90; not currently taking steroidal medication; not pregnant or currently breastfeeding; no history of panic attacks.

Ethical approval for the study procedure was obtained from the Ethics Committee of the Health and Life Sciences Department at Northumbria University.

**Table 4.1** Demographic information for the direct critical social evaluation and indirect critical social evaluation conditions.

	<i>Direct critical social evaluation n=20</i>	<i>Indirect critical social evaluation n=19</i>	<i>p</i>
<i>Age (years)</i>	23.47 (4.15)	23.42 (4.99)	<i>n.s.</i>
<i>Perceived stress</i>	16.20 (6.60)	17.94 (7.83)	<i>n.s.</i>
<i>Trait anxiety</i>	26.05 (10.52)	25.63 (10.11)	<i>n.s.</i>

### *Materials*

#### *The Multitasking Framework*

The Multitasking Framework (see Chapter 2) formed the basis of the stressor paradigm. In addition, social evaluative threat was implemented by the experimenter providing negative feedback throughout the task. Participants were reminded at specific intervals (see Table 4.2) that they were required to work as fast and accurately as possible, to increase the current speed at which they were working and that their score was low and below-average.

#### *Morning mood*

Participants were required to complete a questionnaire on the morning of the laboratory stressor session, which asked questions regarding feelings towards the forthcoming session, as well as their sleep/wake times (see Chapter 2 for scale details).

**Table 4.2** Script and procedure used for both direct critical social evaluation and indirect critical social evaluation conditions.

<i>Time (minutes)</i>	<i>Evaluative comment</i>
+4	“Remember you must be as fast & accurate as you can on all of the tasks”
+8	“Your score is on the low side, you should probably speed up”
+12	“You should really be working faster than this”
+16	“Your score is still below the average”
+18	“You only have 2 minutes remaining and you must get as high a score as you can”

*Procedure*

Participants satisfying the eligibility criteria were invited to provide informed consent. All testing took place at least 1 hour following awakening and between 1200 and 1600, when levels of cortisol are typically lower and more stable (Saxbe, 2008). Participants were instructed not to consume any food or drink (other than water), smoke, or brush their teeth within 30 minutes of attending the laboratory for testing.

On their first visit to the laboratory, which was a minimum of two days and maximum of seven days prior to the test session, participants attended a 5-minute study brief. During this appointment they were notified of what the stressor session would entail: direct critical social evaluation participants were notified that they would stand in front of the researcher and receive critical social evaluation of their behaviour and performance throughout a multitasking task; those in the indirect critical social evaluation condition were told that they would complete the task whilst seated, with the researcher sat behind them, providing social evaluation and assessing their behaviour and performance over their shoulder. Participants were provided with a questionnaire booklet (see Chapter 2), as well as a state mood questionnaire and a sleep quality diary to take away and complete on the morning of the stressor session.

All participants agreed to provide the researcher with their phone number in order to receive a text message on the morning of their test session to remind them of their appointment, and to complete the morning questionnaire.

On the morning of the stressor session, prior to attending, participants were required to complete a morning mood questionnaire. On arrival at the laboratory on the day of the stressor session, participants were seated and the DYNAMAP cuff was placed on their non-dominant arm. They then provided their first saliva sample and completed their first mood questionnaire. After a seated rest period of 10 minutes, to establish a clear baseline (Balodis Wynne-Edwards & Olmstead, 2010), heart rate (HR) and blood pressure (BP) measurements were recorded and participants were given a 2-minute demonstration of the tasks by the researcher, in line with previous uses of the framework (Wetherell & Carter,

2014). Participants were informed that they needed to work as fast and accurately as possible, in order to achieve as high score as they could. They were additionally reminded that they were required to engage with all four of the tasks, and that failure to do so would result in the removal of their data from analysis.

After the demonstration, the second saliva sample was collected, the second HR and BP readings were recorded, and participants completed their second state mood questionnaire. The MTF was reconfigured and once all aforementioned measures were completed, the stressor task commenced.

The cuff remained on the participant's arm for the duration of the laboratory session (1 hour), enabling measures of HR and BP to be obtained at 5-minute intervals during the stressor task. This protocol was applied for both conditions, however, in the indirect critical social evaluation condition, participants were seated at a table, with their back to the researcher. The researcher sat behind them, providing critical social evaluation and taking HR and BP measurements throughout the task. In the direct critical social evaluation condition participants stood behind a podium, facing the researcher who was seated at a desk in front of them, with the computerised task screen projected onto the wall behind the participant (and in front of the researcher). This allowed the researcher to monitor the participant's performance throughout the session. The stressor task lasted 20 minutes. Upon cessation of the stressor the third saliva sample was collected, as well as HR and BP measures, taken immediately (whilst the participant was still standing, in the case of the direct critical social evaluation condition). Participants then completed a third state mood questionnaire. After the stressor, participants were left alone in the room for a 20-minute relaxation period.

During this time participants watched a nature documentary, material similar to that used for relaxation purposes in previous studies (e.g., Evans, Greaves-Lord, Euser, Franken & Huizink, 2012). After 10 minutes, the researcher briefly re-entered the room to collect a fourth saliva sample, state mood questionnaire, and to measure HR and BP. After a further 10 minutes (at the end of the nature documentary) a final saliva sample and state mood questionnaire were completed, and final HR and BP measures were obtained. The completion of these measures marked the end of the study. The procedure lasted for a duration of one hour and the full protocol is outlined in Table 4.3. See Figure 4.1 for visual representation of study set up for both the direct and indirect critical social evaluation condition

**Table 4.3** Study 1 procedure. The time (minutes) of each procedure prior/subsequent to the stress task is displayed in the left column.

<i>Time (minutes)</i>	<i>Procedure</i>
-10	Saliva sample 1
	State mood 1
	Heart rate (HR) and blood pressure (BP) measured, following rest period
-5	Demonstration of the MTF (2 minutes)
-3	Saliva sample 2
	State mood 2
	HR and BP
0	Stressor commencement
	HR and BP
5	HR and BP
10	HR and BP
15	HR and BP
20	Stressor cessation
	HR and BP
	Saliva sample 3
	State mood 3
40	Nature documentary commencement
50	Saliva sample 4
	HR and BP
	State mood 4

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*Treatment of data*

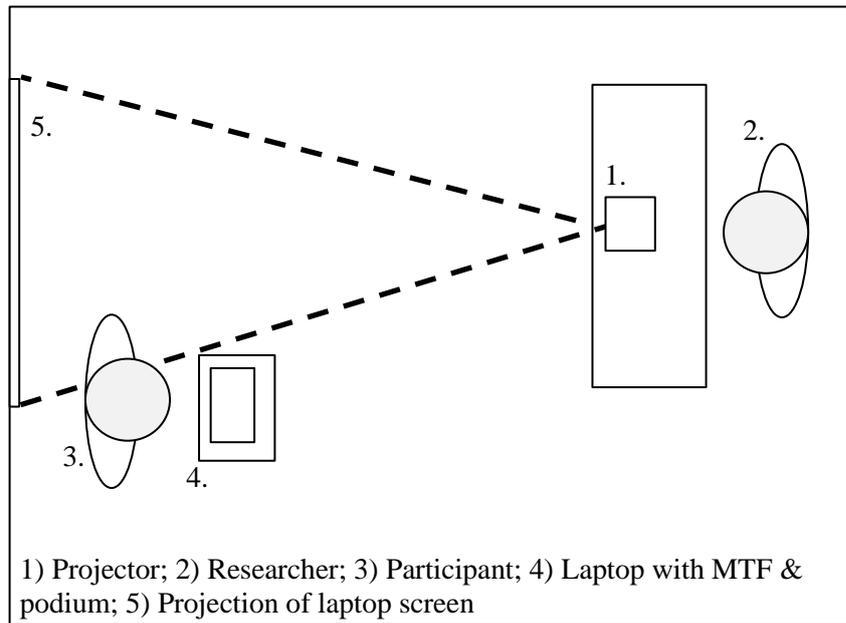
Cortisol levels were investigated using a mixed two-way analysis of variance (ANOVA) with condition (direct critical social evaluation and indirect critical social evaluation) and time (arrival, post-demonstration, stressor cessation, 10 minutes post-stressor, and 20 minutes post-stressor).

Data for cardiovascular parameters (HR, SBP, and DBP) were analysed using mixed two-way ANOVAs with condition (direct critical social evaluation and indirect critical social evaluation) and time (arrival +5 minutes, post-demonstration, stressor cessation, 10 minutes post-stressor, and 20 minutes post-stressor).

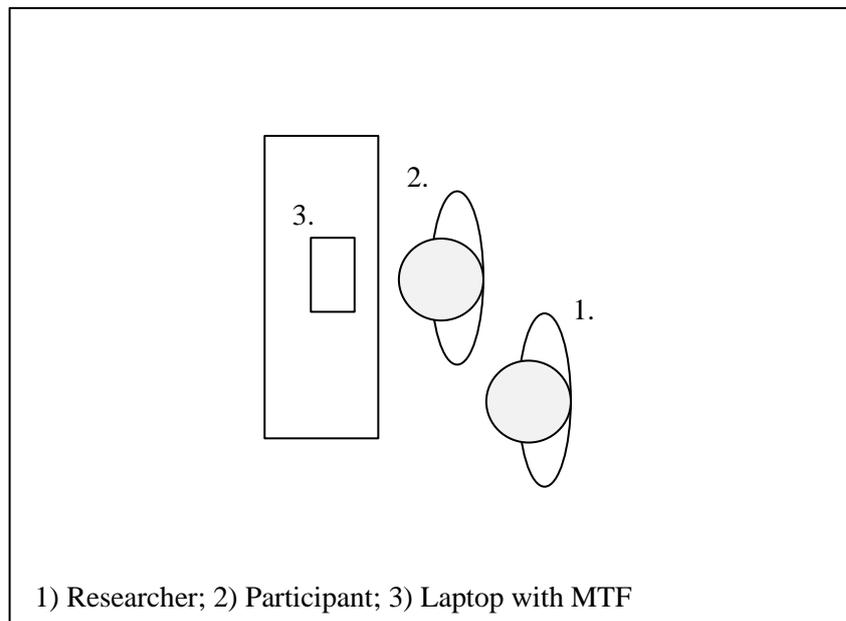
Data for state anxiety were also analysed using a mixed two-way ANOVA with condition (critical social evaluation or no critical social evaluation) and time (arrival, post-demonstration, stressor cessation, 10 minutes post-stressor, and 20 minutes post-stressor).

Significant main effects were investigated using Bonferroni adjusted pairwise comparisons. Significant interaction effects were followed up using Bonferroni adjusted *t*-tests. All analyses were conducted using SPSS 22.

A)



B)



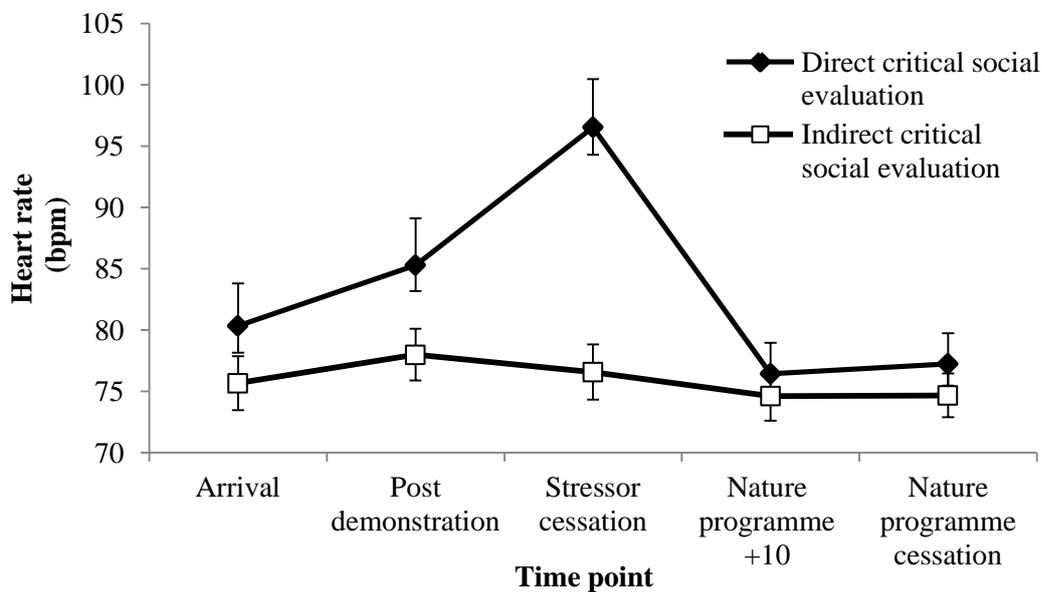
**Figure 4.1** The stressor paradigm for A) direct critical social evaluation (CSE) and B) indirect CSE.

## Results

### *Cardiovascular parameters*

#### *Heart rate*

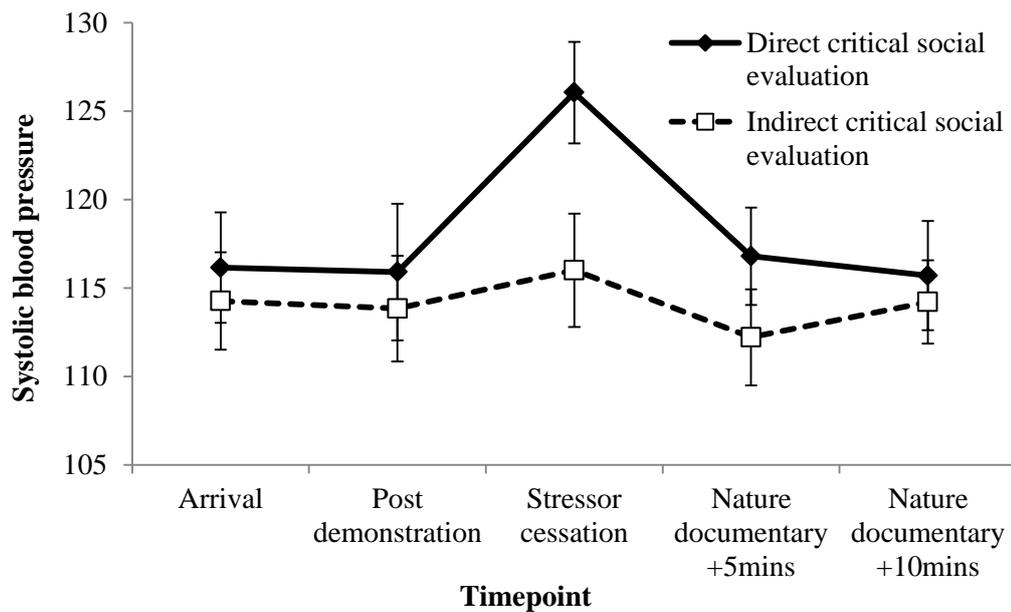
There was a significant main effect of time, [ $F(4,34) = 13.56, p < .001$ , Wilks'  $\Lambda = .39$ ], with a large effect size (partial  $\eta^2 = .66$ ). There was additionally a significant main effect of condition, [ $F(1, 37) = 4.29, p = .045$ , partial  $\eta^2 = .10$ ], and a significant time x condition interaction, [ $F(4,34) = 8.95, p < .001$ , Wilks'  $\Lambda = .49$ ], with a large effect size (partial  $\eta^2 = .51$ ). Following a one-way ANOVA, it was established that cessation of the stressor was the point at which the groups significantly differed, [ $F(1,37) = 10.24, p = .003$ ]. Mean heart rate readings for both groups are presented in Figure 4.2.



**Figure 4.2** Mean (and SE) heart rate for both direct critical social evaluation and indirect critical evaluation conditions, assessed during exposure to the stressor manipulation ( $n = 39$ ).

### *Systolic blood pressure*

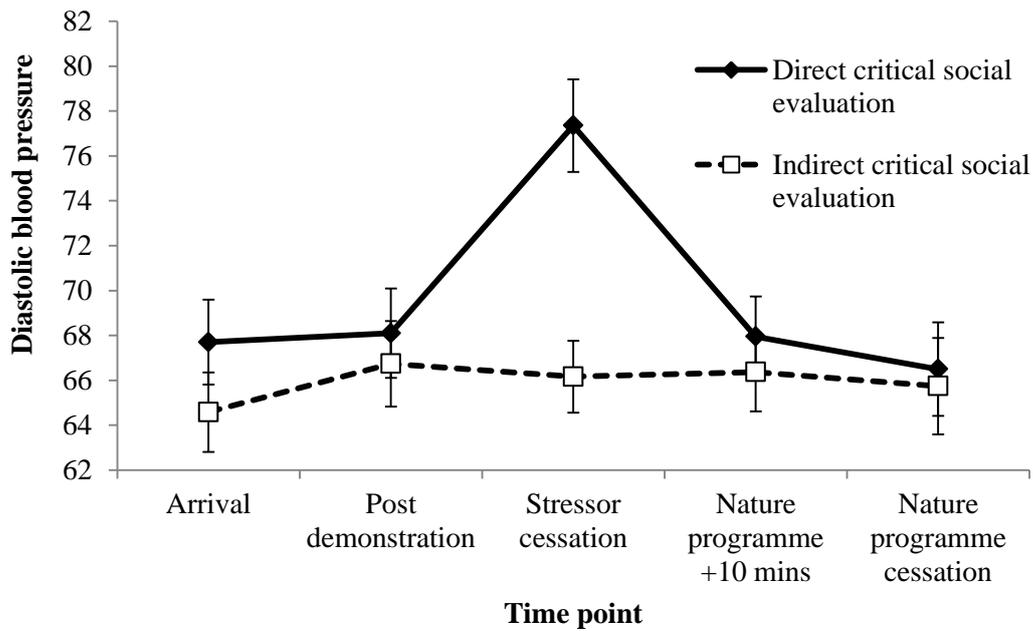
A significant main effect of time was observed, [F (4,34) = 3.27,  $p = .023$ , Wilks'  $\Lambda = .72$ ], with a large effect size (partial  $\eta^2 = .28$ ). However, no significant main effect of condition was found, [F (1,37) = 1.166,  $p = .287$ , partial  $\eta^2 = .03$ ]. There was no interaction effect between time x condition, [F (4,35) = 1.29,  $p = .295$ , Wilks'  $\Lambda = .87$ , partial  $\eta^2 = .13$ ]. Follow up analyses revealed that the only significant differences were observed at time point T7 (cessation of stressor) [F (1,37) = 5.49,  $p = .025$ ]. Mean SBP readings for both groups are presented in Figure 4.3.



**Figure 4.3** Mean (and SE) SBP for the direct critical social evaluation and indirect critical evaluation conditions, assessed during exposure to the stressor manipulation (n = 39).

### Diastolic blood pressure

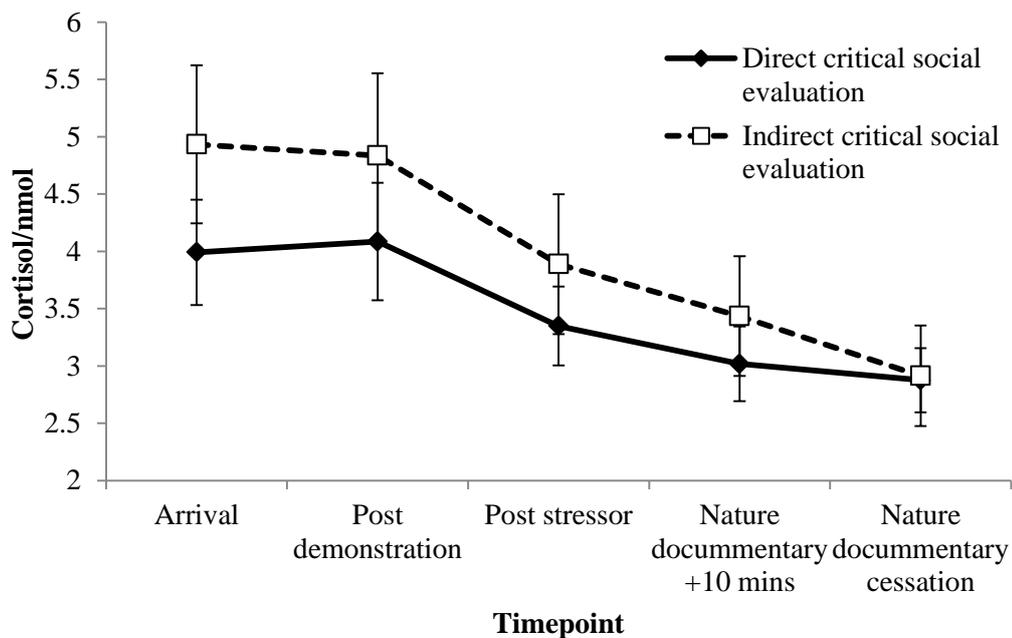
A significant main effect of time was observed, [F (4,34) = 5.45,  $p = .002$ , Wilks'  $\Lambda = .61$ ], with a large effect size (partial  $\eta^2 = .39$ ). There was no significant main effect of condition, [F (1,37) = 2.63,  $p = .113$ , partial  $\eta^2 = .07$ ]. There was, however, a significant time x condition interaction, [F (4,34) = 4.66,  $p = .004$ , Wilks'  $\Lambda = .65$ ], with a large effect size (partial  $\eta^2 = .35$ ). Follow up analyses revealed that the only time point at which the groups significantly differed was at cessation of the stressor, [F (1,37) = 18.17,  $p < .001$ ]. Mean DBP readings for both groups are presented in Figure 4.4.



**Figure 4.4** Mean (and SE) DBP for the direct critical social evaluation and indirect critical evaluation conditions, assessed during exposure to the stressor manipulation (n = 39).

## Cortisol

A significant main effect of time was observed, [ $F(4,29) = 6.35, p = .001$ , Wilks'  $\Lambda = .53$ ], with a large effect size (partial  $\eta^2 = .47$ ). However, changes in cortisol levels followed the typical diurnal decline, observed throughout the day, therefore no cortisol reactivity to the stressor was observed. There was no significant main effect of condition, [ $F(1,32) = .41, p = .527$ , partial  $\eta^2 = .01$ ]. Further, no significant time x condition interaction was observed, [ $F(4,29) = 1.81, p = .154$ , Wilks'  $\Lambda = .80$ , partial  $\eta^2 = .02$ ]. Mean cortisol levels for both groups are presented in Figure 4.5.



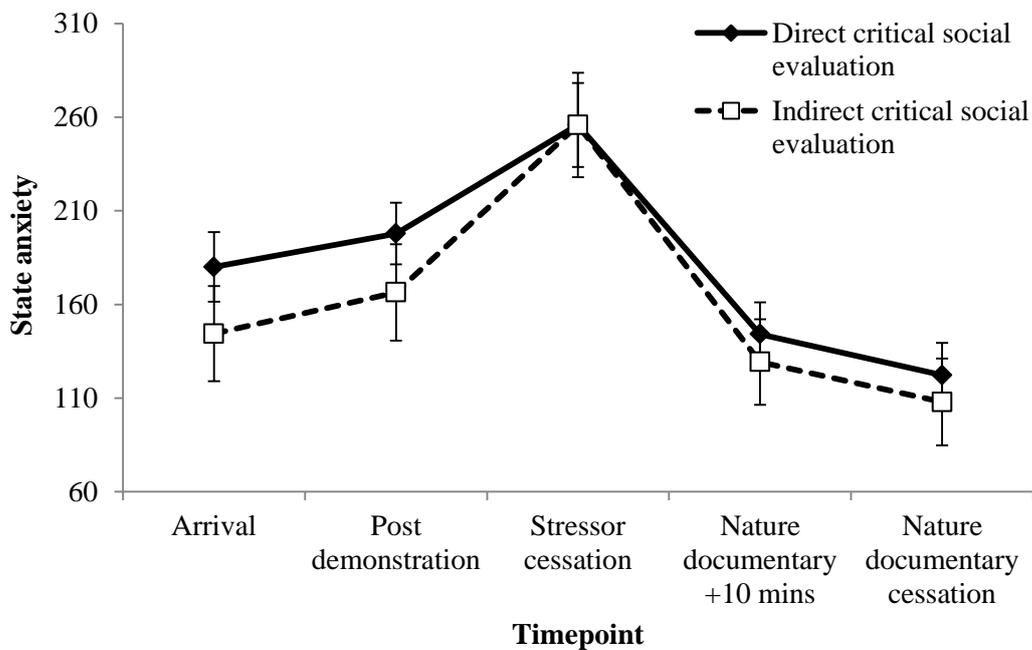
**Figure 4.5** Mean (and SE) cortisol levels for the direct critical social evaluation and indirect critical evaluation conditions, assessed during exposure to the stressor manipulation ( $n = 39$ ).

Mean physiological responses to the stressor paradigm, for both the direct and indirect critical social evaluation groups are presented in Appendix A.

*Psychological measures*

*State anxiety*

There was a significant main effect of time, [F (4,34) = 27.36,  $p < .001$ , Wilks'  $\Lambda = .24$ ], with a considerably large effect size (partial  $\eta^2 = .76$ ). However, there was no significant main effect of condition, [F (1,37) = .50,  $p = .485$ , partial  $\eta^2 = .01$ ]. No time x condition interaction was observed, [F (4,34) = .50,  $p = .739$ , Wilks'  $\Lambda = .95$ , partial  $\eta^2 = .06$ ]. Mean state anxiety scores for both groups are presented in Figure 4.6. Mean psychological responses are reported in Appendix B.



**Figure 4.6** Mean (and SE) state anxiety for the direct critical social evaluation and indirect critical evaluation conditions, assessed during exposure to the stressor manipulation (n = 39).

*Awakening mood on day of stressor*

As indicated in Table 4.4, participants in the direct critical social evaluation condition reported significantly greater feelings of tension than those exposed to indirect critical social evaluation, [F (1,37) = 6.12, p = .018, partial  $\eta^2$  = .14]. They additionally reported feeling less content, [F (1,37) = 5.79, p = .021, partial  $\eta^2$  = .17] less calm, [F (1,37) = 8.40, p = .006, partial  $\eta^2$  = .18] and less happy, [F (1,37) = 6.54, p = .015, partial  $\eta^2$  = .15] than their control group counterparts.

**Table 4.4** Mean (and SD) awakening mood reported on the morning of the stressor for both direct and indirect critical social evaluation groups.

<i>Awakening mood</i>	<i>Direct critical social evaluation (n=20)</i>	<i>Indirect critical evaluation (n= 19)</i>
Calm	50.15(24.90)	71.89(21.77)
Content	46.20(26.52)	63.84(22.96)
Happy	50.45(26.22)	69.89(20.81)
Tense	45.35(21.63)	27.58(23.22)

## **Discussion**

This study aimed to investigate the stress-inducing capabilities of a laboratory stressor, which incorporated combined components demonstrated in previous literature, to elicit a robust stress response. This was investigated by assessing both physiological and psychological responses to the novel stressor paradigm.

Participants were told in advance that they would be critically socially evaluated whilst undertaking a cognitively demanding multitasking exercise. During the task, participants either a) stood facing the researcher while the researcher sat behind a desk, observing them and providing feedback on their performance (direct critical social evaluation), or b) sat at a desk for the duration of the task, with the researcher observing them and providing feedback on their performance from behind (indirect critical social evaluation). Both groups received identical feedback, at the same intervals, and salivary cortisol, heart rate, blood pressure and subjective mood were measured throughout the laboratory session.

As hypothesised, direct critical social evaluation elicited a stronger cardiovascular response, with significantly greater heart rate observed in participants exposed to direct, compared with indirect, critical social evaluation. This is consistent with previous literature investigating cardiovascular responses to stressors using these components (e.g., TSST: Allen, Kennedy, Cryan, Dinan & Clarke, 2014) and demonstrates that facing the researcher while receiving

feedback was considered more physiologically arousing than receiving the same feedback with one's back to the researcher.

The absence of significant group differences in diastolic and systolic blood pressure reactivity to the stressor is a finding which, despite the general trend of the results suggesting elevations in both these parameters upon stressor cessation, is consistent with previous literature; demonstrating that differing cardiovascular response patterns are observed in response to different types of stressors, reflecting the variety of underlying mechanisms which are involved in physiological functioning (e.g., Allen et al., 1987; Willemsen et al., 1998). However, whilst non-significant, large effect sizes observed across these analyses indicate that the non significant effects observed in the present study may be due a lack of power in the sample, with relatively modest sample sizes in both groups. Therefore replication work should attempt to explore this effect in greater depth in a larger population.

Direct critical social evaluation did not increase levels of state anxiety to a greater extent than indirect evaluation; moreover, both conditions led to significant increases in anxiety. Possible suggestions for this are discussed shortly, in relation to possible explanations for the reported cortisol findings. However, despite there being no differences in anxiety during the test session in the laboratory, direct critical social evaluation did influence psychological indices on scales completed on the morning that participants attended the laboratory for the stressor session: interestingly, those who faced forthcoming direct critical social evaluation reported feeling more tense on the morning of the stressor. They additionally reported feeling less content, less calm, and less happy than

individuals who were expecting indirect critical social evaluation. This suggests that they were anticipating a more demanding forthcoming event, and this finding is consistent with previous literature investigating anticipation of a similar socially evaluative laboratory stressor, whereby participants report greater levels of tension, perceived stress, and anxiety on the morning of the planned stressor (Wetherell, Lovell & Smith, 2014).

In contrast to the observed differences between groups in terms of their cardiovascular and psychological responses, hypothesised cortisol increases in response to the stressor were not observed. This was an unexpected finding, as a large body of literature has demonstrated that the components implemented in this stressor paradigm (uncontrollability, motivated performance and social evaluative threat; Dickerson & Kemeny, 2004) reliably elicit HPA axis activation, especially when combined (e.g., Dickerson & Kemeny, 2004).

Elevated cortisol levels are generally detectable after approximately 7 minutes, with levels peaking approximately 15 minutes post stimulus onset (McCann et al., 1993). The stressor used in the current study lasted for a duration of 20 minutes, with saliva samples collected for a further 20 minutes following stressor cessation. Therefore it can be inferred that these findings are not due to protocol-related issues with response detection, but that there may be possible issues with the components of the stressor itself, which have led to an absence of cortisol reactivity.

There are a number of possible explanations for the absence of cortisol reactivity in the present study: the TSST requires participants to stand in front of a panel of assessors and present a free speech after just 5 minutes of preparation,

whereby they need to convince the panel that they are the ideal candidate for a job. This paradigm reliably elicits a large cortisol response (e.g., Kirschbaum, Pirke & Hellhammer, 1993; Allen et al., 2014). During the development of the current paradigm, it was hypothesised that the physiological responses induced by the TSST were due to the full-frontal critical social evaluation domain of the stressor (i.e. standing in front of a panel of assessors whilst completing a task), and this theory guided the inclusion of this method of social evaluation in the current study. However, in light of the present findings, a plausible suggestion could be that it is not merely direct critical social evaluation, that is the key component required to elicit a cortisol response, but it may well be that it is public speaking in front of a panel itself, that is a key activator: if the Social Self Preservation Theory (Dickerson, Gruenewald & Kemeny, 2004) is considered this seems like a very plausible explanation. The theory states that threats to self-esteem, and the respect and acceptance of others alone can activate the HPA axis (Dickerson & Kemeny, 2004). Situations in which individuals' valued attributes are displayed to others, where there is an opportunity for potential social rejection or disapproval if one demonstrates a lack of these valued qualities, are threatening to our social self (Dickerson & Kemeny, 2004). In the current study design, participants stood facing the experimenter, while they completed the cognitive task (no speaking was involved on their part), and the task screen was projected onto the wall behind them. It was hypothesised that this set up, involving full-frontal critical social evaluation of performance would provide sufficient social evaluative threat to elicit such a response. However, it seems likely that the current design, although eliciting a stress response (indicated through

cardiovascular reactivity), did not provide enough of a threat to individuals' 'social self' to activate the HPA axis. This could mean that participants may not have experienced changes in self-esteem and self-regulated emotion (e.g., shame and embarrassment) argued to be a crucial function in stressors attempting to elicit a cortisol response (Dickerson & Kemeny, 2004).

Public speaking is a well-documented stressor, causing cardiovascular activation, increases in ACTH, and redistribution of circulating leukocyte subpopulations (Elsenbruch et al., 2006). The explanations as to why this activity is so effective in eliciting such a strong physiological response often surround the idea that giving a speech presents the challenge of being observed and scrutinised by assessors, which, as outlined in Dickerson and Kemeny's (2004) review, can present a threat to the social self. The risk of social embarrassment and humiliation, causing damage to the social self/reputation, is thought to be a leading reason why public speaking especially, can cause distress in the majority of people (Garcia-Leal, Graeff & Del-Ben, 2014). In a recent study, children who engaged in a public speaking task, believing they were being observed by an audience via a live video link reported concerns about not wanting to "perform badly" or "look silly" in front of the panel when interviewed following the stressor (Cheetham & Turner-Cobb, 2016). These findings support the concept of the fundamental need of humans, as social animals, to be accepted by others (e.g., Baumeister & Leary, 1995), and that public speaking threatens this goal.

Studies have demonstrated that public speaking to an evaluative audience is important when seeking to observe activation of the HPA axis, as performing a speech in a room with an inattentive confederate who is merely in the field of

vision of the participant elicits no cortisol response (Dickerson et al., 2008). This has been further demonstrated by studies which have reduced the level of social evaluative threat during public speaking tasks. For example, the f-TSST, a ‘friendly’ version of the original TSST (Wiemers, Schoofs & Wolf, 2013), is a non-evaluative variation of the TSST, whereby participants are asked to prepare to talk to the panel about their career aspirations, hobbies, and favourite book, for 8 minutes. The committee members are friendly towards the participant: nodding and smiling to provide feelings of support, to ensure that they do not feel negatively judged. This variation, although still involving public speaking, does not activate the HPA axis, most probably due to its non-threatening nature.

It could be therefore be argued that public speaking, coupled with receiving negative/hostile feedback is specifically responsible for the hormonal responses observed following exposure to paradigms including such components, as it has been reported that exposure to the TSST mental arithmetic task, whilst standing in front of assessors, in isolation, does not elicit changes in cortisol levels (Biondi & Picardi, 1999). The argument that the paradigm evaluated in the present study may not have been sufficiently threatening to the social self (Dickerson & Kemeny, 2004) to elicit a response could also be extended to provide an explanation for the lack of differences found between groups for reported state anxiety. Whilst the combination of the multitasking exercise and critical social evaluation did increase state anxiety across both groups, direct critical social evaluation did not increase anxiety to a greater extent than indirect critical social evaluation.

However, the significant differences in morning mood on the day of the planned task do suggest that the knowledge of upcoming direct critical social evaluation was perceived to be more psychologically taxing than anticipating indirect critical social evaluation, with greater reports of negative mood in those anticipating direct critical social evaluation. Furthermore, previous studies have reported greater physiological responses during anticipation of a stressor than during the stressor itself (Birnbaum, 1964). These findings indicate that in addition to differences in selected acute psychobiological responses, appraisals of the forthcoming demand are also different between the two groups, with a profile suggestive of anticipated challenge.

With regard to modifications that could be applied to the present paradigm to stimulate a cortisol response, increasing social evaluation further by including a public speaking task, much like that used in the TSST (Kirschbaum et al., 1993) would be a sensible next step. This would, however, need to be considered with caution, as there are procedural advantages of the present paradigm which are sensitive to the existing stressor components: firstly, due to the requirement of only one researcher, and minimal equipment, the present stressor paradigm benefits from ease of administration and is less labour-intensive than many other stressors (including the TSST: Kirschbaum et al., 1993). In addition, as the ‘cognitively demanding’ component of the stressor holds the ability to modify and manipulate the tasks, with regards to the number, combination and duration of the tasks, the model is less prone to habituation, a predicament reported for some laboratory stressor paradigms (for review, see Grissom & Bhatnagar, 2009). Furthermore, the novel stressor may also be considered more ecologically valid

than a number of previous paradigms as it incorporates multitasking as the cognitively demanding component, which is experienced by the general population on a daily basis. Therefore, the inclusion of a speaking task would need to be carefully tailored in such a way as to maintain both these properties.

### *Conclusions*

The present study sought to develop and evaluate a novel stressor paradigm to assess acute stress responses under controlled laboratory conditions. The results indicate that the stressor is successful in eliciting a physiological stress response. Although the findings suggest that the completion of a cognitively demanding task, whilst being critically socially evaluated by a single researcher, alone, does not trigger HPA axis activation, possible explanations have been discussed. Furthermore the findings regarding anticipation of direct critical social evaluation suggest that in addition to its stress-inducing properties, the paradigm could be a suitable candidate for studies investigating the anticipatory period preceding a stressful event. We can therefore infer that direct critical social evaluation is perceived to be more mentally taxing, as reported on the morning of the task, compared with indirect critical social evaluation. The findings also suggest that the anticipatory stress response differs depending on environmental context and future research should therefore investigate this proposition in greater detail by investigating anticipatory physiological reactivity (e.g., cortisol, specifically the CAR) as well as more in-depth psychological profiles of individuals anticipating a forthcoming demand. Finally, the present findings confirm the feasibility of investigating the anticipatory stress response using this

novel paradigm and it will therefore be implemented for the next stage of the research programme.

## **Chapter 5**

Anticipation of forthcoming demand: A literature review

## Introduction

Stress is linked with psychological distress, whether global, or to a specific stressful event (Chand, 1997). Considerable work has demonstrated both the physiological and psychological arousal-inducing properties of a number of stressors. However, empirical evidence demonstrates that stress responses (observed through cortisol reactivity, for example) can be observed not only in response to direct exposure to a stressor, but also in anticipation of a stressful event (Engert et al., 2013). Further, it has been suggested that this process can prolong the activation of stress mechanisms designed for short-term arousal only.

Whilst it is widely accepted that prolonged activation of the HPA axis (i.e. as a result of chronic stress), leads to deleterious health outcomes, recent studies have indicated that the anticipation of stressful events may also pose as a useful assessment of healthy physiological functioning. For example, in a study assessing stress responses to a laboratory paradigm, chronically stressed caregivers, compared with age-matched low-stress controls, demonstrated significantly greater cortisol increases during the anticipatory period preceding the stressor, but not in peak cortisol levels during the stressor itself (Aschbacher et al., 2013). These findings demonstrate the potential importance of the anticipatory time period and the identification of biomarkers which may be associated with atypical reactivity of stress response mechanisms and future negative health outcomes, associated with over-activation of the stress response systems.

Studies examining the anticipatory response preceding forthcoming acute, naturalistic stressors suggest that this period is often perceived to be highly stressful, and sometimes deemed to be even more stressful than the stressful event

itself (e.g., Greco & Roger, 2003). For example, studies assessing anxiety in patients attending hospital for surgery have reported similar results; in a study of orthopedic surgery patients the most anxiety was observed the day before hospital admission, two days prior to the operation (Johnston, 1980). A further study assessing patients awaiting heart surgery concluded that uncertainty and fear were more stressful for patients awaiting heart surgery than the symptoms of the heart condition itself (Bengston, Herlitz, Karlsson, & Hjalmarson, 1996). Similarly, a qualitative study using focus group interviews with women awaiting diagnoses following an abnormal mammogram reported the waiting period as a type of 'limbo' whereby the women's lives were seriously disrupted with "panic attacks, insomnia, inability to concentrate at work, inability to plan, gastrointestinal upset, tearfulness and preoccupation of fears" (p.45, Thorne, Harris, Hislop, & Vestrup, 1999). Women awaiting an invasive breast biopsy, again, following an abnormal mammogram reported considerable distress during the waiting period between the appointments (Lebel et al., 2003). These findings were supported in a further qualitative study where women retrospectively reflected on their experience of cancer treatment, reporting the waiting period between the first suspicion of cancer and receiving a diagnosis to be a very stressful period, with many participants reporting this to be the most stressful part of the diagnosis-treatment-recovery process (Saegrov & Halding, 2004). Further medical-related evidence comes from a study assessing the waiting period between embryo transfer (when a fertilised embryo is transferred to the uterus) and the pregnancy test in women undergoing IVF treatment, which observed this period to be considerably stressful (Boivin & Takefman, 1995).

The findings that the anticipatory period leading up to medical procedures or results are perceived to be stressful are perhaps not surprising, due to the serious and potentially life-threatening consequences of these anticipated events. The critical element that these events have in common is the degree of uncontrollability involved in each of the stressors, which, as outlined in Dickerson and Kemeny's (2004) review, is a key characteristic of stress responsiveness. The uncontrollable element of stressors of this nature originates from the lack of control the individuals have over the outcome following the waiting period, and the uncertainty surrounding the situation can cause worry and rumination. As alluded to previously, these intrusive cognitions can elicit larger stress responses during the anticipatory period than when the individual is directly faced with the stressor (Birnbaum, 1964). In each of the described scenarios those waiting in anticipation are faced with uncertainty and lack of control over both the duration and the outcome of the waiting period, as well as the possible threat to survival, and whilst it seems clear that an anticipatory stress response does occur in the lead up to certain stressors, surprisingly little is known about the mechanisms that control the magnitude of physiological responses to the anticipation of upcoming events. One potential mechanism involved in the anticipation of events is the cortisol awakening response (CAR). As discussed earlier in this thesis (see Chapter 1), despite a relatively robust CAR in healthy individuals under normal circumstances, varied responses have been observed in those experiencing different types of persistent stress or challenge. For example, increased CARs have been observed in ongoing stressors such as low socioeconomic status (Wright & Steptoe, 2005); chronic stress and worrying (Schlotz, Hellhammer,

Schulz & Stone, 2004); work-related stress (Kunz-Ebrecht, Kirschbaum & Steptoe, 2004); social stress, and lack of social recognition (Wüst et al., 2000). Blunted or attenuated CARs, have been observed in post-traumatic stress disorder (Wessa, Rohleder, Kirschbaum & Flor, 2006); burnout (de Vente, Olf, Van Amsterdam, Kamphuis & Emmelkamp, 2003; Morgan, Cho, Hazlett, Coric & Morgan, 2002); chronic fatigue (Roberts, Wessely, Chalder, Papadopoulos & Cleare, 2004); and in caregivers of dementia patients (de Vugt et al., 2005).

More recently the CAR has also been demonstrated to be sensitive to day to day differences in activity in healthy individuals who are not experiencing chronic stress. For example, greater CAR responses have been reported on work days, characterised by increased demands, compared with non-work days that are associated with relatively fewer demands (Kunz-Ebrecht, Kirschbaum, Marmot & Steptoe, 2004). Significantly increased CARs have also been reported on days containing daily stressors, such as arguments and home-related overloads, compared to stressor-free days (Stawski, Cichy, Piazza & Almeida, 2013). The preparatory responses associated with stressors of this nature, together with evidence from studies observing modifications to the CAR in those reporting chronic stressors, have led to speculation of the specific functions of the CAR.

The observation that those with upcoming stressful events exhibit greater CARs on the morning of a stressor indicate that cortisol can increase, not only in direct response to a stressor (e.g., Dickerson & Kemeny, 2004), but also in the period preceding a challenging event (Engert, 2013; Rohleder et al., 2007). The specific role of the CAR remains relatively unknown, however, given that the CAR represents a cortisol increase at the beginning of the human activity phase

(Powell & Schlotz, 2012), the phenomenon may serve an adaptive role in the preparation of an individual for forthcoming demand within the upcoming day, referred to as the ‘anticipation hypothesis’.

### *The anticipation hypothesis*

The ‘anticipation hypothesis’ postulates that increased morning levels of cortisol may reflect a greater need for resources and energy to cope with the forthcoming challenges of the day ahead. For example, glucocorticoids enable a plethora of physiological processes, which facilitate a state of enhanced arousal (Schulz, Kirschbaum, Pruessner & Hellhammer, 1998), which is beneficial for upcoming tasks that require differing magnitudes of arousal. In support of this concept, a potential explanation for the links between the CAR and anticipation of the forthcoming day comes from evidence of a neuronal mechanism associated with awakening. That is, post-awakening increases in cortisol may be associated with the process of awakening via activation of memory representations of the self (Powell & Schlotz, 2012). This process enables orientation with time and space and, together, could elicit activation of the HPA axis (Wilhelm, Born, Kuehlka, Schlotz & Wüst, 2007). This theory has been supported by further suggestions that the CAR may elicit activation of prospective memory representations, enabling the individual to orientate themselves with anticipation of demands of the upcoming day (Fries, Dettenborn & Kirschbaum, 2009), although this particular topic area has not been widely studied through empirical testing.

Further support for the anticipation hypothesis comes from findings that the CAR response only appears to occur following rising from bed and commencing the daily routine, but not when the same individuals are woken

during the night (Dettenborn, Rosenloecher & Kirschbaum, 2007). This could indicate that the CAR only occurs in the face of upcoming demands (i.e. when getting up for the day), but not in cases when increased arousal is unnecessary (e.g., in the middle of the night).

### *Worry and rumination*

As well as the impact of anticipation of forthcoming demand on biomarkers of stress reactivity, repetitive negative thoughts (RNT), collectively including cognitive states such as worry and rumination, have also been associated with strong effects of elements of stress reactivity. Worry is defined as “a chain of thoughts and images, negatively affect-laden and relatively uncontrollable. The worry process represents an attempt to engage in mental problem-solving on an issue whose outcome is uncertain but contains the possibility of one or more negative outcomes. Consequently, worry relates closely to fear processes.” (p.10. Borkovec, Robinson, Pruzinsky & DePree, 1983).

Rumination, described as “the process of thinking perseveratively about one’s feelings and problems” (p.400. Nolen-Hoeksema, Wisco & Lyumbomirsky, 2008) refers to these thoughts post stressor, and refers to a series of thoughts and behaviours which lead individuals to focus on their emotions and renders them unable to focus on distracting activities which could relieve symptoms (Nolen-Hoeksema & Morrow, 1991). Previous studies have demonstrated that worry and rumination are highly correlated and are both associated with anxiety, with similar processes driving both constructs (Ehring & Watkins, 2008). Whilst a considerable amount of research has focused predominantly on the link between chronic rumination and maladaptive consequences such as depression and anxiety

(e.g., the response styles theory: Nolen- Hoeksema & Morrow, 1991) less is known about the effects of rumination following acute stress. Research suggests that persistent worrying and rumination prolongs stress-related physiological activation, mediating the well-established relationship between stress and disease (Brosschot, van Dijk & Thayer, 2002). As such, these processes have been coined ‘perseverative cognition’, referring to intrusive repetitive thoughts about past or future, anticipatory stressors.

Anticipation requires the ability to ruminate about events which could occur months or years from the present, and it has been argued that the cognitive preoccupation with upcoming tasks itself can pose as a cognitive stressor in its own right (Ennis, Kelly & Lambert, 2001; Schlotz et al., 2004) and, therefore, subsequently facilitate HPA axis activation (Roger & Najarian, 1998). In consideration of the evolutionary perspective, the ability to conceptualise and accurately anticipate potential challenges or threats is adaptive, as the negative affect associated with anticipating a stressful event allows for appropriate modifications to be made with regards to behaviour, cognition and physiology. This process not only means the individual is prepared for the forthcoming demand, but in some situations the forward-planning this requires, combined with the motivation to take measures to avoid the event, could even result in bypassing the threat altogether (Aspinwall & Taylor, 1997; Schulkin, 2011). However, despite the advantages of appropriate levels of anticipation, inappropriate anticipation or rumination (leading to excessive or prolonged responses) can lead to maladaptation of physiological mediators, as described earlier in this thesis (see Chapter 1). With regards to the psychological effects of anticipation, studies have

suggested that negative mood induction may amplify dysfunctional attitudes (Fries et al., 2009), demonstrating another example of the negative long-term implications of this prolonged activation.

#### *Anticipation of naturalistic stressors*

Naturalistic studies assessing cortisol profiles in individuals preparing for a particular challenging or stressful event have further added to the body of literature regarding anticipation of forthcoming demand: for example, in a sample of competitive ballroom dancers preparing for a competition, waking levels of cortisol were significantly greater on the competition day compared with control days (Rohleder et al., 2007). In line with previous studies observing greater CARs on ‘stressful’ days (e.g., Stawski et al., 2013), cortisol diurnal profiles also remained significantly higher over the course of the competition day compared with the control day profiles (Rohleder et al., 2007), demonstrating that the impact of the stressor demands also extends to effect diurnal secretion of cortisol.

Longitudinal data has demonstrated that medical graduates preparing to undertake a specialist medical exam, allowing them to be ‘ranked’, and essentially determining whether or not they would be accepted for specialist training, secreted greater levels of cortisol in the period leading up to the exam day, and on the exam day itself compared with the control days following the event (Gonzalez-Cabrera, Fernandez-Prada, Iribar-Ibabe & Peinado, 2014). Interestingly, whilst cortisol levels progressively increased during the preparatory period for the examination, the medical graduates additionally reported higher levels of anxiety and a progressive increase in self-perceived stress in the seven-month period leading up to the examination, compared with the days following

the exam. These findings strongly support the concept of anticipation of a stressful event prolonging the observed stress response.

Another naturalistic stressor paradigm which has been assessed with regards to anticipatory stress-inducing properties is competition. Whilst competition elicits robust stress responses (e.g., in footballers: Alix-Sy, Scanff & Filaire, 2008) the onset of the response to competitive situations is elicited before the competitive event commences and, as such, competition also provides a useful tool for assessing anticipatory stress responses (e.g., Alix-Sy et al., 2008; Filaire et al., 2009). These studies have observed increases in cortisol during the period directly leading up to the competition, for example; in a study assessing psychological and physiological reactivity during both practice and competition rounds in elite golfers, significantly greater cortisol, somatic anxiety and lower self-confidence were observed during the competition compared with the practice round (McKay, Selig, Carlson & Morris, 1997). In studies of Judo, competitors report increased anxiety and cortisol secretion in the lead up to a competition (Filaire, Sagnol, Ferrand & Maso, 2001; Salvador, Suay, Gonzalez-Bono & Serrano, 2003). Further, Filaire and colleagues (2001) observed significantly higher cortisol before an international competition compared with a national one, supporting the concept that the greater the intensity of the threat (in this instance, to the social-self), the greater the cortisol response (Dickerson & Kemeny, 2004). Judo is a particularly useful competitive sport to assess anticipatory processes as Judo fights sit below the threshold that elicits exercise related HPA responses, typical of other sports which involve short duration spurts of exercise (Filaire et al., 2001).

In a recent review of cortisol and competition (Casto & Edwards, 2016) higher levels of cortisol were reported on competition days prior to the warm up compared with time-matched samples collected on neutral non-competition days. This pattern has been observed across a number of sports, both those in which exercise-induced HPA axis activation is a possibility (e.g., football) and those where the physical demands of the activity are less likely to evoke an exercise-induced HPA response (e.g., Judo), suggesting a genuine effect of stress, and not merely exercise related induction of this response. Conversely, competition also appears to allow for vicarious experiences of anticipation in those invested in the event. For example, one study demonstrated that team coaches demonstrate cortisol increases throughout the competition period (Kugler, Reinjes, Tewes & Schedlowski, 1996) although for players themselves, it seems that participation is crucial, with no observed cortisol responses in players watching from the bench whilst their team competes (Edwards & Casto, 2013).

As highlighted above, naturalistic studies can provide useful knowledge regarding anticipation of everyday demands and associations with the CAR; furthermore, these stressors often encompass the prospect of one's valued attributes being evaluated and critiqued (e.g., when participating in a competition: Casto & Edwards, 2016), with the risk of social rejection and being shamed. These activities all pose a threat to the 'social self' and, as such, comprise a key characteristic of stress activation (e.g., Dickerson & Kemeny, 2004). These 'assessment' stressors provide a useful model of naturalistic stress, due to the importance of the assessment with regards to personal and professional outcomes for these individuals. Such evaluations of one's valued attributes can be argued to

cause high levels of acute stress, as the events hold the properties defined as crucial in the activation of the HPA axis: social evaluation and uncontrollability (Dickerson & Kemeny, 2004).

However, the drawback with stressors of this nature is that whilst these events are often perceived as highly stressful, they are, to some extent, a routine or voluntary part of these individuals' lives. For example, in elite athletes, competition is not unusual, and in fact is a key aspect of one's role as an elite athlete (Bille et al., 2006). The same could be argued for competitive ballroom dancers. Medical students on the other hand take regular exams throughout their training; so again, examinations are not atypical for this sample. The issue of routine could, however, introduce the potential for habituation to occur (e.g., Kirschbaum, 1995), and whilst the outcomes of such events may be of considerable importance to the individual, they are not situations that involve a threat to survival (i.e. an immediate need for fight or flight).

Studies assessing the psychobiological response in individuals taking part in extreme, high-risk sports, defined as activities involving a high potential for personal injury or death, could, however, address these issues to some degree. For example, rock climbing, which is considered a 'high-risk' sport (Watson & Pulford, 2004), elicits a psychobiological stress response, and the level of response for both physiological and psychological parameters appears to increase with the level of risk of the type of climb (Draper, Jones, Fryer, Hodgson & Blackwell, 2010). In lead rope climbing, which involves the climber leading the climb from the ground, participants demonstrate greater  $VO_2$  max, cortisol responses, self-reports of perceived demand, and mental and physical challenge

compared with a less risky climbing activity, such as top rope climbing (Hodgson et al., 2009).

Skydiving, another popular extreme sport has consistently been demonstrated to elicit a physiological stress response (e.g., Chatterton, Vogelsong, Lu & Hudgens, 1997; Hare, Wetherell & Smith, 2013, Meyer et al., 2015; Taverniers et al., 2011; Thatcher, Reeves & Dorling, 2003; Yonelinas, Parks, Koen, Jorgenson & Mendoza, 2011). Specifically, skydiving elicits activation of the HPA axis, and has, therefore, more recently, been used as a naturalistic model of acute stress (e.g., Carlson, Dikecligil, Greenberg & Mujica-Parodi, 2012; Hare et al., 2013; Yonelinas et al., 2011). Unlike many other stressors, skydiving carries a true risk of injury or death, which from an evolutionary perspective provides a more ecologically valid paradigm for examining a true ‘fight or flight’ response.

Interestingly, in both skydiving and rock climbing, level of experience does not appear to affect the magnitude of physiological response observed in participants. Climbers attempting a lead climb route for the first time observed the same pattern of physiological arousal as climbers who had previously carried out the same climb (Draper et al., 2010). A similar pattern has been observed in skydiving populations, with a previous study from our group observing no significant differences in cortisol profiles pre and post skydive when comparing novice solo skydivers and experienced jumpers, with experience levels ranging from 30 to 1000 jumps (Hare et al., 2013). Similar findings have been reported for heart rate during the skydive period, with both novice and experienced skydivers demonstrating equivalent responses, suggesting a resistance to habituation to

potentially life-threatening stimuli (Allison et al., 2012). Conversely, a recent study has suggested that whilst the level of experience does not extinguish the cortisol response, there may be a comparably flatter response and faster recovery in experienced compared to novice skydivers (Meyer, 2015). However, key methodological factors may explain the discrepant findings: the group of novice skydivers Meyer and colleagues (2015) recruited were completing a tandem skydive, unlike the majority of the previous studies reporting no signs of habituation, which recruited solo novice jumpers (e.g., Allison et al., 2012; Hare et al., 2013). An early skydiving study also reported the habituation of the cortisol response to successive jumps in one day (Deinzer et al., 1997), seemingly supporting Meyer's (2015) suggestion of a flatter response following repeated exposure. However, again, the protocol adopted for the study by Deinzer and colleagues (1997) differed from the majority of previous studies in the respect that cortisol was not assessed in direct proximity of the skydive, but instead, at 20 minute intervals across the jumping day, during which participants completed 3 parachute jumps. This meant that all participants provided samples at the same times, whether they were jumping shortly after providing the sample (i.e. within the hour), or whether they were jumping later in the day. These differences in sample times in relation to the onset of the stressor, therefore prevent any meaningful interpretation of the effects of the skydive on acute cortisol reactivity. Furthermore, half the participants had their jumping day postponed twice, due to poor weather conditions and, again, it is possible that this may have influenced observed cortisol responses to some degree, particularly with regards to anticipation.

Together, these findings (Meyer et al., 2015; Deinzer et al., 1997) do not convincingly contradict previous evidence that skydiving, as a stressor paradigm, is resistant to habituation (Allison et al., 2012; Hare et al., 2013). Furthermore, observations that novice jumpers report greater anticipatory state anxiety prior to a skydive than their experienced counterparts, despite exhibiting similar patterns of physiological response, demonstrates possible discordance between physiological and psychological stress responses. In line with these findings, climbers who had previously climbed a particular route reported significantly lower pre-climb cognitive and somatic anxiety compared with those climbing the route for the first time (Draper et al., 2010). These findings are of interest as they seem to contradict the general belief that physiological stress responses are triggered by situations which are novel and unpredictable (Mason, 1968) as many of these participants are experienced in their sport and the activities described are therefore neither novel nor unpredictable to them. It is however suggested that for stressors which present a genuine threat to survival, the fight or flight response is absolutely necessary, regardless of the number of times the activity has been experienced, as complacency of the stress response could lead to fatal consequences.

The stressors discussed so far reflect those which can be assessed under naturalistic, yet uncontrolled conditions, and which provide the opportunity to observe the organic preparatory stage in the period preceding a stressor. As the varying levels of severity of stressors indicate (i.e. some which have uncertain and potentially fatal outcomes, whilst others involve considerably less severe consequences), it is likely that this will, in turn, impact the magnitude of observed

response (Dickerson & Kemeny, 2004). However, all of the studies discussed so far illustrate the strength of the stress response, and highlight that the effects of a stressor are not restricted only to the direct exposure itself, but also include the prelude of the event.

#### *Anticipation of laboratory stressors*

Whilst the study of naturalistic stressors has provided insight into the effects of anticipation under these conditions, anticipation of such stressors cannot be manipulated or controlled, and as such, there may be great variance in the conditions under which anticipation is experienced and observed. Research assessing anticipatory periods in the lead up to stressor exposure under the controlled conditions of the laboratory, are limited. However, a small number of studies have demonstrated the integrity of the anticipatory stress response, even under artificial conditions.

In a functional magnetic resonance imaging (fMRI) study, feelings of dread were assessed in participants during the waiting period of an adverse event, an electrical shock to the dorsum of their foot (Berns et al., 2006). Participants were given the opportunity to choose between two sets of voltage and time delay (e.g., “90% in 3 seconds” or “60% in 27 seconds”). The findings revealed that the majority of participants opted for the shorter delay more than 50% of the time, and in some cases, individuals recurrently selected the higher voltage in cases where it would shorten the waiting period. Interestingly the brain imaging findings demonstrated that waiting did not change the response to the shock itself, and that there was no difference in voltage sensitivity between those rated as mild (who dreaded only to the extent of shortening the delay, but were not willing to

take more voltage to shorten the delay) and extreme ‘dreaders’ (who preferred more voltage sooner, to less voltage later). The results suggest that participants had a preference for the shorter delay due to the relief this would provide from anticipatory dread of an unpleasant outcome. This study observed immediate anticipation of an event which participants were told would occur while they were at the laboratory. Therefore, whilst the findings are interesting and provide insight into potential avoidance behaviour related to the anticipation of stressful events, in order to observe longer-term effects of anticipation of a forthcoming demand, a stressor paradigm is required which will allow the manipulation and assessment of a longer anticipatory period.

Other studies have attempted to monitor anticipation in the periods immediately preceding laboratory stressors: for example, 3 minutes (Davidson, Marshall, Tomarken & Henriques, 2000) and 5 minutes (Gramer & Sprintschnik, 2007) prior to exposure. Only one study has assessed anticipation of a socially evaluative laboratory stressor the day prior to attending the laboratory and on the day of the stressor, to provide a comprehensive profile for an anticipatory stress response (Wetherell, Lovell & Smith, 2014). The study observed greater cortisol awakening responses on the day of the stressor compared with the pre-stressor day, demonstrating that presence of a physiological anticipatory response towards the event.

Although there is a paucity of studies assessing anticipation of manipulated forthcoming demand, the evidence so far demonstrates that anticipation does lead to activation of psychological (e.g., anxiety) and biological (e.g., cardiovascular and endocrine) mechanisms. However, little is known about

the consequences which follow an anticipated stressor that is no longer a threat (i.e. the threat is anticipated but the stressor is avoided prior to exposure). Failure to recover following stress can cause prolonged activation of physiological processes which are designed to cope with short term stressors, and this can lead to maladaptive responses such as burnout (Pruessner et al., 1999) and cardiovascular disease (for review, see Dimsdale, 2008). Furthermore, effective recovery and return to baseline has been argued to be one of the most important factors in preventing adverse negative consequences (Brosschot, Gerin & Thayer, 2006). Studies which have focused attention on the recovery period following a stressful event have generally done so following actual exposure to the anticipated stressor, but have not assessed the recovery process following termination of the threat before the stressor has materialised.

As has been discussed, the anticipatory period in the lead up to a stressor can be perceived as stressful, and in some cases is rated as even more unpleasant than the stressor itself. Although few studies have assessed this concept, it has been demonstrated that exposure to the anticipated threat can relieve the unpleasant dread which can be experienced in the lead up to a stressor (Berns et al., 2006). It could therefore be possible that failure to experience the anticipated stressor may present variation in the observed recovery period, different to responses observed in individuals who do experience anticipated exposure.

Unsuccessful recovery from anticipatory stress responses could, in turn, play an important role in the relationship between stress and deleterious health outcomes. One study exploring this concept did so by assessing cardiovascular and psychological responses in individuals who either anticipated and

subsequently completed a socially evaluative stressor (giving a free speech to an evaluator) or only anticipated the stressor, which was subsequently aborted before exposure (Vaugh, Panage, Mendes & Gotlib, 2010). The findings suggested that those who anticipated but did not experience the stressor observed a reduction in self-reported negative affect (NA) compared with their baseline, and with participants who were exposed to the stressor. However, in individuals whose NA did not recover, high heart rate levels were maintained during the ‘recovery’ period. These results demonstrate that the effects of anticipation can prolong the activation of the stress response, demonstrated through reports of greater NA and heart rate in the recovery period following termination of the waiting period preceding exposure.

The studies discussed in this review demonstrate the importance of the investigation of the anticipatory response to forthcoming demand, and the potential usefulness of a paradigm which allows for the controlled manipulation of anticipation towards such an event. The following two study chapters will address this suggestion by assessing the role of anticipation in psychobiological stress responding to both a socially salient laboratory stressor, and a naturalistic stressor.

## **Chapter 6**

Study 2: Physiological and psychological responses to a forthcoming socially  
evaluative laboratory stressor

The findings of the following study were presented at The Northumbria Research  
Annual Conference 2015 (poster presentation).

## Introduction

Chapter 5 provided a review of the literature addressing the concept of anticipation of stressful or challenging events. Whilst there is, at this stage, paucity in the literature directly assessing anticipatory responses during this period, some studies have reported psychological and physiological response profiles during the period preceding forthcoming challenging events, suggestive of anticipated demand. Greater cortisol secretion, self-reported anxiety and stress have been observed during the college examination period (Weekes et al., 2008); greater CARs have been observed in trainee teachers on the day of observation assessments (Wolfram et al., 2013); and greater waking levels of cortisol have also been observed in dancers on the day of a ballroom competition (Rohleder et al., 2007; Allison et al., 2012).

Whilst naturalistic studies are useful in allowing the observation of responses in a real-world setting, the lack of overall control is somewhat undesirable for those wishing to investigate this response in a standardised manner. Therefore, the utilisation of an ecologically valid, and socially salient stressor, allowing the manipulation of forthcoming demand under controlled conditions, is required in order to fully measure this response.

The first study of this research programme (see Chapter 4) sought to develop an ecologically valid laboratory stressor, which would reliably elicit a stress response. The stressor elicited a significant psychobiological stress response (increased cardiovascular function, indicative of sympathetic arousal). Further, in relation to the anticipatory hypothesis, participants who were anticipating full-frontal critical social evaluation reported significantly greater levels of tension,

and lower levels of happiness, calmness and contentedness, before experiencing the stressor, compared with their control group counterparts. Therefore, the stressor paradigm, in addition to providing a tool for assessing stress reactivity, provides a model of anticipation of forthcoming demand fitting the aforementioned criteria.

This study will therefore assess anticipatory responses to the laboratory stressor paradigm developed in Study 1, which provides a controlled environment in which the duration and potential magnitude of anticipation of a stressful event can be manipulated. Anticipation and reactivity on the day of the stressor will also be assessed in the context of the recovery day, to investigate whether anticipatory responses are associated with specific patterns of recovery of the response.

## **Aims**

The aim of the present study was to assess psychobiological responses during an anticipatory period for a forthcoming acute laboratory stressor paradigm, developed in the previous study of this research programme. The paradigm has been found to not only elicit a psychobiological stress response, but also psychological responses suggesting anticipation of a forthcoming demand (greater self-reported tension, lower happiness, calmness and contentedness). Additional psychobiological responses were assessed in the lead up to and recovery period following the event that was both anticipated and experienced.

## **Hypotheses**

It was hypothesised in the present study that individuals would demonstrate greater physiological and psychological reactivity on the morning of the stressor, compared with the other three sampling days (the day before, the day after, and a control/neutral day). It was further hypothesised that a profile indicative of anticipation of forthcoming demand would be observed for the psychological indices assessment, such as greater anxiety, tension, stress, upset and worry, and lower reports of calmness, happiness, contentedness and relaxation prior to stressor exposure.

## Method

### *Participants*

The sample comprised 31 healthy adults, ranging in age from 18-38 years ( $M_{age} = 24.4$ ,  $SD_{age} = 5.18$ ). One participant withdrew from the study after the first sampling day due to sleep disturbance. 30 participants remained in the final analysis (10 males, 20 females).

Participants were volunteers who responded to email advertisements, poster advertisements placed across Northumbria University, as well as undergraduate students who volunteered via the University's online research participation pool. Volunteers were screened for the eligibility criteria, including being aged between 18-40 years of age, and confirmation of the following: Resting blood pressure which did not exceed 140/90; not currently taking steroidal medication; not pregnant or currently breastfeeding; no history of panic attacks.

Participants were compensated for their time with a choice of either 6 participation points (Northumbria University psychology students) or a £10 high street shopping voucher.

Ethical approval for the study procedure was obtained from the Ethics Committee of the Health and Life Sciences Department at Northumbria University.

### *Materials*

#### *Stressor manipulation*

This study utilised the laboratory stressor paradigm developed in Study 1 (see Chapter 1 and 2 for full details).

### *Questionnaires*

Details of the scales completed by participants are presented in Chapter 2. In order to thoroughly assess differences in state variables across each day, in addition to assessing the overall state anxiety scores, items from the morning state mood questionnaires, and subsequent state anxiety questionnaires were analysed individually to measure calmness, tension, upset, relaxation, contentedness, worry, stress, and happiness. Sleep-related items were also analysed to assess mental alertness and physical tension at sleep onset the previous night, as well as morning wellness. All significant effects are presented in the results section.

### *Procedure*

Participants satisfying the eligibility criteria were invited to the laboratory for a short (10 minute) briefing. During this time, participants were told that they would be completing a stressor task which would be cognitively demanding, whilst standing in front of a researcher who would be monitoring their behaviour and performance throughout the task. They were notified that they would receive critical social evaluation of their performance throughout. Willing participants provided informed consent and were given data collection packs to take away with them. These packs included 4 sets of salivette saliva collection tubes (6 samples for each of the 4 sampling days), a questionnaire booklet (see Chapter 2), state anxiety questionnaires, a sleep quality diary and a saliva sample collection diary. Participants received training on how to provide the saliva samples, and were instructed to accurately record saliva collection times. All participants were required to provide samples the day before returning to the laboratory for the stressor (day 1), the day of the stressor (day 2), the day after (day 3), and on a

control day (day 4: a typical day which was at least two days after day 3). For sampling times, see Table 6.1.

All participants agreed to provide the researcher with their mobile phone number in order to receive reminders each evening before a sampling day, prompting them to put their salivette tubes out in preparation for the next morning.

On the day the participants attended the laboratory for the stressor session, the Study 1 protocol was replicated, including the number and timing of saliva samples provided during the 1 hour-long session (see Chapter 2). In addition, following the stressor, participants provided a further two days of samples (the day after the stressor and control day), before returning their samples to the laboratory and receiving a debrief. See Chapter 2 for treatment of saliva samples.

**Table 6.1** Sampling times for saliva samples and state anxiety questionnaires.

<i>Time point</i>	<i>Saliva / self-reports</i>
<i>Immediately upon awakening</i>	Saliva sample 1
<i>+30 minutes</i>	Saliva sample 2
<i>+45 minutes</i>	Saliva sample 3
<i>+1 hour</i>	Saliva sample 4 Sleep diary and state mood 1
<i>+6 hours</i>	Saliva sample 5 State mood 2
<i>Bed time</i>	Saliva sample 6 State mood 3

*Treatment of data*

In order to maximise the sample size participants were included in analyses for which they had provided a full set of measures. As stated in an earlier chapter (see Chapter 2),  $AUC_G$  was calculated using five specific samples, a number greater than required to calculate CAR magnitude. Therefore, due to missing samples at one or more of the required time points (due to insufficient saliva volume and/or missing samples), a large number of participants required removal from analysis of  $AUC_G$ . Due to a considerable drop in sample size when all four sampling days were included, the decision was made in the present study to maximise sample size by focusing only on day 1 (pre stressor) and day 2 (day of stressor) for  $AUC_G$  analysis only.

Repeated measures ANOVAs were conducted to analyse the samples collected during the stressor manipulation (state anxiety, heart rate, SBP, DBP and salivary cortisol). For state anxiety and cortisol, the time points included in the analysis were: arrival at the laboratory, post demonstration, stressor cessation, 10 minutes post stressor (during relaxation period), and 20 minutes post stressor cessation (at the end of the laboratory session). Heart rate, SBP and DBP were assessed upon arrival at the laboratory, post demonstration, upon stressor commencement, stressor +5 minutes, stressor +10 minutes, stressor +15 minutes, stressor cessation, 10 minutes post stressor (during relaxation period), and 20 minutes post stressor (at the end of the laboratory session).

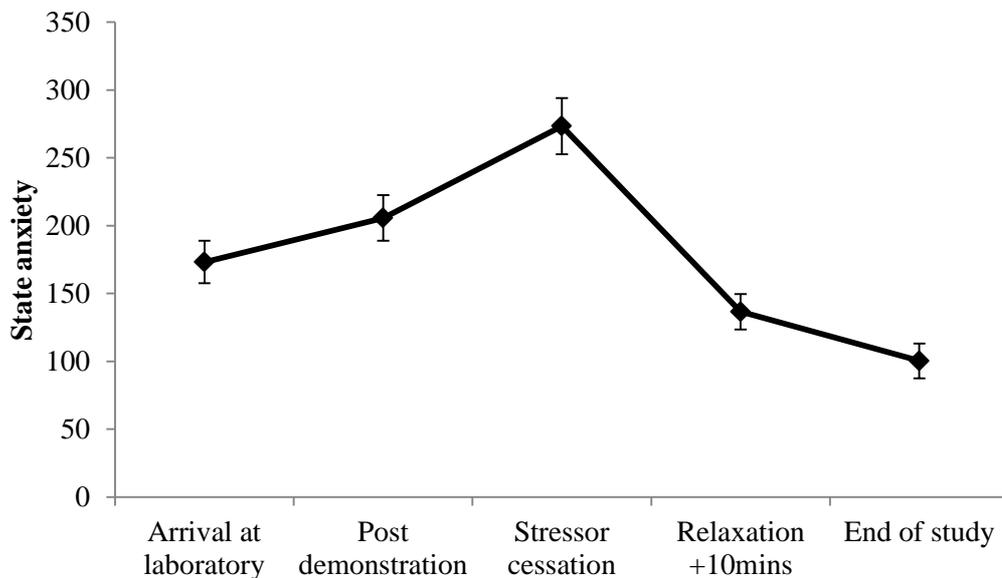
## Results

### *Stressor manipulation*

Mean stress reactivity for each of the following state indices are presented in Appendix C.

### *State anxiety*

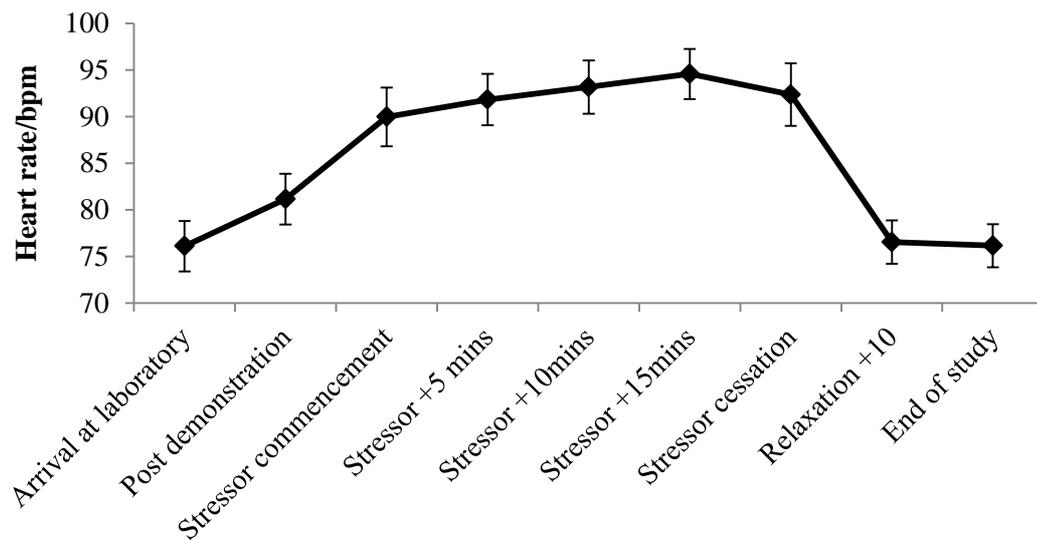
There was a significant main effect of time [ $F(4,23) = 19.03, p < .001$ , Wilks'  $\Lambda = .23$ ], with a large effect size (partial  $\eta^2 = .77$ ). Post hoc revealed significantly greater anxiety post stressor demonstration compared with arrival at the laboratory ( $p = .019$ ), post stressor compared with post demonstration ( $p < .001$ ), upon arrival at the laboratory compared with 10 minutes into the relaxation period ( $p = .012$ ), upon arrival at the laboratory compared with the end of the relaxation period ( $p < .001$ ), and post stressor compared with post stressor demonstration ( $p < .001$ ). See Figure 6.1.



**Figure 6.1** Mean (and SE) state anxiety, assessed during exposure to the stressor manipulation ( $n = 27$ ).

### Heart rate

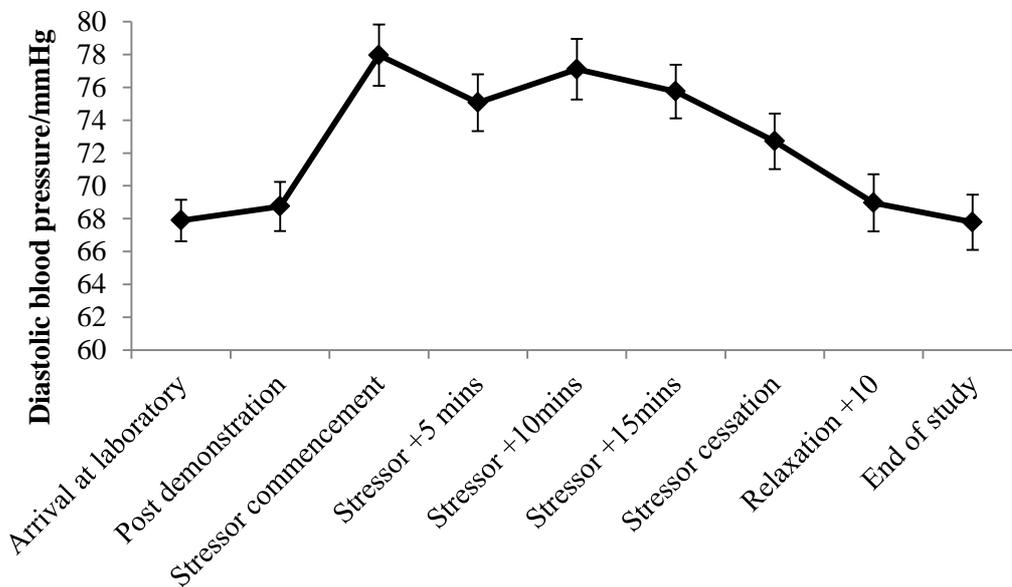
There was a significant main effect of time on heart rate, [ $F(8,20) = 19.82, p < .001, \text{Wilks' } \Lambda = .11$ ], with a considerably large effect (partial  $\eta^2 = .89$ ). Pairwise comparisons revealed significant increases in heart rate from measures obtained upon arrival at the laboratory, to the end of the laboratory session. Heart rate significantly increased from arrival at the laboratory to post demonstration ( $p < .001$ ), from post demonstration to stressor commencement ( $p < .001$ ), and continued to increase significantly ( $p = < .001-.030$ ), until significant decline was observed at stressor cessation. See Figure 6.2.



**Figure 6.2** Mean (and SE) heart rate (bpm) assessed during exposure to the stressor manipulation ( $n = 28$ ).

### *Diastolic blood pressure (DBP)*

There was a significant main effect of time, [F (8,20) = 5.36,  $p = .001$ , Wilks'  $\Lambda = .32$ ] with a large effect size (partial  $\eta^2 = .68$ ). Pairwise comparisons revealed significant increases in DBP from arrival at the laboratory compared with stressor commencement ( $p < .001$ ), and significant increases were observed during the stressor ( $p = <.001-.023$ ), with DBP remaining elevated until stressor cessation, where recovery was observed ( $p = .027$ ). See Figure 6.3.

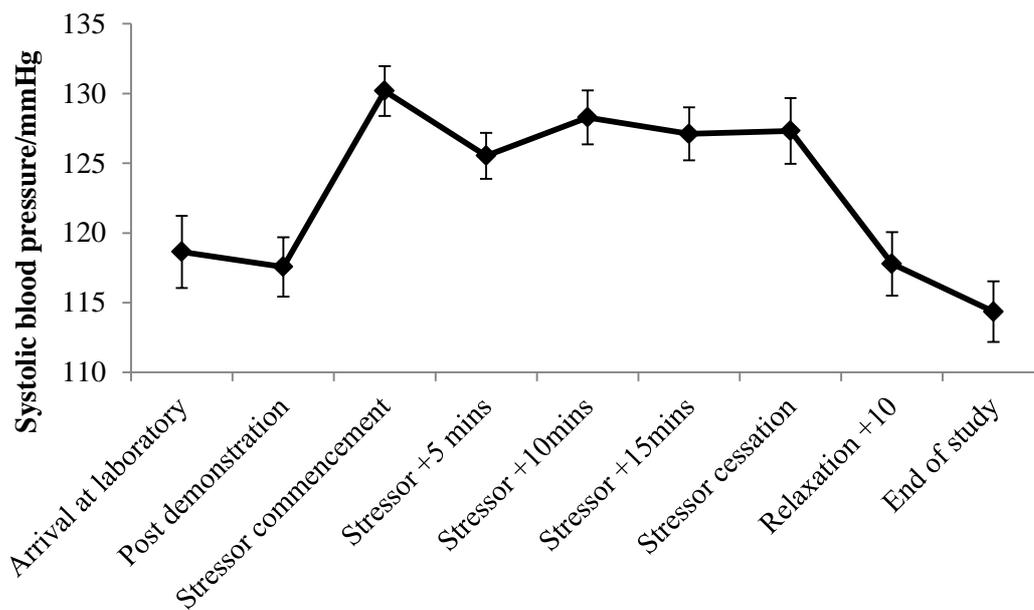


**Figure 6.3** Mean (and SE) diastolic blood pressure (mmHg) assessed during exposure to the stressor manipulation ( $n = 28$ ).

### *Systolic blood pressure (SBP)*

There was a significant main effect of time, [F (8,20) = 15.31,  $p < .001$ , Wilks'  $\Lambda = .14$ ] with a large effect size (partial  $\eta^2 = .86$ ). Pairwise comparisons revealed the following: Lower SBP on arrival at the laboratory compared with stressor commencement ( $p < .001$ ), stressor +5 minutes ( $p = .008$ ), stressor +10 minutes ( $p = .001$ ), stressor +15 minutes ( $p = .001$ ); and stressor cessation

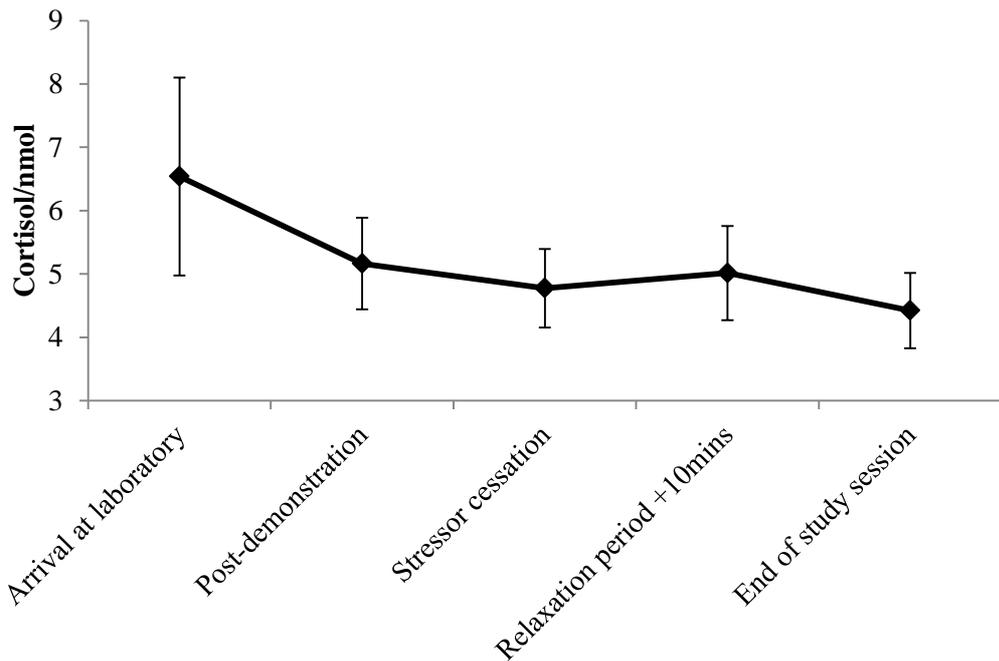
( $p = .002$ ). Lower SBP was observed post stressor demonstration compared with stressor commencement ( $p < .001$ ), stressor +5 minutes ( $p = .001$ ), stressor +10 minutes ( $p < .001$ ), stressor +15 minutes ( $p < .001$ ); and stressor cessation ( $p = .001$ ). Higher SBP was observed at stressor commencement compared with stressor +5 minutes ( $p = .005$ ), relaxation period +10 minutes ( $p < .001$ ), and the end of the study ( $p < .001$ ). Higher SBP was also reported at stressor +5 minutes compared with relaxation period +10 minutes ( $p = .002$ ), at the end of the study ( $p < .001$ ). Higher SBP was observed at stressor +10 minutes compared with the relaxation period +10 minutes ( $p < .001$ ); and at the end of the study ( $p < .001$ ). Higher SBP was also observed at stressor +15 minutes compared with relaxation period +10 minutes ( $p < .001$ ) and at the end of the study ( $p < .001$ ). Higher SBP was observed at stressor cessation compared with relaxation period +10 minutes ( $p < .001$ ) and at the end of the study ( $p < .001$ ). Higher SBP was finally reported at relaxation period +10 minutes compared with the end of the study ( $p < .001$ ). See Figure 6.4.



**Figure 6.4** Mean (and SE) systolic blood pressure (mmHg) assessed during exposure to the stressor manipulation (n = 28).

### *Cortisol*

There was no significant main effect of time on cortisol reactivity throughout the stressor session, [F (4,22) = 2.28, p = .093, Wilks'  $\Lambda$  = .71, partial  $\eta^2$  = .29]. See Figure 6.5.



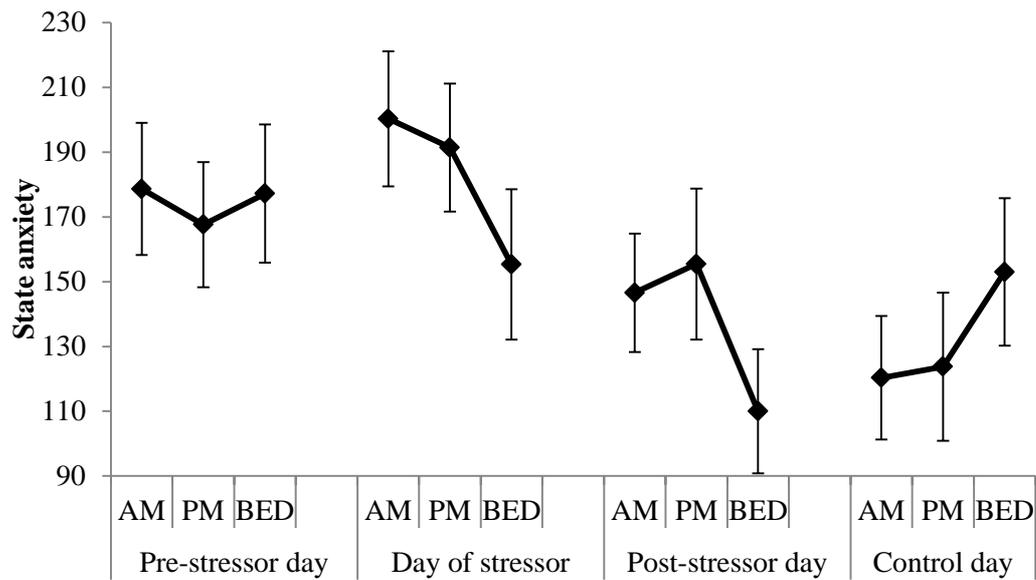
**Figure 6.5** Mean (and SE) cortisol (nmol) assessed during the stressor manipulation (n = 26).

*Diurnal variables*

*Psychological indices*

*State anxiety profile across the day*

A significant main effect of day was observed, [F (3,21) = 4.05, p = .020, Wilks'  $\Lambda$  = .63] with a large effect size (partial  $\eta^2$  = .37). Pairwise comparisons for the significant main effect of day revealed significantly greater reports of state anxiety on the day of the stressor, compared with the post-stressor day (p = .024), and control day (p = .026). There was however, no significant main effect of time, [F (2,22) = 1.47, p = .251, Wilks'  $\Lambda$  = .88, partial  $\eta^2$  = .12] or time x day interaction [F (6,18) = .74, p = .429, Wilks'  $\Lambda$  = .74, partial  $\eta^2$  = .26]. See Figure 6.6.

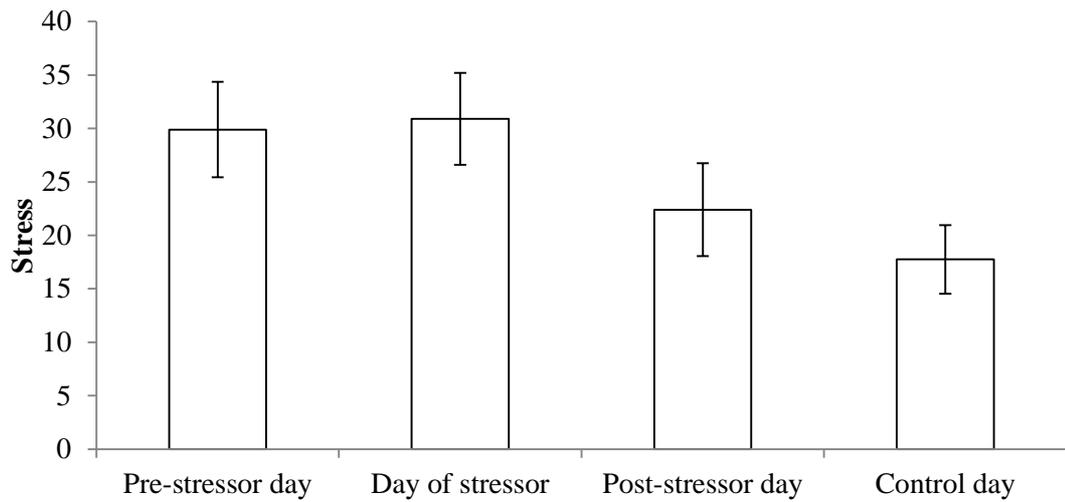


**Figure 6.6** Mean (and SE) state anxiety assessed in the morning, at waking +6 hours and bedtime, over the four sampling days (n = 27).

*Psychological mood reported in the morning*

*Stress*

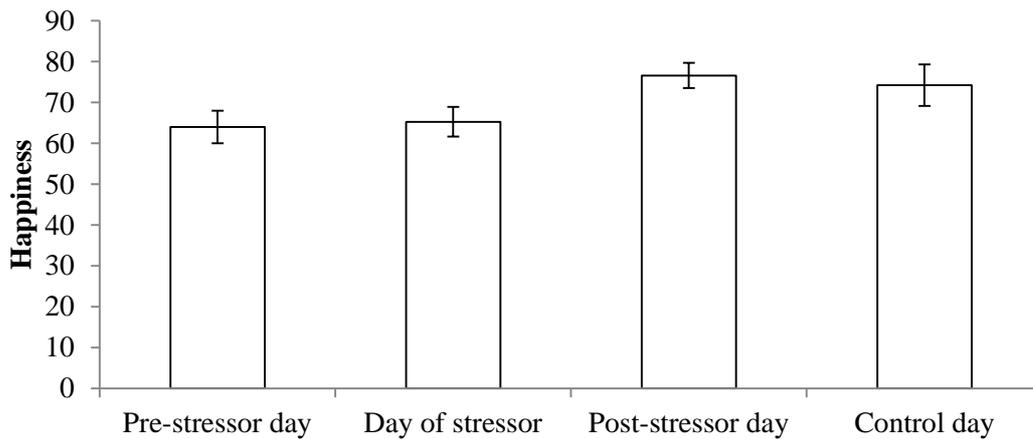
There was a significant main effect of day on self-reported stress, [F (3,25) = 4.32, p = .014, Wilks'  $\Lambda$  = .66] with a large effect size (partial  $\eta^2$  = .34). Pairwise comparisons revealed significantly greater reports of stress on the pre-stressor day compared with control day (p = .009), day of stressor compared with post-stressor day (p = .040), and the day of the stressor compared with the control day (p = .004). See Figure 6.7.



**Figure 6.7** Mean (and SE) self-reported stress assessed in the morning, at waking +6 hours and bedtime, over the four sampling days (n = 28).

### *Happiness*

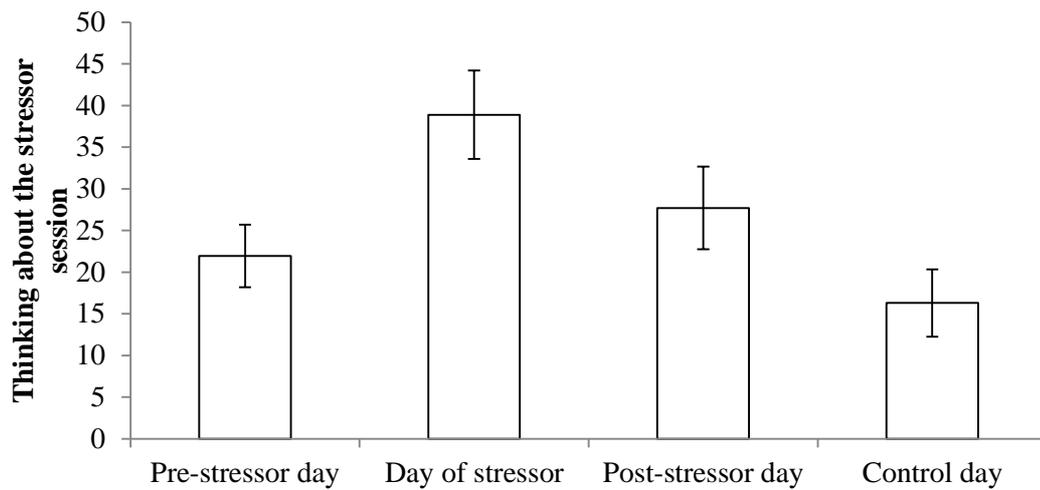
There was a significant effect of day on self-reported happiness, [F (3,25) = 4.34, p = .014, Wilks'  $\Lambda$  = .66] with a large effect size (partial  $\eta^2$  = .34). Pairwise comparisons revealed significantly higher scores for happiness reported on: post-stressor day compared with pre stressor day (p = .006), the control day compared with the pre-stressor day (p = .040), and on the post-stressor day compared with the day of the stressor (p = .005). See Figure 6.8.



**Figure 6.8** Mean (and SE) self-reported happiness assessed in the morning, at waking +6 hours and bedtime, over the four sampling days ( $n = 28$ ).

#### *Thinking about the stressor*

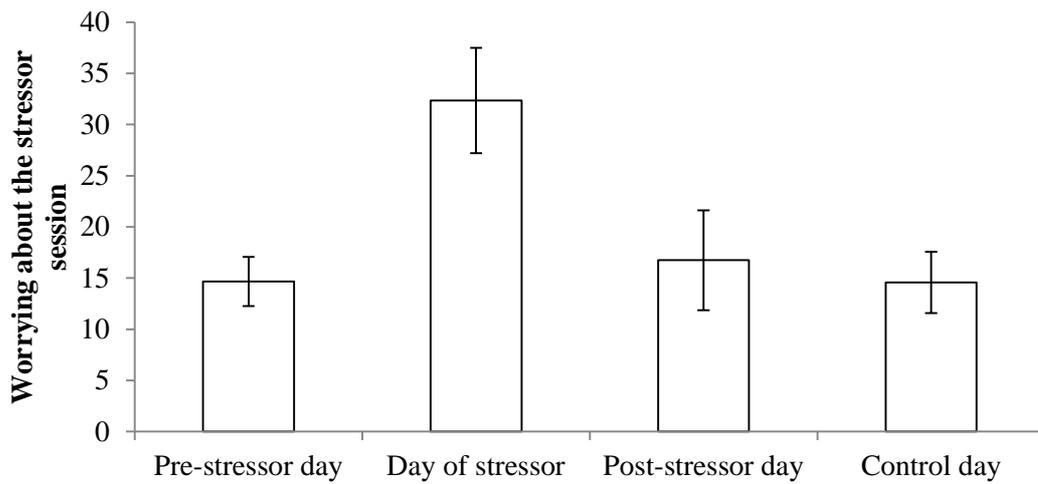
There was a significant main effect of day on the extent to which participants reported thinking about the stressor session, [ $F(3,24) = 6.33$ ,  $p = .003$ , Wilks'  $\Lambda = .56$ ] with a large effect size (partial  $\eta^2 = .44$ ). Pairwise comparisons revealed significantly greater reports of thinking about the stressor session on; the day of the stressor compared with the pre-stressor day ( $p = .004$ ), the day of the stressor compared with the post-stressor day ( $p = .048$ ), the post-stressor day compared with the control day ( $p = .010$ ), and the day of the stressor compared with the control day ( $p = <.001$ ). See Figure 6.9.



**Figure 6.9** Mean (and SE) self-reported ‘thinking about the stressor session’, assessed in the morning, across the four sampling days (n = 27).

#### *Worrying about the stressor*

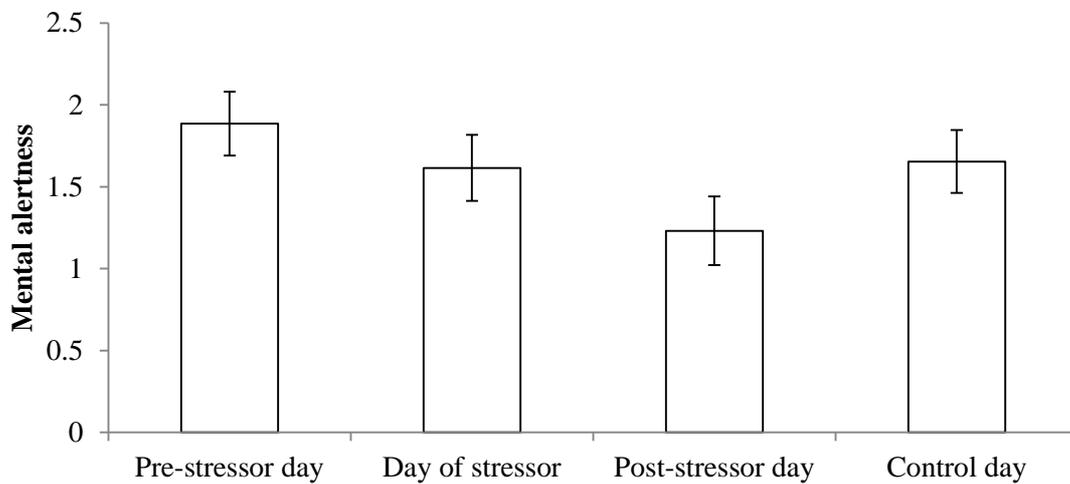
There was a significant effect of day on self-reported worrying about the stressor, [F (3,20) = 4.50, p = .014, Wilks’  $\Lambda$  = .60] with a large effect size (partial  $\eta^2$  = .40). Pairwise comparisons revealed significantly greater reports of worrying on the morning of the stressor compared with the pre-stressor day (p = .001) the day of the stressor compared with the post-stressor day (p = .006), and the day of the stressor compared with the control day (p = .003). See Figure 6.10.



**Figure 6.10** Mean (and SE) self-reported ‘worrying about the stressor session’, over the four sampling days (n = 23).

*Mental alertness*

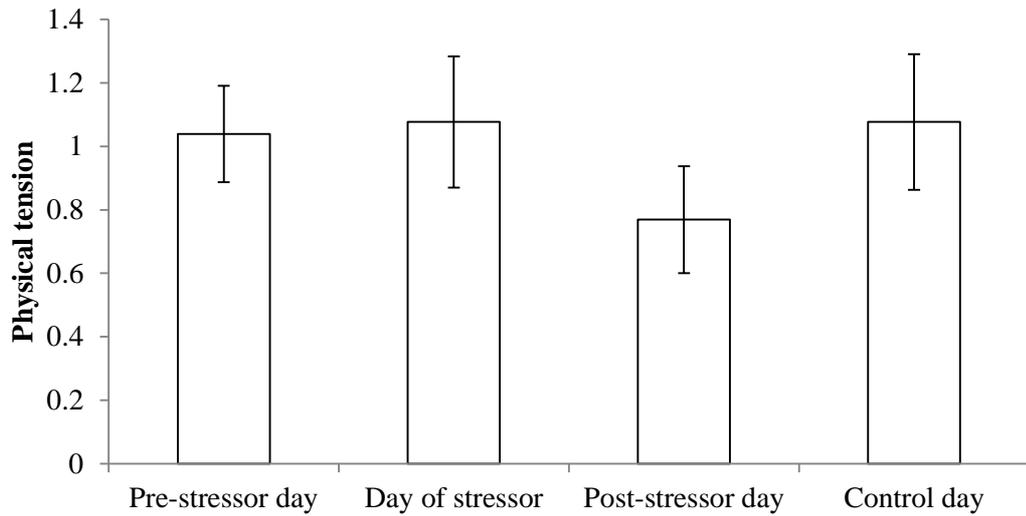
There was no main effect of day on morning reports for mental alertness, [F (3,23) = 1.70, p = .196, Wilks’  $\Lambda$  = .82, partial  $\eta^2$  = .18]. See Figure 6.11.



**Figure 6.11** Mean (and SE) self-reported mental alertness assessed over the four sampling days (n = 26).

### *Physical tension*

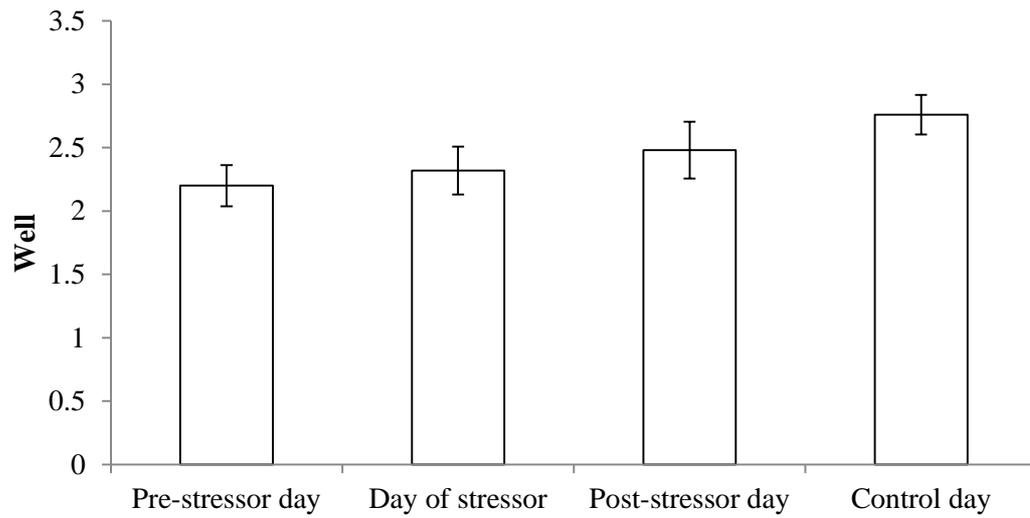
There was no significant effect of day on morning reports of physical tension, [F (3,23) = .712, Wilks'  $\Lambda$  = .92,  $p$  = .555, partial  $\eta^2$  = .09]. See Figure 6.12.



**Figure 6.12** Mean (and SE) self-reported physical tension assessed over the four sampling days ( $n = 26$ ).

### *Wellness*

There was no significant effect of day on reported 'wellness' in the morning, [F (3,22) = 2.081,  $p$  = .132, Wilks'  $\Lambda$  = .78, partial  $\eta^2$  = .22]. See Figure 6.13.



**Figure 6.13** Mean (and SE) self-reported scores of morning wellness tension assessed over the four sampling days ( $n = 25$ ).

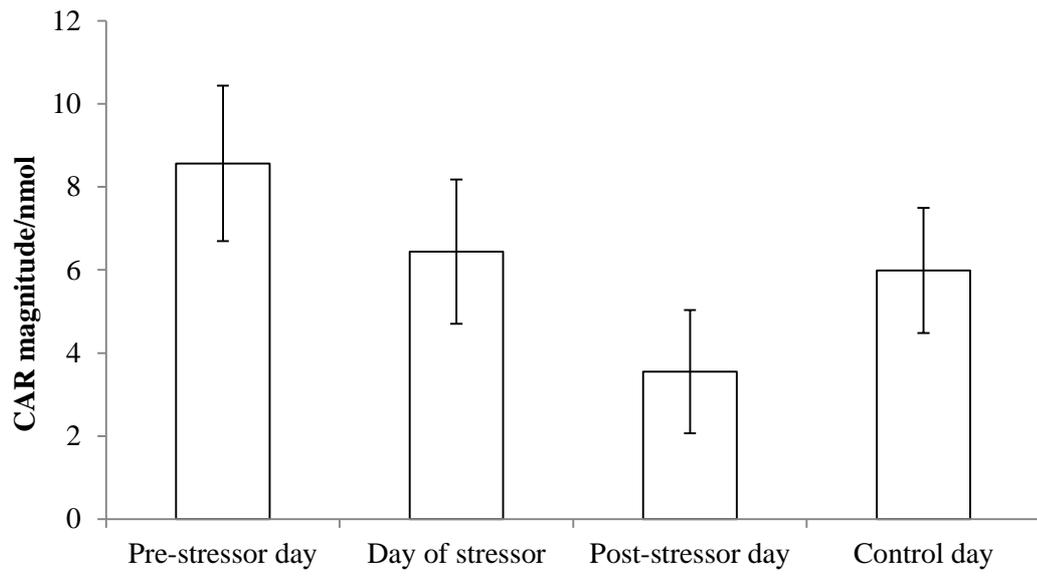
#### *Cortisol indices*

##### *Diurnal profiles*

A paired samples t-test revealed no significant differences in  $AUC_G$  between pre-stressor day ( $M = 5816.67$ ,  $SD = 3753.40$ ) and day of stressor ( $M = 5751.38$ ,  $SD = 2959.98$ ), [ $t(16) = .12$ ,  $p = .909$ ].

##### *CAR magnitude*

There was no significant main effect of day on CAR magnitude, [ $F(3,18) = 1.99$ ,  $p = .152$ , Wilks'  $\Lambda = .75$ , partial  $\eta^2 = .25$ ]. See Figure 6.14.



**Figure 6.14** Mean (and SE) CAR magnitude assessed across the four sampling days (n = 21).

## Discussion

The importance of recovery following activation of stress mechanisms has been extensively highlighted across the literature to date, with empirical evidence demonstrating a link between stress exposure and disease. It has, more recently, been suggested that it is not merely the stress response itself that causes these deleterious health outcomes, but that it is, more specifically, prolonged or inappropriate activation of these systems which leads to negative consequences. Recent studies have suggested that anticipatory psychological indices are also capable of initiating the activation of stressor mechanisms in advance of the experience and prolonging the activation of these physiological responses, establishing the importance of broadening knowledge of this area, which is currently sparsely studied.

The aim of the present study, therefore, was to assess psychobiological responses during both the anticipatory period leading up to, and the recovery period following, an acute laboratory stressor paradigm, which in Study 1, was demonstrated to elicit psychological responses suggestive of an anticipatory response. Both the acute stressor phase (i.e. direct responses to the stressor) and anticipation of the stressor were assessed. This was achieved by assessing psychobiological indices, from waking to bedtime, over four sampling days: the day before exposure to the stressor; the day of the stressor; the day following the event and on a control day. As far as could be controlled, these four sampling days were identical in terms of planned activities, with the exception of participation in a novel laboratory stressor on the day of the stressor, and

therefore, differences in observed responses can be attributed to the anticipation of, and exposure to, the stressor.

With regard to acute stress reactivity to the laboratory paradigm, there was a significant effect of time on state anxiety, with increases in anxiety observed between arrival at the laboratory and following the demonstration of the stressor task, and between the end of the demonstration and stressor cessation. Anxiety returned to baseline within ten minutes of stressor cessation, during the relaxation period, and was at its lowest at the end of the study. These findings concur with those observed in Study 1, demonstrating considerable replicability in this parameter.

Upon analysis of the cardiovascular parameters, as was the case in Study 1, a significant effect of time was also observed for heart rate, with levels increasing following the demonstration of the stressor, and remaining elevated until ten minutes following commencement of the relaxation period. Systolic blood pressure (SBP) also significantly increased during the stressor session, with increases observed between arrival at the laboratory and stressor commencement. As with heart rate, SBP remained elevated until the relaxation period.

The observed increases in SBP responses were consistent with the results reported in Study 1, whereby post-stressor activation was observed. However due to the greater number of measurements included in the present study protocol, the findings from the present study revealed increases at the onset of the stressor. Similarly, the present study also observed significant increases in diastolic blood pressure (DBP) during stressor exposure, which again, concurs with the findings reported in Study 1. Together, these findings demonstrate that the stressor

paradigm is effective in triggering SBP and DBP responses, evidenced by elevated readings for these indices during stressor exposure, in the present study. It should be noted that whilst these time points were intervals at which we would hypothesise changes due to increase (and later decrease) in demand (stressor commencement and stressor cessation), stressor commencement was the first time point of assessment following transition from sitting to standing, a change in position which has been demonstrated to affect cardiovascular readings (e.g., Eser, Khorshid, Gunes & Demir, 2007). However, the stressor cessation reading was taken while participants remained standing, which demonstrates that changes observed at this stage were a result of the stressor and not a change in position. It should also be noted that the increases in blood pressure observed upon stressor commencement are not consistent with blood pressure reactions to the transition from sitting to standing, with this activity typically causing a decrease in readings, not an increase (Eser et al., 2007; Olufsen, Ottesen, Tran, Ellwein, Lipsitz & Novak, 2005). No cortisol reactivity was observed in response to exposure to the stressor paradigm. However, this finding was expected due to the non-significant effects reported in Study 1.

Dickerson & Kemeny (2004) suggest that in addition to the role of social evaluative threat as a key component in eliciting activation of the HPA axis, there is a dose-response relationship between the degree of social evaluation involved in a stressor, and the magnitude of cortisol response. As reported in Study 1, the stressor paradigm presented here did not elicit a cortisol response; which, as discussed in the corresponding chapter may have been due to an insufficient level of social evaluation in the paradigm (i.e. the stressor was not threatening enough

to the social self: Dickerson & Kemeny, 2004). As discussed in the context of Study 1, this could explain why some studies examining psychobiological responses to laboratory stressors have observed cortisol responses, whilst others have not. For example, the TSST (Kirschbaum et al., 1995), involves a rigorous assessment including the presence of a panel of a minimum of 3 assessors, providing feedback while the participant delivers a free speech, and completes a mental arithmetic task. In studies exactly, or very closely, replicating the paradigm, cortisol responses have been observed (e.g., Zoccola & Dickerson, 2011), whilst studies which have presented modified versions, manipulating (often reducing) social evaluative threat, have yielded cortisol reactivity of a reduced magnitude (for review, see Biondi & Picardi, 1999).

Although a variety of laboratory stressor paradigms have been devised in order to examine the acute stress response, few laboratory stressor studies have assessed participants during the anticipatory period of the event. However, in a recent study adopting a stressor protocol based on the TSST, not only were cortisol responses observed in direct response to the acute stressor, but greater CARs were also observed on the morning of the stressor, compared with the day prior (Wetherell et al., 2014).

Based on the findings from previous studies observing greater physiological (Wetherell et al., 2014) and psychological (Rohleder et al., 2007) arousal on the morning of challenging events, together with observations that the stressor designed in Study 1 prompted more thinking and worrying about the stressor on the day of the event, it was hypothesised that self-reported anxiety would be greater on the day of the stressor, compared with the other three

sampling days. This hypothesis was supported with greater reports of anxiety being reported on the day of the stressor, compared with the day after the stressor, and a control day. These findings are consistent with those from previous studies observing higher levels of stress in the anticipatory period preceding novel, naturalistic stressors (e.g., skydiving: Hare et al., 2013; and climbing: Pijpers, Oudejans, Holsheimer & Bakker, 2003). The morning reports of state mood also suggest that the day of the stressor was perceived to be more psychologically taxing than the other sampling days; participants reported feeling significantly more stressed on the day of the stressor, and the day before the stressor, compared with the control day. Greater levels of stress were also observed on the day of the stressor compared with the day after the event. The finding that those preparing for a stressor report higher subjective stress during this period is consistent with other studies that have assessed novel stressors in the laboratory (e.g., Juster, Perna, Marin, Sindi & Lupien, 2012). In contrast, participants also reported feeling less happy the day before the stressor and on the day of the stressor, compared with the day after the stressor, and on the control day. Taken together, these data demonstrate a profile of greater levels of distress and lower positive mood on the day of anticipated stressful events.

Previous studies have observed increased CARs and greater daily secretion of cortisol on days when individuals are anticipating a forthcoming stressor (Lovell, Wetherell & Smith, 2014). In addition, as the stressor paradigm is associated with reports of greater perceived demand and greater levels of thinking about the event, it was hypothesised that anticipation of the stressor would elicit a greater cortisol response. The findings of the present study, do not,

however, support this hypothesis, with no significant differences observed for CAR magnitude or overall diurnal cortisol secretion across any of the sampling days. This finding does not support previous literature which has demonstrated greater CARs on the mornings of stressful events; however, there are possible explanations for this finding. Typically, empirical studies demonstrating elevated CARs and cortisol secretion over the day have focused attention on a variety of stressors which involve a high level of social-evaluative threat. For example, for elite athletes anticipating a competition, the forthcoming event can be described as an anxiety-arousing situation (Salvador et al., 2003); under these conditions, the abilities of competitors are being observed and assessed by others, and they are competing to either win or lose. In situations such as these, it could be argued that the outcome of the stressful event (i.e. the competition) would have considerable consequences for the individual. That is, due to the importance of the ensuing consequences, the anticipatory response may reflect concern for the effects of the outcome post exposure. This element of threat is not one which is included in the stressor paradigm implemented in this study. Despite efforts to include a high level of social evaluation, there were no real or perceived post-stressor consequences of poor performance during the stressor session once the session was complete (i.e. participants attended the laboratory, and were free to remove themselves from the threat once the study was over, or indeed at any point during testing).

In keeping with the aim to assess anticipation of forthcoming demand, participants were also asked to report how much they were thinking about and worrying about the stressor session. The results demonstrate that participants were

thinking more about the stressor on the morning of the laboratory visit compared with the day prior to and the day following the stressor, and the control day. There were also significantly greater reports of thinking about the study the day after the stressor compared with the control day (3-5 days later) which could suggest that participants may have ruminated about the session for a short period after the event. Reports of greater levels of worrying on the morning of the stressor were also observed, compared with all other sampling days.

The finding that participants reported greater anxiety and generally less positive mood on the morning of the stressor compared with the other sampling days demonstrates that whilst no HPA axis activation occurred during the anticipatory period, participants were, as suggested by the findings of Study 1, anticipating a forthcoming demand, with reports of more stress and less happiness on the day they were attending the laboratory. These findings are also in line with those found in other studies, whereby individuals anticipating an acute stressor later in the day have reported more negative psychological feelings that morning compared with other days (e.g., Wetherell et al., 2014).

The observation that subjective psychological responses are not parallel with cortisol reactivity is not a novel phenomenon, with a number of studies demonstrating a lack of concordance between these indices both in naturalistic stressors (e.g., Hare et al., 2013) and under laboratory conditions (Gruenewald, Kemeny, Aziz & Fahey, 2004). Moreover, this finding has also been reported following a review of studies assessing cortisol reactivity and self reported psychological responses to acute stressors (Dickerson & Kemeny, 2004), with no associations found between cortisol responses and subjective distress. The authors

concluded that despite clear observations that social-evaluative stressors elicit a significantly greater physiological response than any other form of stressor, they were not perceived to be more distressing than any of these stressors. These findings support the notion that the diverse ranges of profiles of stressors elicit an equally broad range of response patterns, which reflect the varying underlying mechanisms underpinning the stress response.

The present study represents the first to have obtained a comprehensive diurnal profile over the days surrounding a novel stressful event, not only for cortisol indices but also for a range of psychological parameters, including anxiety, stress, happiness, and those directly related to anticipation, such as the degree to which participants were thinking and worrying about the stressor. This allowed for thorough investigation and analysis of the anticipation and recovery periods, which has not previously been assessed with this level of detail.

Further, to ensure that responses were indicative of feelings towards the stressor and not external variables, in the first instance participants were asked when booking the stressor session, to choose a period in which they had no other events which could be considered either stressful or atypical to them. To control for unexpected events which could fall under one of these categories, on each day participants were required to report whether they would describe the day as typical to them, and if not, were asked to provide details. There were no reports of changes to planned non-eventful days and, as such, in the absence of retaining participants in a controlled environment, it is assumed that days included in the study were typical to the individuals.

Whilst the present study expanded the sampling window surrounding a stressor, future work could improve on the methodology employed here by increasing the level of assessment during the study to gain a more comprehensive profile of everyday activities and their potential effects on study outcomes. For example, utilising actigraphy for every participant, to objectively measure sleep and waking, would enable more accurate data assessing activities and allow for cross-examination of the timings of saliva samples to improve on monitoring of adherence to sampling protocols.

A further modification would be to increase the perceived anticipation of the stressor by adopting the procedure used for the TSST, either in full (i.e. to carry out the procedure when participants attended the laboratory) or to simply brief participants that this would be the procedure they would experience when attending the laboratory (i.e. to elicit anticipation of a public speaking stressor whereby assessment would be undertaken by 3 assessors). This modification could evoke greater anticipatory responses reflecting the cortisol reactivity previously observed in direct response to the TSST due to the increase of perceived social evaluative threat. However, at this stage, there is no evidence to suggest that the anticipation of a stressor known to increase cortisol (e.g., the TSST: Kirschbaum et al., 1993) would increase basal or waking cortisol levels any more than the current stressor paradigm, which was already associated with increased psychological anticipatory processes.

### *Conclusion*

This was the first study to assess the anticipatory and recovery period surrounding exposure to a novel laboratory stressor, to thoroughly assess both the physiological and psychological response to a forthcoming stressor. The stressor paradigm was associated with acute stress reactivity, specifically psychological and cardiovascular responses, but not cortisol reactivity. Moreover, anticipation of the stressor paradigm was associated with increased reports of stress and negative emotions. No differences in basal HPA responding were observed and this may reflect a lack of control over extraneous factors, or the fact that the manipulated stressor had no significant consequences for the participant and, as such, as an adaptive function, HPA responding was not necessary.

In summary, the findings of the present study illustrate the importance of research assessing acute laboratory stressors to extend their sampling protocol to include the anticipation and recovery periods preceding and following the stressor task, as this may help address the gaps in the literature and add to knowledge of prolonged activation of the stress response. The following chapter will examine the anticipatory response to a naturalistic stressor paradigm, which will be characterised by greater levels of uncontrollability and, unlike the stressor utilised in the present study, will involve significant potential consequences for participants engaging with the activity.

## **Chapter 7**

Study 3: An investigation of the psychobiological anticipatory response in individuals preparing for a solo skydive

The findings of the following study have been reported at:

Craw, O, Smith, M.A, Wetherell, M.A (2016). Weather to Fly: The psychobiological effects of anticipation of a forthcoming skydive. The Psychobiology Section of the British Psychological Society Annual Scientific Meeting, the Lake District.

## Introduction

Assessing anticipation under controlled conditions whereby anticipation can be manipulated (as demonstrated in Study 2) is useful in the experimental examination of the possible mechanisms underlying this response. However, in order to investigate how individuals respond to stressors under natural circumstances (i.e. with little input from the researcher) anticipation should also be measured in the period preceding naturally occurring stressors: that is, stressors which participants experience without experimental intervention and, as suggested in the previous chapter, which are characterised by a sufficient amount of perceived threat to the individual, to observe HPA axis reactivity. Chapter 5 presented and discussed a number of naturalistic stressors which have been assessed with regards to anticipatory stress responses: for example, in the period preceding specialist medical training examinations (Gonzalez-Cabrera, Fernandez, Iribar-Ibabe & Peinado, 2014); ballroom competitions (Rohleder et al., 2007); and league football matches (Alix-Sy et al., 2008).

An appropriate naturalistic paradigm for use as a tool to assess anticipation requires the capability to elicit a robust physiological response. One activity which has been observed to elicit robust stress reactivity, is skydiving. Skydiving, a reasonably popular, high-risk, extreme sport, has attracted research interest in an attempt to understand the psychological profile of individuals who choose to participate in the sport. Not only does the high-risk characteristic of skydiving present a more intense challenge than many other stressors (Yonelinas et al., 2011), but skydiving triggers an acute stress response, evidenced through increased cortisol secretion in response to tandem skydiving (Carlson, Dikecligil,

Greenberg & Mujica-Parodi, 2012; Meyer et al., 2015) solo static line skydiving (Hare et al., 2013) and solo skydiving from a balloon, from a slightly lower altitude (Taverniers et al., 2011). Furthermore, the activity increases reports of anxiety in novices (Allison et al., 2012; Hare et al., 2013) with first-time jumpers reporting greater anxiety immediately prior to boarding the plane than experienced jumpers. Similar findings have been reported in larger samples, also comparing novice and experienced skydivers, whereby novices demonstrated a greater contrast between pre and post jump subjective anxiety, while experienced jumpers exhibited lower anxiety and greater happiness before the skydive (Price & Bundsen, 2005). Our previous work demonstrated that the physiological aspect of the response is observed whether the participant is taking part in their first, or 1000<sup>th</sup> skydive (Hare et al., 2013), and this has been demonstrated elsewhere (Allison et al., 2012), although those with greater experience (>30 jumps) did not report differences in anxiety in immediate proximity of the skydive (Hare et al., 2013). Interestingly, whilst the point of experience at which anxiety habituates is not known, a larger scale study observed significant increases in pre jump anxiety in skydivers who had completed up to 10 jumps ( $M_{\text{jumps}} = 5$ ), demonstrating that the response does not rapidly habituate with experience (Boldak & Guskowska, 2013). These findings demonstrate robust resistance to habituation with regard to cortisol reactivity, and a reasonable resistance to habituation of psychological indices. Previous studies have consistently endorsed skydiving as a salient naturalistic stressor, eliciting a robust psychological and physiological stress response and, as such, the activity is deemed a suitable stressor for the purpose of assessing anticipation and recovery of the stress response.

Due to the nature of the sport, whereby participation in the activity is heavily dependent on external variables such as the weather and daylight hours, the activity provides a unique paradigm whereby participants may anticipate and encounter the stressor, or where the anticipation may be aborted prior to exposure (i.e. before the jump). This naturally occurring anticipation paradigm offers the opportunity to study the recovery period in those who anticipate and either do or do not experience a planned stressor, without intervention from the researcher. The present study will therefore directly assess individuals' physiological and psychological responses during both the anticipatory period preceding, and the recovery period following, novice participation in a solo skydive.

## **Aims**

The primary aim of the present study was to investigate psychological and physiological activity during the anticipatory period preceding, immediate exposure to, and the recovery period following, a naturalistic stressor which has been demonstrated to elicit a robust stress response: skydiving.

## **Hypotheses**

It was hypothesised that partaking in a solo skydive would elicit a physiological stress response upon direct exposure, followed by a brisk recovery, demonstrating activation of the HPA axis. It was further hypothesised that there would be daily variation in both physiological and psychological reactivity, suggestive of an anticipatory response to the forthcoming skydive. Based on previous research findings, greater cortisol output, greater negative emotions (such as anxiety and stress) and lower positive emotions (such as happiness and calmness) were hypothesised on the morning of the planned skydive, reflective of anticipation of forthcoming demand.

## Method

### *Participants*

The study comprised 31 healthy adults; 3 were removed due to non-adherence to saliva sampling protocol and/or failing to provide samples and complete questionnaires (one of these participants was included in all analyses except those assessing cortisol), and 4 failed to return their samples to the researcher. A total of 24 participants were therefore included in data analysis, (15 males, 9 females), ranging in age from 18-28 ( $M_{\text{age}} = 21.4$ ,  $SD_{\text{age}} = 2.26$ ).

Participants were recruited from university skydiving clubs in the North East of England. Students who were enrolled on upcoming Ram Air Progression System (RAPS) solo skydiving courses were invited to participate via email. No participants had any previous skydiving experience and were all novice jumpers. Eligibility criteria included those aged 18-40, with a resting blood pressure which did not exceed 140/90 and who were not currently taking steroidal medication.

### *Materials*

Details regarding the questionnaires issued to participants, as well as the saliva collection technique used for this study can be found in the general methods section (see Chapter 2).

Participants completed a total of 30 saliva samples: 6 on day 1, day 3 and day 4, and 11 on day 2 (if they completed the planned skydive).

In addition to completing the questionnaire booklet (as described in Chapter 2), participants were required to complete state anxiety questionnaires (Marteau & Bekker, 1992) at the time of their saliva samples: one in the morning (which also

contained items regarding sleep quality and anticipation of the forthcoming skydive); one 6 hours post awakening, and finally; one at bedtime. Participants were also equipped with actiwatches in order to measure activity as a proxy of sleep and wake times (for details regarding the actigraphy equipment, see Chapter 2).

### *Procedure*

The study was advertised to local university skydiving clubs via email and social media. Volunteers satisfying the eligibility criteria were invited for a briefing, where they were provided with details regarding the study and were subsequently given data collection packs to take away (including questionnaires as described in Chapter 2, and Salivette saliva collection tubes). The study protocol spanned over 4 days, 3 consecutive days and one control day: one day of researcher-collected samples, the day of the planned skydive (day 2), whereby participants were woken by the researcher at the parachute centre and all samples from waking until 1 hour post-jump were obtained and frozen immediately (participants self-collected the sample at bed time on this day). Three further days of self-collected samples and questionnaires were obtained: the day before the skydive (day 1), the day after the skydive (day 3), and a ‘typical’ control day (day 4: a day of the participants’ choosing which was 3-5 days after the third day of sampling).

On days 1, 3 and 4, participants provided 6 saliva samples in the domestic setting: immediately upon awakening; 30 minutes post awakening; 45 minutes post awakening; 1 hour post awakening; 6 hours post awakening; and immediately before bed. The researcher met participants at the airfield on day 1

and collected the bedtime sample on this day. Day 2 (the day of the planned skydive) involved an identical sampling protocol; however, as stated previously, all samples were collected at the parachute centre by the researcher. Participants who completed a skydive provided 6 additional samples: 1 hour prior to the jump; 30 minutes prior to the jump; immediately prior to the jump; immediately post jump; 30 minutes post jump; and 1 hour post jump. See Table 7.1 for sampling procedure for day 2.

The RAPS solo skydiving course completed by participants involved approximately 8 hours of ground training by a British Parachute Association qualified instructor before taking part in a solo static line skydive. The jump involved the participant leaving the aircraft at 3,500 feet (approximately 15 minutes of flight time). The main parachute was deployed using a device called a 'static line'. This is a length of webbing attached to the aircraft at one end, and the bag (in which the main parachute is kept) at the other. On the instructor's command, participants vacated the aircraft, and as they fell away the static line pulled the main parachute out and this started the deployment. The canopy used was a modern square parachute made from technically advanced materials and specifically designed to allow the jumper to steer the canopy to the landing area. All participants had a radio fitted to their helmet, in the event that they required assistance during the landing process. There were no instances of malfunctions or complications during the skydive for any participants recruited for this study.

The course duration lasted from Friday evening to the following day (Saturday) in all cases. Students taking part in the skydiving course completed half of their training (4 hours) on the Friday evening, slept at the airfield, and

subsequently completed the second half of the training on the Saturday morning. Following successful completion of the training, participants would then complete their first solo skydive on Saturday afternoon (weather and daylight depending).

For samples collected at home, participants were instructed to store them in their own fridges and return them to the researcher as soon as possible, following collection. The return of the final samples and questionnaire booklet marked the end of the study. Participants were reimbursed for their time with a £10 high street shopping voucher.

**Table 7.1** The testing procedure on the day of the skydive (day 2).

<i>Time point</i>	<i>Samples/scales completed</i>
<i>Waking</i>	Saliva sample 1
<i>Waking +30 minutes</i>	Saliva sample 2
<i>Waking +45 minutes</i>	Saliva sample 3
<i>Waking + 1 hour</i>	Saliva sample 4 Morning mood questionnaire/sleep diary [Breakfast] [Lunch]
<i>Waking + 6 hours</i>	Saliva sample 5 State anxiety questionnaire 1
<i>1 hour pre skydive (when putting on equipment)</i>	Saliva sample 6 State anxiety questionnaire 2
<i>30 minutes pre skydive (when sitting on flight line)</i>	Saliva sample 7 State anxiety questionnaire 3
<i>Immediately pre skydive (walking out to board plane)</i>	Saliva sample 8 State anxiety questionnaire 4
<i>Immediately post skydive (walking back to remove equipment)</i>	Saliva sample 9 State anxiety questionnaire 5
<i>30 minutes post skydive (before skydive debrief from instructor)</i>	Saliva sample 10 State anxiety questionnaire 6
<i>1 hour post skydive (after skydive debrief from instructor)</i>	Saliva sample 11 State anxiety questionnaire 7 [Dinner]

*Treatment of data*

Following removal of data based on non-adherence checks (see Chapter 2) 24 participants were included in final analyses, though as evidenced in differences in sample size across analyses, some data were missing, as participants were only included in analyses for which they included complete data. Although two naturally-occurring groups were expected to emerge (1. those who anticipated a skydive and completed as planned, and 2. those who anticipated a skydive and did not complete as planned) due to unexpected circumstances, it was deemed appropriate, in line with the research question, to categorise participants into one of three groups, based on duration of anticipation. The groups which emerged during testing were those who; ‘jumped as planned’ (n = 6: jumped on day 2 as per original protocol), ‘anticipated with doubt’ (n = 11: these participants were told by the skydiving instructor on the evening of day 1 that due to poor weather forecast that they may not jump the next day), and those who ‘knew they were not skydiving the next day’ (n = 7; notified by the instructor that they would definitely not be jumping the next day, due to extremely poor weather forecast). However, two participants who jumped on day 3 (not day 2, as planned) also provided the samples immediately before and after the skydive, and so are also included in the acute skydive analysis for the skydive day (n = 8), but are included in the ‘anticipated with doubt’ group for diurnal analyses.

Despite the planned protocol, there was a considerable level of variability in the conditions for each participant with regards to anticipation timescale and

actual activity. All participants provided samples for the 4 days; however, the variability in actual activity on the day of the planned skydive was difficult to control. For example, some participants who did not jump on the planned day, decided to return to jump on the following day, and therefore they would potentially experience extended or a second wave of anticipation towards the new planned skydive day and this data would therefore not be directly comparable with that of those who participated in their skydive as planned, or those that did not return.

The pre jump and planned jump days were considered the most standardised in terms of levels of anticipation and this guided the decision to focus the main analysis on these two days: that is, there were no differences between the groups in planned activities on the pre jump day, up until the evening, whereby some were notified of the probability of skydiving the following day. At this point, the derived groups emerge depending on the information given by the instructor: participants were either a) expecting to jump the next day, as planned (the 'jumped as planned' group) b) expecting to jump the next day, but with doubt (the 'anticipated with doubt' group), or c) no longer anticipating to jump the next day (the 'knew they were not jumping the next day' group). In order to assess anticipation of a forthcoming stressor the main analyses will therefore focus on the pre skydive and planned skydive days only. However, a summary of the findings from the complete four sampling days will be provided for comparative purposes only.

### *Statistical analysis*

The effects of the skydive were assessed by conducting repeated measures ANOVAs including six time points (1 hour pre-jump, 30 minutes pre-jump, immediately pre-jump, immediately post-jump, 30 minutes post-jump and 1 hour post-jump) for both cortisol and state anxiety. CAR magnitude was calculated for the pre skydive and planned skydive days and a 3 way (expecting to jump x anticipating with doubt x not expecting to jump) mixed ANOVA was conducted to establish differences in responses across these days. CAR magnitude and  $AUC_G$  were not calculated for participants who failed to provide data for all the time points required to calculate these variables.

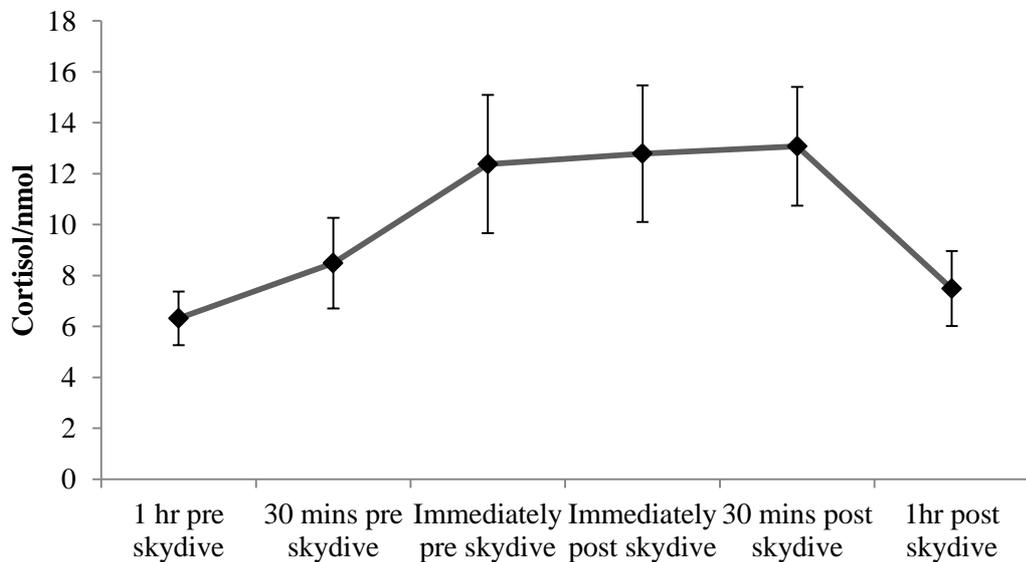
## Results

### *Acute measures (skydive)*

Appendix D presents diurnal data observed for each of the following indices, collected over both the day prior to the planned skydive (day 1) and the day of the planned skydive (day 2).

### *Cortisol*

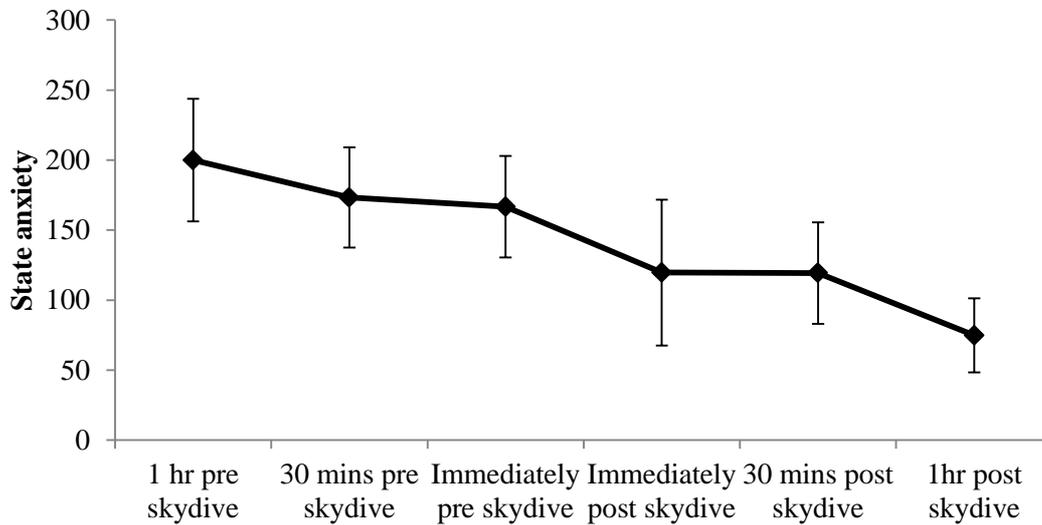
As Figure 7.1 indicates, there was a significant main effect of time, [ $F(5,3) = 22.15, p = .014, \text{Wilks' } \Lambda = .03$ ], with a large effect size, (partial  $\eta^2 = .97$ ). Post hoc revealed a significant increase in cortisol between 1 hour pre-jump and 30 minutes post-jump ( $p = .024$ ) and decreases between; immediately pre-jump and 1 hour post-jump ( $p = .042$ ), immediately post-jump and 1 hour post-jump ( $p = .009$ ), and finally 30 minutes post-jump and 1 hour post-jump ( $p = .004$ ).



**Figure 7.1** Mean (and SE) cortisol secretion during the 1 hour period preceding the skydive, and 1 hour following the event ( $n = 8$ ).

*State anxiety*

There was no significant main effect of time, [F (5,1) = .73, p = .707, Wilks'  $\Lambda$  = .22, partial  $\eta^2$  = .78]. See Figure 7.2.

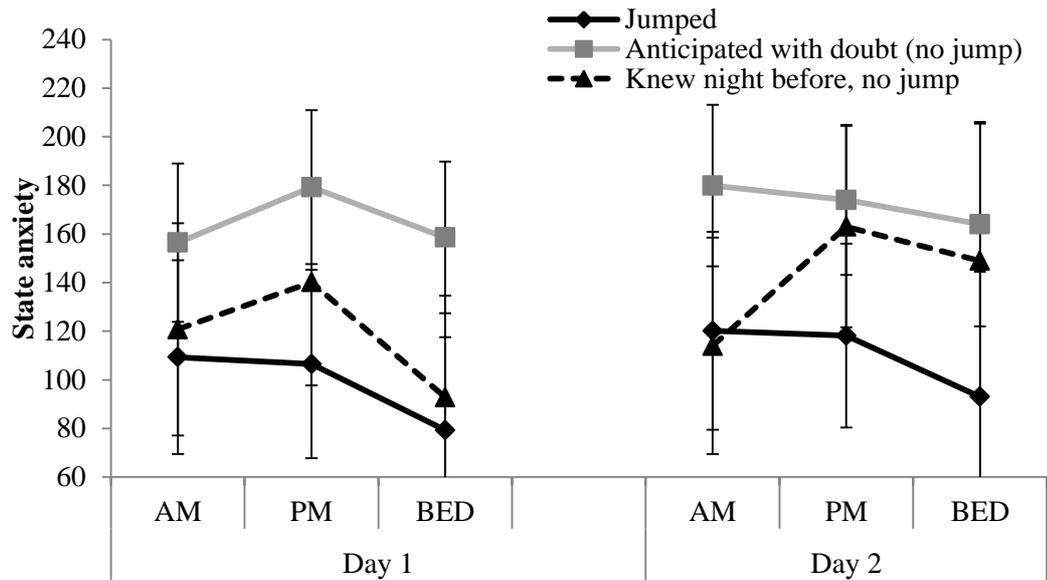


**Figure 7.2** Mean (and SE) state anxiety during the 1 hour period preceding the skydive, and 1 hour following the event (n = 6).

*Diurnal measures*

*State anxiety*

There was no significant effect of day on state anxiety, [F (1,17) = 1.39, p = .255, Wilks'  $\Lambda$  = .93, partial  $\eta^2$  = .08]. There was also no significant main effect of group, [F (1,17) = 1.36, p = .283, partial  $\eta^2$  = .14], and no interaction between day x group, [F (2,17) = .15, p = .865, Wilks'  $\Lambda$  = .98, partial  $\eta^2$  = .02]. See Figure 7.3.

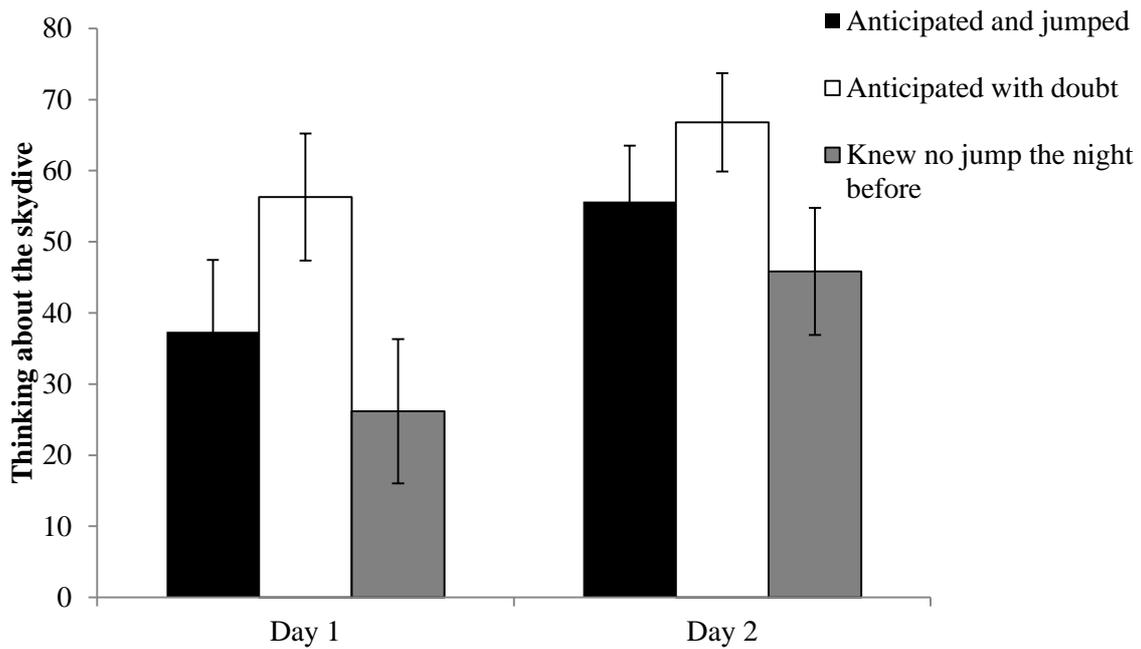


**Figure 7.3** Mean (and SE) state anxiety for the day prior to the skydive (day 1), and day of the planned skydive (day 2),  $n = 20$ .

*Mood following waking*

*Thinking about the skydive*

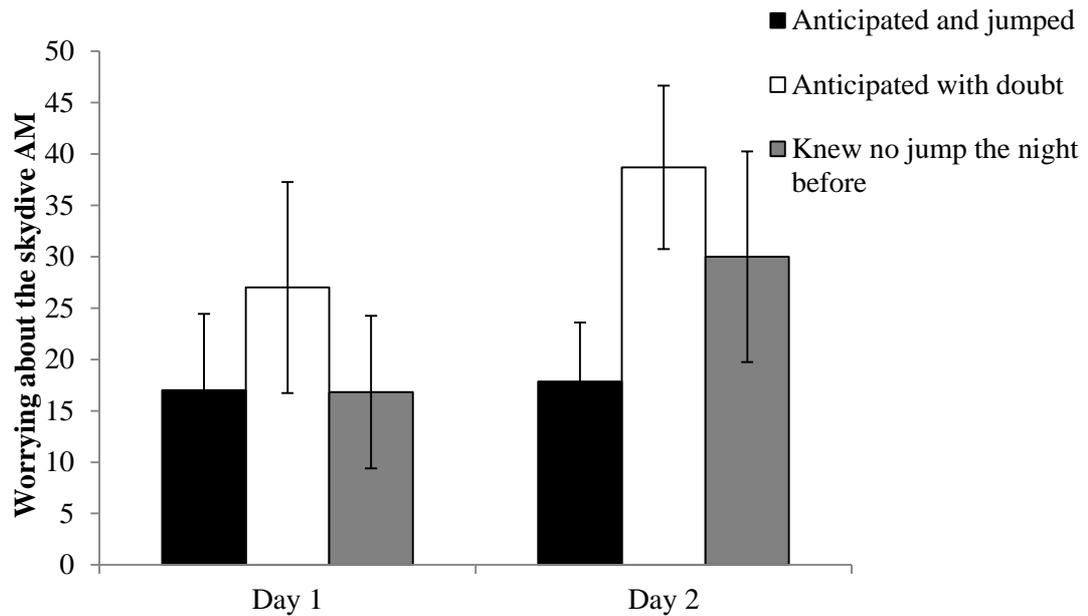
Figure 7.4 illustrates there was a significant main effect of day, with participants reportedly thinking more about the skydive on day 2 than on day 1, [ $F(1,19) = 4.42, p = .049, \text{Wilks' } \Lambda = .81$ ] with a small effect size (partial  $\eta^2 = .19$ ). There was also a significant effect of group, [ $F(1,19) = 5.38, p = .014$ ] with a large effect size (partial  $\eta^2 = .36$ ). There was however, no significant day  $\times$  group interaction, [ $F(1,19) = .16, p = .851, \text{Wilks' } \Lambda = .98, \text{partial } \eta^2 = .02$ ]. Post hoc revealed a significant difference between the ‘anticipated with doubt’ and ‘knew night before no jump’ groups ( $p = .014$ ).



**Figure 7.4** Mean (and SE) scores for ‘thinking about the skydive’, reported in the morning, on the day prior to the skydive (day 1), and the day of the planned skydive (day 2), n = 22.

*Worrying about the skydive*

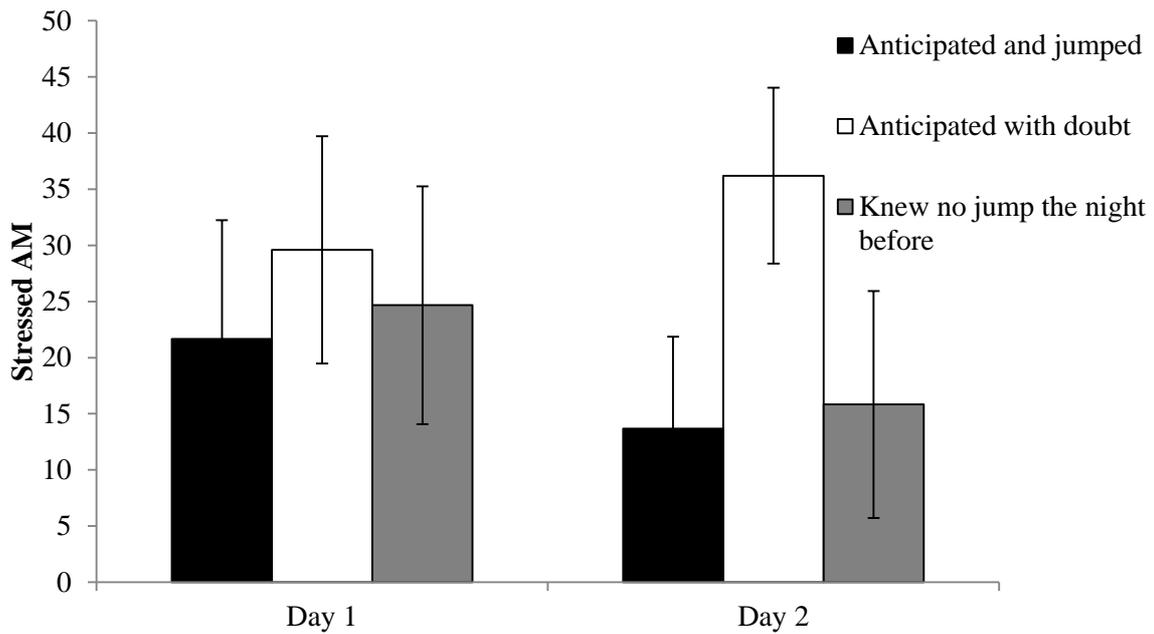
With regards to self-reported worrying on the morning of day 1 and day 2, there were no significant effects observed between; days, [F (1,19) = 1.85, p = .190, Wilks’  $\Lambda$  = .91, partial  $\eta^2$  = .09]; group, [F (1,19) = 1.74, p = .202, partial  $\eta^2$  = .16]; and no interaction, [F (1,19) = .35, p = .711, Wilks’  $\Lambda$  = .97, partial  $\eta^2$  = .04]. See Figure 7.5.



**Figure 7.5** Mean (and SE) scores for ‘worrying about the skydive’, reported in the morning, on the day prior to the skydive (day 1), and the day of the planned skydive (day 2),  $n = 22$ .

### *Stress*

There were no significant differences between the three groups with regards to self-reported stress on the morning of day 1 and day 2 [ $F(1,19) = .30$ ,  $p = .591$ , Wilks’  $\Lambda = .99$ , partial  $\eta^2 = .02$ ]. There was also no main effect of group [ $F(2,19) = 1.23$ ,  $p = .315$ , partial  $\eta^2 = .11$ ], or day x group interaction for this variable [ $F(2,19) = .76$ ,  $p = .481$ , Wilks’  $\Lambda = .93$ , partial  $\eta^2 = .07$ ]. See Figure 7.6.



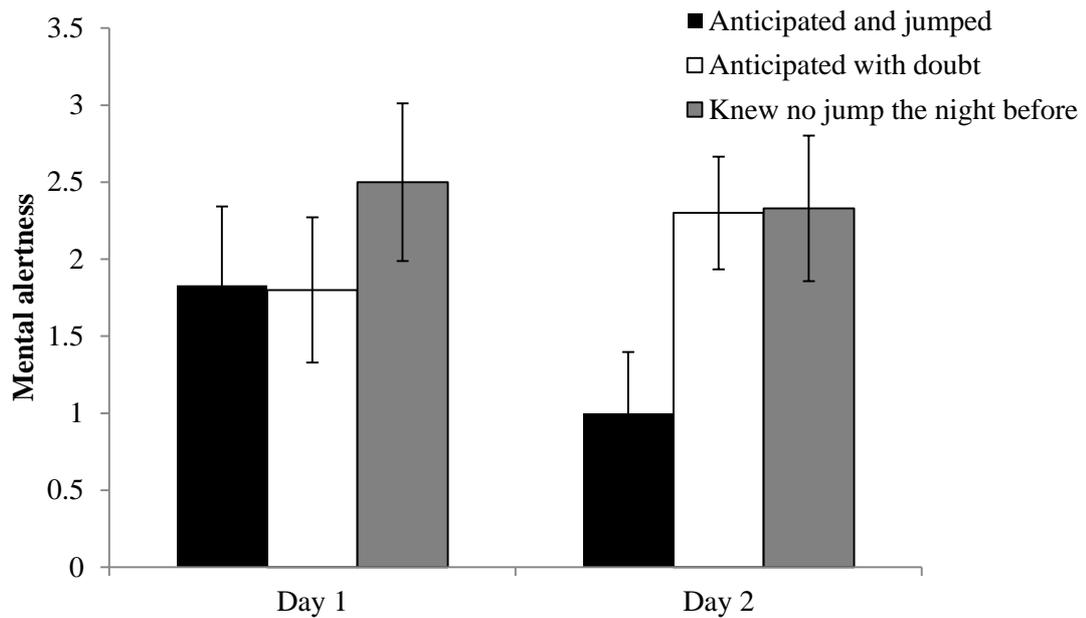
**Figure 7.6** Mean (and SE) self-reported stress reported in the morning, on the day prior to the skydive (day 1) and the day of the planned skydive (day 2),  $n = 22$ .

### *Happiness*

No significant effect of day was observed [ $F(1,19) = 2.83, p = .109$ , Wilks'  $\Lambda = .87$ , partial  $\eta^2 = .13$ ]. There was also no significant main effect of group [ $F(2,19) = .59, p = .565$ , partial  $\eta^2 = .06$ ] or a significant day x group interaction [ $F(2,19) = .05, p = .953$ , Wilks'  $\Lambda = .10$ , partial  $\eta^2 = .01$ ].

### *Mental alertness*

There were no significant differences found for mental alertness on the morning of day 1 or day 2 [ $F(1,19) = .20, p = .658$ , Wilks'  $\Lambda = .99$ , partial  $\eta^2 = .01$ ], and no effect of group [ $F(2,19) = 2.08, p = .152$ , partial  $\eta^2 = .18$ ]. There was also no day x group interaction [ $F(2,19) = 1.19, p = .326$ , Wilks'  $\Lambda = .89$ , partial  $\eta^2 = .11$ ]. See Figure 7.7.



**Figure 7.7** Mean (and SE) self-reported mental alertness reported in the morning, on the day prior to the skydive (day 1), and the day of the planned skydive (day 2),  $n = 22$ .

#### *Physical tension*

With regards to self-reported physical tension on the morning of day 1 and day 2, there were no significant differences [ $F(1,19) = .74$ ,  $p = .399$ , Wilks'  $\Lambda = .96$ , partial  $\eta^2 = .04$ ]. There was a trend for an effect of group, [ $F(2,19) = 3.35$ ,  $p = .057$ , partial  $\eta^2 = .26$ ], but no day x group interaction, [ $F(2,19) = .19$ ,  $p = .830$ , Wilks'  $\Lambda = .98$ , partial  $\eta^2 = .02$ ]. There was however, a significant difference between reported physical tension in the 'anticipated and jumped' and the 'anticipated with doubt' groups, with those anticipating a jump reporting significantly less physical tension than those anticipating with doubt ( $p = .018$ ).

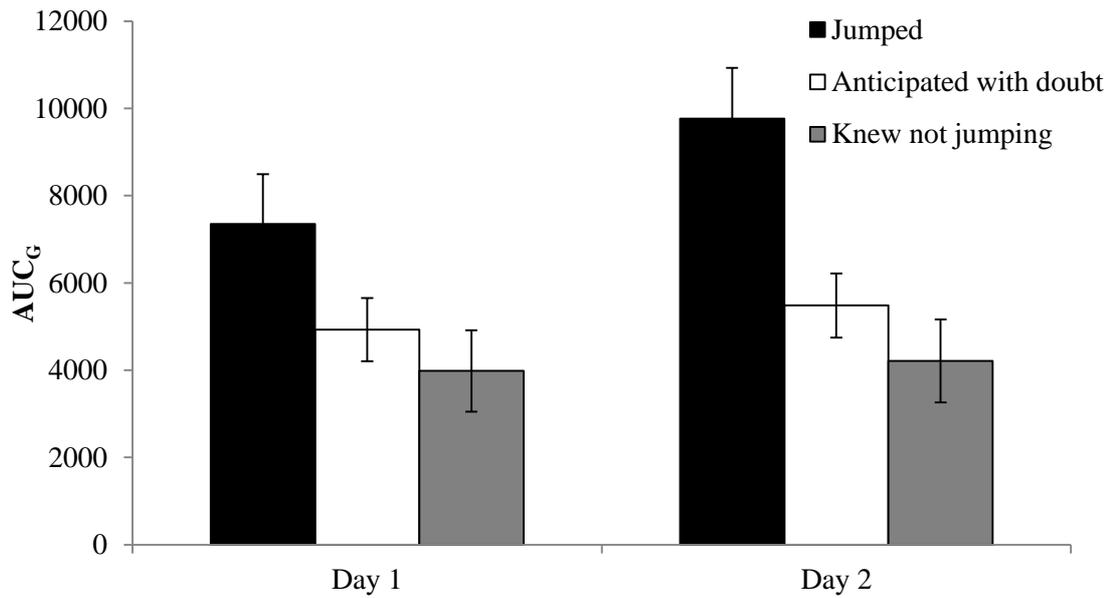
### *Wellness*

For self-reported wellness, there were no significant effects found for day [F (1,19) = .33,  $p = .571$ , Wilks'  $\Lambda = .98$ , partial  $\eta^2 = .02$ ], group [F (1,18) = .93,  $p = .414$ , partial  $\eta^2 = .09$ ] and no interaction between these variables [F (2,19) = 1.24,  $p = .312$ , Wilks'  $\Lambda = .88$ , partial  $\eta^2 = .12$ ].

### *Diurnal cortisol secretion*

#### *AUC<sub>G</sub>*

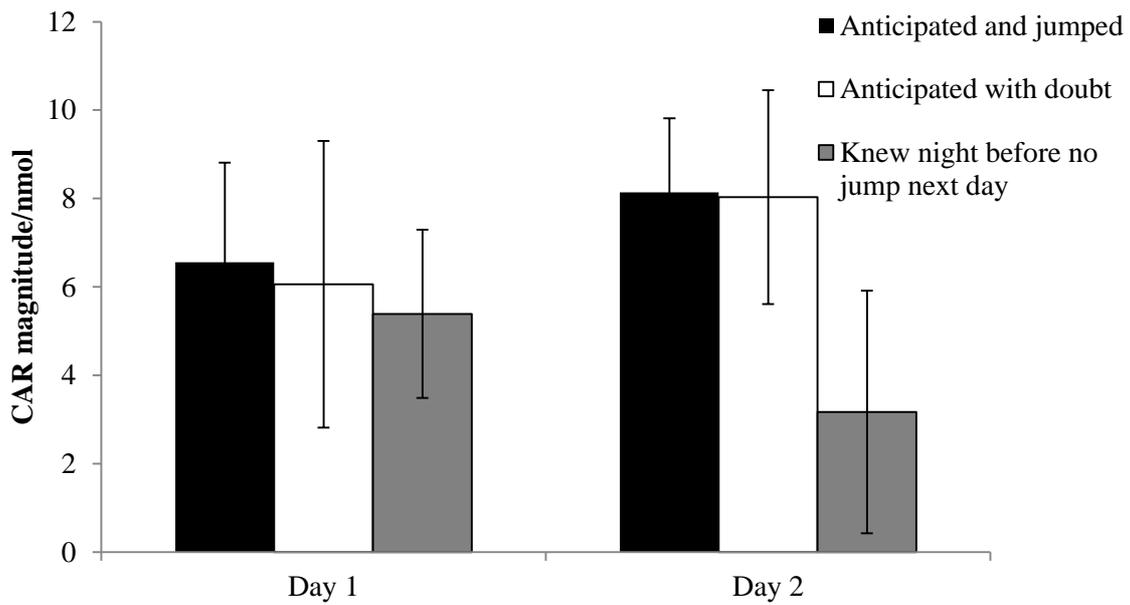
Figure 7.8 indicates a significant main effect of day [F (1,17) = 6.59,  $p = .020$ , Wilks'  $\Lambda = .72$ ] with a large effect size (partial  $\eta^2 = .28$ ), and a main effect of group [F (1,17) = 5.48,  $p = .015$ ] also with a large effect size (partial  $\eta^2 = .39$ ). There was, however, no day x group interaction, [F (2,17) = 2.16,  $p = .146$ , Wilks'  $\Lambda = .80$ , partial  $\eta^2 = .20$ ]. Post hocs revealed that AUC<sub>G</sub> was greater on the day of the planned skydive compared with pre-jump day ( $p = .020$ ). Further, significantly greater AUC<sub>G</sub> was observed in the 'jumped as planned' group compared with those who 'anticipated with doubt' ( $p = .017$ ) and those who 'knew they were not jumping next day' ( $p = .005$ ).



**Figure 7.8** Mean (and SE)  $AUC_G$  observed on the day prior the skydive (day 1), and the day of the planned skydive (day 2),  $n = 20$ .

#### *CAR magnitude*

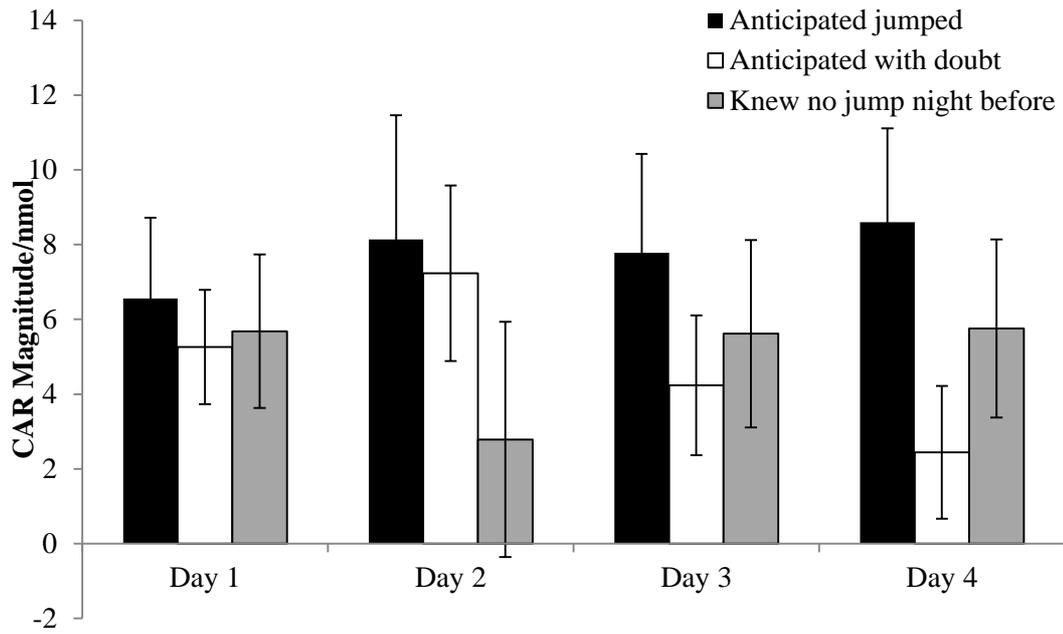
No significant effect of day on CAR magnitude was observed [ $F(1,18) = .06$ ,  $p = .818$ , Wilks'  $\Lambda = .10$ , partial  $\eta^2 = .00$ ]. There was also no main effect of group, [ $F(1,18) = .91$ ,  $p = .420$ , partial  $\eta^2 = .09$ ], and no significant day x group interaction, [ $F(2,18) = .54$ ,  $p = .591$ , Wilks'  $\Lambda = .94$ , partial  $\eta^2 = .06$ ]. See Figure 7.9.



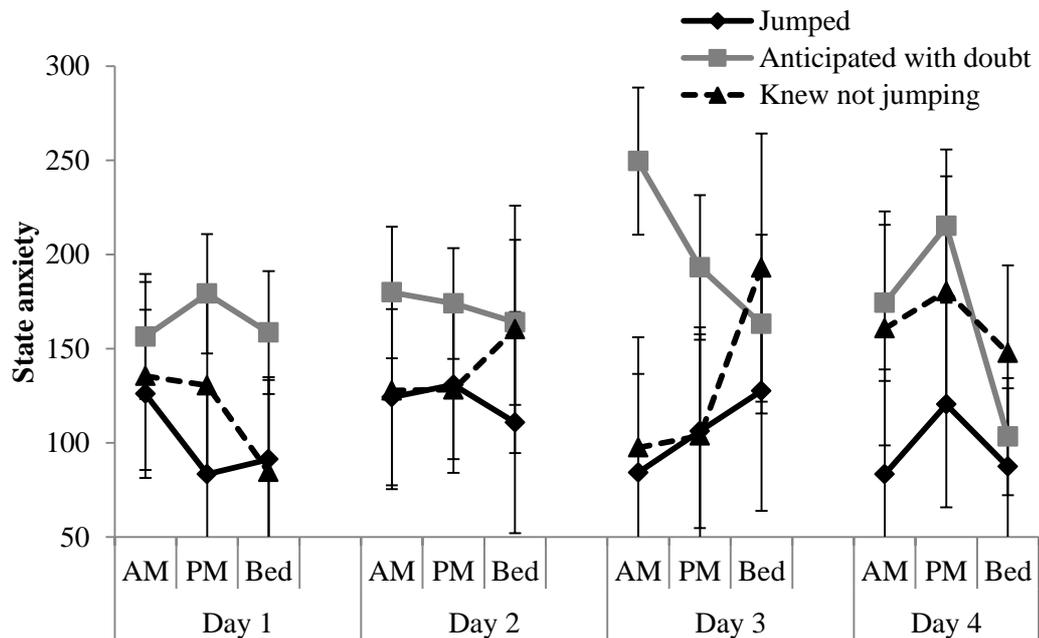
**Figure 7.9** Mean (and SE) CAR magnitude observed across the three groups on the day prior the skydive (day 1) and the day of the planned skydive (day 2), n = 21.

*Results obtained for the full four sampling days*

As previously explained, despite the protocol being delivered as closely to the planned protocol as possible, there were a number of within-group variables from day 2 onwards, that prevented meaningful comparisons between the groups with regard to post day 2 assessments. However, for completeness, this section will include an overview of the data collected over the full four days of the planned protocol. See Figure 7.10 for a visual summary of the CAR magnitude means (and SE), and Figure 7.11 for mean (and SE) state anxiety.



**Figure 7.10** Mean (and SE) CAR magnitude observed across the complete set of four sampling days (n = 21).



**Figure 7.11** Mean (and SE) state anxiety observed across the complete set of four sampling days (n = 18).

## Discussion

Studies that have assessed the preparatory period preceding stressful naturalistic events have interpreted observed stress reactivity as an anticipatory response towards the stressor. Consistencies in reports of higher waking cortisol values observed on challenging or stressful days (e.g., Rohleder, 2007; Wolfram et al., 2013) lend support to the general consensus that this phenomenon serves to prepare individuals for the forthcoming demand of the day. However, few studies have considered the changes which may be detectable between the day of exposure and the day prior to the stressful event.

The aim of the present study therefore, was to assess the psychobiological responses to the anticipation of a skydive by comparing the day of exposure with the day preceding the event. That is, participants were individuals who had volunteered to partake in their first static line (solo) skydive, and thus completed the two days of sampling in proximity of the planned jump. Despite the two planned groups expected to naturally occur as a result of the largely weather-dependent nature of the sport (i.e. those who anticipated and jumped, and those who anticipated yet did not jump), due to external factors the participants were finally categorised into three groups: 1) those who anticipated and jumped (as planned); 2) individuals who anticipated with doubt (and did not jump); and 3) those who knew the night before the planned skydive (the evening of the first sampling day) that they would not be jumping the following day.

Based on the findings of previous studies (e.g., Chatterton et al., 1997; Hare et al., 2013; Meyer et al., 2015), it was hypothesised that there would be significant, rapid increase in cortisol secretion in response to the skydive,

followed by a relatively brisk decline within the hour following the jump. This hypothesis was supported by the findings of the present study, with significant increases in cortisol within the hour before the jump, and clear, marked recovery following the event. This finding is consistent, not only with previous studies with skydivers (e.g., Hare et al., 2013; Meyer et al., 2015), but also demonstrates healthy functioning of the HPA axis, with effective, appropriate increases in response to the stressor, and efficient shutting down of the system once the threat has passed (e.g., McEwen, 1998). State anxiety however, interestingly, did not change significantly during the immediate lead up to or the recovery period following the skydive. The trend for greater anxiety prior to the jump, and a decline following the event, however, is a finding which concurs with those reported in previously studies, indicating a strong anticipatory response immediately prior to completing the skydive, and a brisk decline immediately following the experience (Hare et al., 2013; Chatterton et al., 1997).

With regards to anticipatory responses towards the skydive, it was hypothesised that greater cortisol reactivity would be observed on the day of the planned skydive, compared with the day before the event. This hypothesis was supported by the overall cortisol secretion across the day, with  $AUC_G$  levels being significantly higher in participants who jumped on the day of the planned skydive, compared with the groups who both anticipated a skydive with doubt, and those who went to bed the night before having been told they definitely would not be skydiving the next day. There were, however, no significant differences in CAR magnitude observed across days or groups, despite increased CARs being observed on the morning of other naturalistic stressful events (e.g., in trainee

school teachers on the day of observed classes: Wolfram et al., 2013). Notable differences were observed in the means however, with both those preparing to jump as planned, and those who anticipated the jump with doubt demonstrating greater CAR responses on the day of the skydive compared with the previous day, whereas the group who woke knowing they were not jumping that day had a smaller CAR than the day before. This pattern of results, whilst not significant, is in line with predictions based on previous literature, whereby the removal of threat is associated with the recovery of the cortisol response, represented in this case through the reduction in the CAR. Both the group who were expecting to jump, and the group anticipating with doubt, woke with the knowledge that they may be jumping that day, whether or not there was doubt, and this anticipation of a (possible) skydive appears to have been impacting upon the CAR response.

The second hypothesis regarding anticipation was that the psychological state measures reported across the day would present a profile suggestive of anticipation of a stressful event, including greater reports of state anxiety on the day of the skydive, and mood responses indicative of an anticipatory response (such as thinking more about the skydive, greater levels of stress and so on). There were, however, no significant differences in levels of state anxiety or self-reported mood states of happiness and stress across the two sampling days. Significant differences were, however, observed for measures assessing anticipation of the event; there was a significant effect of day observed for the extent to which individuals reported thinking about the skydive, with participants thinking about the skydive more on the morning of the jump compared to the day before. Interestingly, post hocs revealed that those who were anticipating the

skydive with doubt reported thinking about the skydive more than both those who had no reason to doubt that they would jump, and those who knew they would not jump that day. Furthermore, although not significant, these individuals also reported greater levels of worrying about the skydive than both other groups, as well as greater state anxiety on the morning of the (possible) skydive.

Overall, with regards to addressing the research question, the findings are mixed. No significant differences were observed in CAR magnitude across days, however, this is not sufficient evidence to dismiss a link between the CAR and anticipation of a forthcoming stressful event. Firstly, it must be noted that the sample sizes for each group were small, with only 6 participants actually completing the skydive as planned (i.e. on the second day of sampling), 10 anticipated a jump the following day, with doubt, and 6 knew they would not be skydiving the following day. The greater overall secretion of cortisol observed on the day of the planned skydive in those who completed the jump was expected due to the acute stress reactivity observed in direct response to the skydive. That is, although the cortisol measures surrounding the skydive were not included in the  $AUC_G$  equation, higher cortisol levels prior to and following the acute anticipation and recovery period would contribute to elevated diurnal secretion across the day. This finding supports the notion that cortisol plays a role in the preparation for forthcoming demanding situations.

One of the aims of the study was to assess the recovery period both in relation to a) anticipation, and execution of expected stressor exposure and b) anticipation of a stressor whereby threat was subsequently removed prior to exposure. Unfortunately, due to the considerable variability in the activities

undertaken by participants on the day following the scheduled jump, assessment of the recovery period was not possible or appropriate in the present study. Whilst every effort was made to control the expectancy participants had regarding the probability of jumping the following day (i.e. to ensure that all participants went to bed anticipating a skydive, regardless of the final outcome), influences beyond the control of the researcher presented unexpected variations in expectations for the following day. Parachute Centre personnel made participants aware in cases when the probability of skydiving the following day was non-existent (for example, when a severe storm was forecast), or when it was unlikely, but not impossible that they would be completing their skydive the next day (for example, when the forecast weather conditions were poor, but there was a possibility of suitable intervals for skydiving). This group however, anticipating with doubt, demonstrated further variation, as there was no consistency in the stage at which participants were finally notified that they would certainly not be jumping. For example, in some cases, participants were notified early in the morning (on the day of the planned skydive), immediately following collection of their morning saliva samples; others were told shortly before providing their sample 6 hours post awakening; whilst others were anticipating with doubt until later in the afternoon. In addition, within this group, a small group of participants made the decision on the day of the planned (but aborted) skydive, that they would return to attempt to jump the following day (sampling day 3, originally the planned recovery day). Again, there was considerable variation between these individuals, as some participants made this decision early on in the day (and thus may have been mounting an anticipatory response from this point onwards), while others

made this decision considerably later on in the day, which may have been only shortly before providing their bed time saliva sample. Due to the extensive heterogeneity, occurring from the day of the planned skydive onwards, confounded by a relatively small sample size, the decision was made to focus only on the pre skydive and skydive days, as up to this stage, any observed differences between the days and groups should be due to anticipation of the skydive, with relatively fewer possible confounds.

Whilst the results were not statistically significant, the pattern that emerged for those anticipating with doubt suggests a trend demonstrating that uncertainty is more psychologically taxing than knowing that something stressful will certainly happen (or that it will not). As discussed in Chapter 5, effective anticipation of a forthcoming stressor enables an individual to appropriately prepare for the event and, in some cases, the forward-planning required can lead to complete avoidance of the stressor altogether (Aspinwall & Taylor, 1997; Schulkin, 2011). However, uncertainty regarding whether a future threat will present itself, disrupts this process, diminishing the efficiency of preparation, leading to psychological distress (Grupe & Nitschke, 2013). It can also be argued that uncertainty encompasses a considerable degree of uncontrollability, one of the key stressor domains found to reliably heighten the stress response in Dickerson & Kemeny's (2004) extensive review of stressor paradigms.

Uncertainty involves a considerable lack of control over whether or not exposure to the threat will occur, which could in turn, magnify the perceived threat even further (Dickerson & Kemeny, 2004). The concept that uncertainty is more stressful than absolute certainty of a stressful event has been demonstrated

in previous literature, with a number of studies reporting uncertainty to be a powerful stressor in its own right (e.g., Greco & Roger, 2003). Early studies investigating temporal uncertainty (uncertainty about *when*, not *whether* the event would happen), for example, observed that participants preferred receiving immediate electric shocks rather than unpredictable ones (Badia, McBane, Suter & Lewis, 1966). In another relatively early study, electrical plant workers who were unknowingly exposed to asbestos and were assessed after being notified that they may develop lung cancer were monitored during the period of uncertainty following notification of their exposure (Barak, Achiron, Rotstein, Elizur & Noy, 1997). During the 12-month period following this insight, the workers reported serious psychological anxiety, sleep disturbances and nightmares, and considerable uncertainty about whether or not they would become ill. It was concluded that the extreme uncertainty of illness was responsible for these negative effects on their health and wellbeing. In further support of the concept of uncertainty presenting itself as a stressor, 12 months following notification of exposure, 50% of the sample, had been diagnosed with Post-Traumatic Stress Disorder (PTSD), despite the trauma relating to a (possible) future event, and not a retrospective trauma, as is typically associated with PTSD (Barak et al., 1997). In a similar study, medical personnel who had been exposed to blood or body fluids of Ebola patients were monitored during the period of ‘waiting-to-know’ whether or not they had contracted the disease (Locsin & Matua, 2002). Ebola, which has an incubation period of 3-21 days (WHO, 2016), presents symptoms during this time if a ‘contact person’ has contracted the disease. Due to the nature of the exposure-symptom time frame, during this period, participants were

experiencing high levels of uncertainty, the outcome of which, could present fatal consequences. Emerging themes following analysis of interviews with the participants included those of anticipation, hope, hopelessness and fear, as well as the concept described by the authors as “anticipating death while hoping for life”. Whilst both of these studies represent situations where individuals are anticipating very distressing, potentially fatal consequences, they clearly demonstrate that uncertainty of the most extreme kind can cause more stress than knowing something stressful is certain.

The finding that the potential threat to survival is stressful is not surprising, however, the concept that uncertainty is perceived to be more stressful than the certainty of a unpleasant outcome is a concept that has also been observed under less extreme or severe conditions. A laboratory study seeking to create uncertainty involving the presentation of distressing and neutral images to participants who either were or were not provided with the ability to predict the nature of the next photograph (distressing or neutral), reported increased heart rate in the group without the ability to predict which type of image they would be shown next (Greco & Roger, 2003). This demonstrated increased arousal in response to uncertainty. Furthermore, in a recent study examining event uncertainty (i.e. uncertainty as to whether or not an event would occur) participants were presented with a computerised task whereby they were required to guess whether or not the image of a snake was ‘hiding’ under a rock (Berker et al., 2016). If participants chose a rock that had a snake beneath it, they received an electric shock to their hand. The task was presented in a way which allowed participants to learn which rocks were most likely to have snakes under them,

allowing them to avoid these shocks. However, the odds changed throughout the experiment, and the stimuli were manipulated to provide either 0%, 50%, or 100% chance of receiving a shock. Stress responses were observed through skin conductance (whereby skin conductance tracked uncertainty), state anxiety (with greater anxiety reported by those with a 50% chance of a shock), and pupil dilation, which was used as an indicator of sympathetic stimulation. The findings demonstrated that a 50% chance of receiving a shock was perceived to be more stressful than 0% or 100% chances, supporting previous literature regarding the effect of uncertainty on the stress response.

The present study sheds light on the anticipatory response preceding a naturalistic stressor, and demonstrates the usefulness of skydiving as a model for assessing this response. Despite the unexpected methodological changes which presented themselves in the present study, the unplanned grouping categories which emerged did allow for preliminary analysis of the differences between group profiles in those anticipating a skydive, based on the level of anticipation they were experiencing. The planned protocol aimed to assess and compare both anticipatory and recovery processes in the context of participants anticipating a demand that they would either experience or not experience. Whilst the current study was still able to assess anticipation, the original protocol would have allowed for the additional investigation of the recovery period, to ascertain whether the dynamic of recovery is associated with anticipation, and whether the recovery profile differs when an individual either a) is faced with the anticipated threat, or b) is not exposed to an anticipated threat. Unfortunately, given the small

sample sizes, empirical testing of these differences was not appropriate in this instance; however, such variability in outcomes is typical of naturalistic studies.

The data obtained in the present study are representative of a real-world setting, whereby events cannot be meticulously controlled in the way they can in the laboratory environment. However, the researcher was present to collect samples both on the evening of the first day of sampling, and on the full day of the planned skydive, thus obtaining more control over sampling than is often possible in research of this nature. Whilst a lack of full control is not desirable, this property is also a strength of naturalistic studies, where the data have been collected in as standardised a way as possible, whilst allowing for the flexibility required to assess true behaviour of the individuals being investigated, and the ability to collect ecologically valid data. It is, therefore, clear that increased sample sizes would increase the level of interpretation possible using the current protocol.

An unavoidable, yet important consideration in the present study is that all participants had volunteered to take part in a solo skydive, and therefore the sample recruited for the study was not necessarily representative of the wider population. For instance, people taking part in high-risk activities are often considered high sensation seekers, defined as those with “the tendency to seek novel, varied, complex and intense sensations and experiences and the willingness to take risks for the sake of such experience” (Zuckerman, 1994). As these individuals seek novelty and risk, it could be argued that they do not perceive novel, ‘risky’ experiences as much of a threat compared to those who would instead choose to avoid such experiences.

Another separate, but linked issue is that skydiving is a unique stressor, not only due to its high-risk nature, but also because unlike the majority of stressors assessed in the laboratory, the sport is socially-sanctioned and perceived as an enjoyable activity to those who participate (Franken et al., 2006), rather than one that is unpleasant. Therefore it is possible that the reactivity observed in response to skydiving is more complex in terms of the other possible underlying processes, such as excitement. The impact of anticipating a positive activity on physiological functioning has been examined in very few studies, however, elevated cortisol has been observed in anticipation of Christmas, in children (Flinn, Nepomnaschy, Muehlenbein & Ponzi, 2011), indicating that excitement is capable of eliciting reactivity similar to that observed in response to stressful stimuli. The following chapter will address this topic in greater detail.

### *Conclusion*

Despite limitations relating to sample size, the current study supports previous research demonstrating that skydiving elicits a robust physiological response. In addition, the findings of the present study contribute to the literature regarding anticipation of forthcoming acute stressful events, demonstrating greater cortisol secretion across the day of the a planned stressor, and observing a trend for greater CAR magnitude on the morning of a stressful event which is either relatively certain (i.e. the group who anticipated and jumped as planned) or is anticipated with doubt, compared with knowledge that they will certainly not be faced with such an experience.

As hypothesised, the findings indicate a profile reflective of anticipation, particularly with regards to psychological indices, whereby anticipated

uncertainty regarding the skydive was associated with significantly greater reports of thinking about the study on the morning of the planned skydive, and a trend towards greater worrying, and feelings of stress.

With regards to the planned assessment of longer-term recovery of the stress response (across the following day and a control day later in the week), actual measurement of this effect was not possible, due to the heterogeneity across the sample on the planned recovery day. However, the findings demonstrate the impact of psychological appraisals on anticipation, and could, therefore, suggest that there may also be an influence of appraisals on the dynamic of the recovery period following the elicitation of both the anticipatory and acute stress response. This should therefore be considered and investigated by future studies attempting to measure the full duration of the stress response to a planned event; from anticipation of the event, through to recovery.

## **Chapter 8**

Study 4: Investigating psychobiological anticipatory responses to a positive,  
reward stimulus

## Introduction

The physiological stress response in humans has been well investigated, with a plethora of studies investigating this response in the context of both artificial and naturalistic stressors (e.g., Kirschbaum et al., 1995; Rohleder et al., 2007). In recent years, there have been suggestions of previously undetected, prolonged activation of the acute stress response in healthy individuals, which have arisen from findings that have been indicative of an anticipatory response preceding a forthcoming stressor. The previous two studies therefore investigated these suggestions by assessing the anticipatory response during the period preceding two stressors that are typically associated with stress responding: social evaluation, under laboratory conditions (e.g., Feldman, Cohen, Hamrick & Lepore, 2004) and skydiving (e.g., Hare et al., 2013). Collectively, the findings were indicative of an anticipatory response in the period leading up to the events, and further suggested that this response may be adaptive to the specific resources required for particular activities (based on the type and magnitude of threat).

Whilst there is general consensus that the CAR is an adaptive phenomenon that may serve to prepare the individual for the demands of the upcoming day (Fries et al., 2009), the precise physiological role of this distinct stage of the cortisol circadian rhythm is still relatively unknown. The CAR has been associated with the anticipation of forthcoming events, demonstrating relatively robust associations, although the majority of studies seeking to investigate the role of the CAR in this relationship, to date, have typically done so in relation to adverse, stressful events found to elicit physiological arousal (for review, see Chapter 5).

Due to these observed associations between forthcoming events and the CAR, it has been speculated that the CAR may be associated with prospective memory (Fries et al., 2009), that is, the ability to remember to carry out actions at a later time (Ellis, 1996). This suggestion has been supported by studies examining the CAR in participants preparing for challenging events: for example, competitions (Filaire, Alix, Ferrand & Verger, 2009), and in day to day functioning such as in relation to work days compared with weekends (Kunz-Ebrecht, Kirschbaum, Marmot & Steptoe, 2004). Greater waking cortisol secretion has also been observed on the day of a ballroom competition (Rohleder et al., 2007), further indicating a role of cortisol in the preparation of forthcoming demand. Although support for this concept is found in studies of forthcoming stressful stimuli, support also comes from studies reporting links between CAR magnitude and general cognitive function in older adults (Almela, van der Meij, Hidalgo, Villada & Salvador, 2012) and toddlers (Saridjan et al., 2014). These findings therefore raise the question as to whether the CAR is associated with the memory activation of positive or neutral tasks for the forthcoming day, rather than those perceived as a challenge or threat, exclusively.

Whilst mostly exploratory in nature, a small number of studies have indicated that physiological responses may play a greater role in daily functioning and preparation for forthcoming events than previously considered. It has been suggested, for example, that physiological responses do not only occur in relation to stressful situations, but that they may also be sensitive to social challenges with different affective states, such as excitement and positive affect (Flinn, Nepomnaschy, Muehlenbein & Ponzi, 2011). For example, studies have

demonstrated physiological reactivity in response to positive stimuli including playing computer games (Mounier-Vehier et al., 1995), listening to music (Gerra et al., 1998), and anticipating food consumption (Ott et al., 2011). Computer games have also been observed to elicit increases in cardiovascular reactivity and blood pressure in children and adolescents (Modesti et al., 1994), as well as in adult populations (Mounier-Vehier et al., 1995). Whilst some studies have interpreted this reactivity as a stress response, others have suggested a key role of positive excitement in this relationship (Hebert, Beland, Dionne-Fournelle, Crete & Lupien, 2005).

Ott and colleagues (2011) investigated the effect of food anticipation induced by meal announcement, on cortisol levels. Subsequent to assignment to either the 'anticipation' or 'no anticipation' condition, the anticipation group were told they would be served a breakfast buffet two hours later. Anticipation was induced by preparing the food in front of this group of participants, while the 'no anticipation' group were told they would need to remain fasted until the end of the experiment. Serum cortisol was assessed throughout the testing period, and results revealed an attenuation of the typical diurnal decline in participants anticipating a meal. The findings subtly indicate that positive anticipation of a rich meal increases cortisol levels in healthy adults, although it must be noted that these responses were not as large in magnitude as those observed in response to adverse, stress-inducing stimuli.

In another study, anticipation of a positive event was observed through assessing cortisol responses in children during the Christmas holidays (Flinn et al., 2011). Christmas holidays present a unique and naturally occurring paradigm

whereby children do not attend school, or have school work to complete. This time period is considered one of excitement and anticipation of treats and gifts, making the event a suitable candidate for measuring the anticipation of a day which is regarded as highly positive and exciting for children. Greater cortisol reactivity was observed in the two days preceding Christmas day, compared with typical days. Reports of this research are brief, so the precise methodology adopted for this study is not known; however, the concept of cortisol reactivity observed in response to positive events, has also been reported elsewhere. In a recent study with a more rigorous methodology, CAR magnitude was assessed in children who anticipated receiving a gift in a simple prospective memory task (Bäumler et al., 2014). Children provided saliva samples on two non-consecutive days. They were told by their parents the night before the task day that they would receive a gift the following day, but only if they reminded their parent of this as soon as they woke the next morning. More pronounced CARs were observed on the day that children were required to complete the task, supporting both the theoretical concept of prospective memory associations with the CAR, and also the role of the CAR in relation to upcoming positive events.

Although these findings have not been replicated at this stage, they support the theoretical model suggesting that firstly, physiological response patterns, similar to those observed in response to adverse stimuli, have also been observed in response to positive and neutral stimuli; and second, that the CAR is not exclusively associated with forthcoming adverse, stressful events. That is enhanced CARs may also be associated with positive expectation or excitement,

and could play an adaptive role, not only when under threat, but also in everyday physiology (Clow, Hucklebridge, Stalder, Evans & Thorn, 2010).

The extant findings described so far, whilst novel and relatively exploratory at this stage; therefore allude to adaptive properties of the CAR beyond those which have previously been evidenced. Furthermore, this concept is consistent with findings of generalised emotional responses reported in studies assessing cardiovascular responses to both positive and negative affective stimuli (Waldstein et al., 2000; Warner & Strowman, 1994). Alternatively, these previous findings suggest that cortisol reactivity observed in anticipation of a pleasant stimulus (whether observed via increased CARs or a direct stimuli-induced arousal) may present an altered pattern or magnitude of response, compared to those elicited by negative stimuli; and this is a possibility that also requires investigation.

As highlighted in the previous chapter, prolonged and unnecessary activation of the stress response, and subsequent over-exposure to moderators of the response, is associated with a plethora of negative health outcomes (McEwen, 2003). It remains unknown as to whether cortisol elevations in response to positive stimuli share the same underlying mechanisms engaged in the stress response, or whether positive stimuli elicits similar patterns of anticipatory response in the period preceding the event. It is therefore necessary to explore this concept further, in order to establish the pathways through which this response may or may not occur in healthy adults anticipating a pleasant or neutral stimulus. Investigation of this relationship may serve to provide an indication as to whether there is a requirement for future studies to explore the consequences of both

prolonged reactivity to positive (or neutral) stimuli, and to uncover whether the potential consequences differ from those associated with repeated exposure to adverse and challenging events.

The present study sought to investigate this proposition, by assessing both psychological and physiological responses in anticipation of a pleasant stimulus, a reward. The study adopted a modified version of the protocol utilised in the study by Bäumlér et al. (2014), focusing on the investigation of the response in healthy adults, in order to ascertain whether the response observed in children is replicable in the adult population.

## **Aims**

The aim of the current study was to assess psychobiological responses during the anticipatory period leading up to a forthcoming pleasant stimulus, which required participants to engage with a simple memory task (remembering to send a text message within a specific time frame) in order to receive a ‘reward’.

## **Hypotheses**

Based on the previous reward study in children (Bäumler et al., 2014) which observed greater CARs on task/positive event days, it was hypothesised that participants would have a greater CAR on the day of their task, compared with the other three days of data collection.

## Method

### *Participants*

The sample comprised 45 healthy adults. Three participants failed to return their samples to the researcher and a further participant reported being non-compliant with the morning saliva samples and mood state questionnaires. Their data were therefore removed from the data set for all analyses, leaving data for 41 participants (10 males, 31 females), ranging between 18 and 40 years of age ( $M_{\text{age}} = 26.35$  years,  $SD_{\text{age}} = 8.09$ ). Participants were recruited from the student population at a university in the North East of England, from the general public via posters across the university campus and social media advertisements, as well as via subsequent snowball sampling. Volunteers were screened for the eligibility criteria; including being aged between 18-40 years of age, and confirmation of no current engagement with any of the following: providing longstanding care for another person with chronic illness; managing any long term stressors ; managing serious medical or inflammatory illness; taking steroidal medication; pregnant or currently breastfeeding; currently undergoing (or had recently undergone) hormone replacement treatment.

Ethical approval for the study procedure was obtained from the Ethics Committee of the Health and Life Sciences Department at Northumbria University.

### *Materials*

The questionnaires and saliva sampling procedures used for this study are reported in the general methods chapter (see Chapter 2).

Participants provided saliva samples (Sarstedt, Germany) over 4 days (5 samples per day = 20 samples). They were additionally given a saliva sampling diary to complete as they provided samples, morning mood questionnaires (including questions regarding sleep quality) for each of the 4 days, and finally a questionnaire booklet (see Chapter 2). Participants completed all materials for the study in the domestic setting.

### *Procedure*

Volunteers satisfying the eligibility criteria were invited to take part in the study. All participants provided written informed consent. Participants attended the laboratory for a 10 minute briefing where they were provided with detailed instructions regarding the study procedure, and given the necessary training for the collection and storage of saliva samples. This included a demonstration of how to provide samples using Salivettes (Sarstedt, Germany), and the request that they keep all samples in the fridge until returning them to the researcher at their earliest possible convenience. The importance of adherence to the strict times stated for the collection of saliva samples was emphasised to participants, as well as the behaviours that should be avoided prior to providing samples (including drinking caffeinated, alcoholic or sugary beverages; eating; brushing teeth; using mouthwash; and smoking).

Participants were instructed to choose 2 consecutive days over 2 consecutive weeks (e.g., Tuesday and Wednesday in week 1, Thursday and Friday in week 2) for their data collection days. They were requested to choose days that they would describe as 'typical' for them (i.e. not on a day which was planned to be more eventful than usual). The morning questionnaire allowed

participants to report any unexpected significant events that could have made the day less 'typical' than usual.

On each of the 4 days, participants were required to provide saliva samples at 5 time points (see Table 8.1 for outline of sampling procedure). To maximise adherence and as a means of assessing the timing of samples, participants were asked to complete a saliva sampling diary, where they were instructed to write the precise time at which they provided each sample. They completed a state mood questionnaire and sleep quality diary each morning, as well as further state mood questionnaires, at intervals when they provided subsequent saliva samples (see Table 8.1). In order to aid adherence to the sampling procedure, participants received a text reminder the night before each sampling day, advising them to prepare their samples for the following morning.

During the briefing, in addition to being provided with the materials for the study, participants were notified that on a day during the study period they would receive a text containing instructions for a task they would be required to complete. It was not disclosed to participants at this stage that the task was a manipulation to assess the effect of an upcoming task on the CAR. They were told that the task would be to send a text response to the same phone number at a specific time on the following day (e.g., task text sent on day 1 for completion on day 2). Participants were told that if they remembered to complete this task, they would receive an additional 'bonus' £10 voucher, and would be entered into a prize draw for an iPad mini. The text read "Please text this number saying '£10' between 1400 and 1430 tomorrow in order to receive an additional £10 voucher and to enter the prize draw for an iPad mini. Please note, only texts received

between 1400 and 1430 will be eligible for the additional voucher and prize draw entry. Thank you.” Participants were counterbalanced to receive the task text either in week 1 (day 1) or week 2 (day 3).

On completion of day 4, participants returned the samples to the researcher and were debriefed as to the true aims of the study. They were reimbursed with either a £10 or £20 shopping voucher, depending on whether or not they completed the task. A total of 11 participants did not complete the task; however, two of these participants subsequently did not return their saliva samples or questionnaire data; therefore, nine participants who failed to complete the text task were included in the analysis as non-completers.

**Table 8.1** Study sampling protocol applied for each of the four samplings days.

<i>Time point</i>	<i>Samples and materials completed</i>
<i>Immediately upon waking</i>	Saliva sample 1
<i>+30 minutes</i>	Saliva sample 2
<i>+45 minutes</i>	Saliva sample 3
	State mood 1
	Sleep diary
<i>+6 hours</i>	Saliva sample 4
	State mood 2
<i>Bedtime</i>	Saliva sample 5
	State mood 3

### *Treatment of data*

As with the previous studies in this research programme, some participants did not return any materials to the laboratory ( $n = 3$ ) and one admitted to not completing samples or materials at the appropriate times. In order to maximise sample size, participants were included in analyses for which they had provided a full set of measures. Sample sizes are therefore individually reported for each of these indices.

$AUC_G$  and CAR magnitude values were calculated as described in the general methods chapter (see Chapter 2).

A one-way ANOVA was conducted to assess diurnal secretion of cortisol ( $AUC_G$ ) with day as the repeated measures factor (days 1, 2, 3, and 4). CAR magnitude was also analysed using a one-way ANOVA with day as the repeated measures factor (days 1, 2, 3, and 4).

State anxiety was analysed using a two-way ANOVA with day (days 1, 2, 3, and 4) and time (morning, waking +6 hours, and bed time). Morning state mood and anticipation items (stress, happiness, mentally alert, physically tense, wellness, thinking about the study and worrying about the study) were each analysed individually using one-way ANOVAs with one repeated measures factor: day (days 1, 2, 3, and 4).

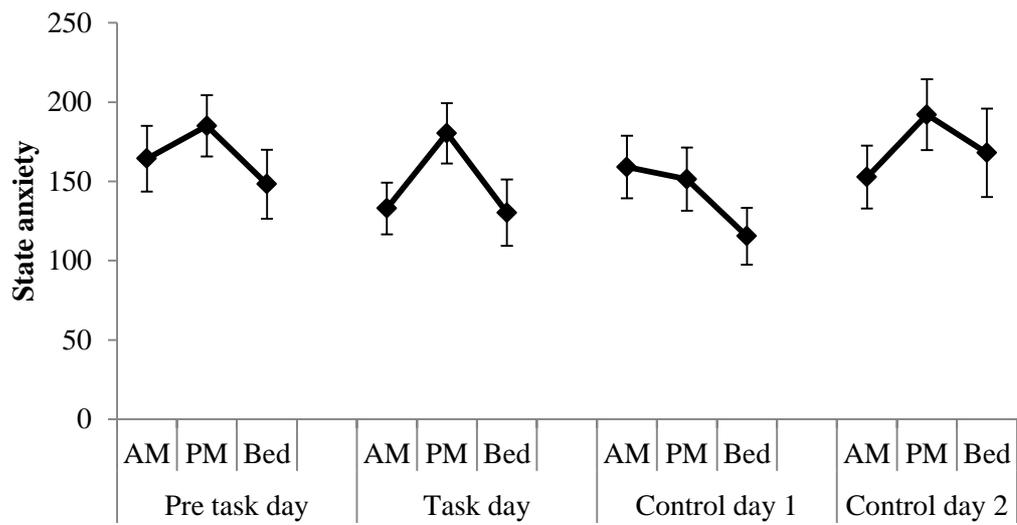
## Results

Mean diurnal cortisol patterns and psychological indices observed across the four sampling days are presented in Appendix E.

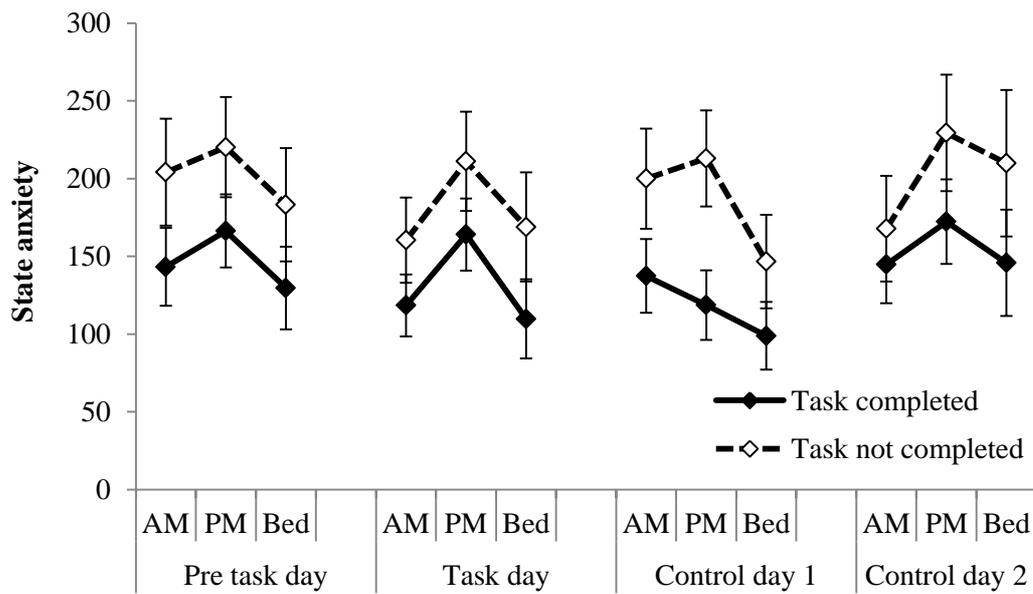
### *State anxiety*

Figure 8.1 indicates there was a main effect of time, [F (2,24) = 6.93,  $p = .004$ , Wilks'  $\Lambda = .63$ ] with a large effect size (partial  $\eta^2 = .37$ ), but no main effect of day, [F (3,23) = 2.16,  $p = .121$ , Wilks'  $\Lambda = .78$ , partial  $\eta^2 = .220$ ]. There was no significant day x time interaction, [F (6,20) = .82,  $p = .567$ , Wilks'  $\Lambda = .802$ , partial  $\eta^2 = .198$ ]. Post hocs revealed significantly greater reports of anxiety 6 hours post awakening compared with waking ( $p = .022$ ), waking compared with bedtime ( $p = .002$ ), and 6 hours post awakening compared with bedtime ( $p = .002$ ).

As not all participants successfully completed the task (i.e. some failed to respond to the text message as instructed), it was deemed appropriate to assess whether there were differences in state anxiety in those who did and did not complete the task (and provided complete state anxiety data). This analysis revealed no significant effects (with the exception of the previously reported main effect of time). See Figure 8.2.



**Figure 8.1** Mean (and SE) state anxiety for days 1, 2, 3 and 4 (n = 26).



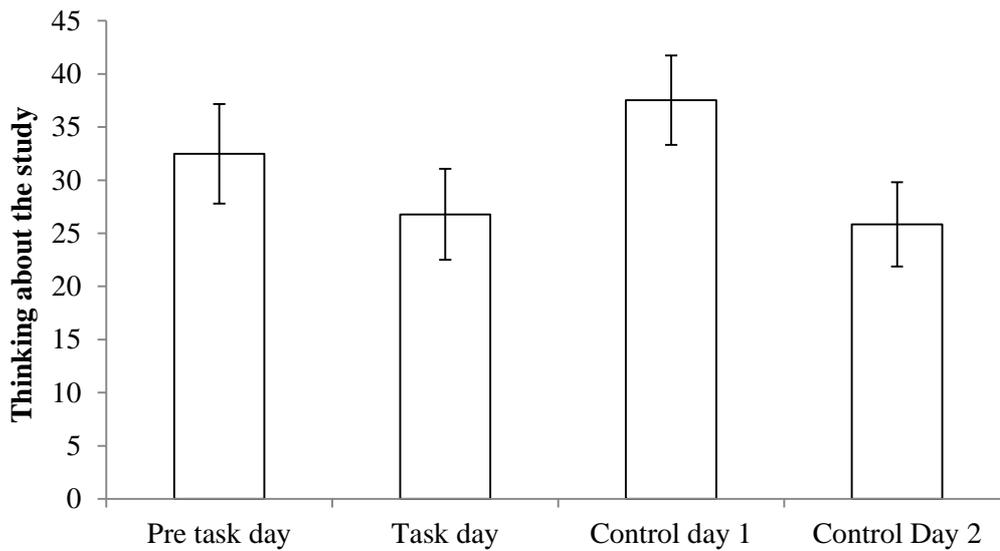
**Figure 8.2** Mean (and SE) state anxiety for participants who did (n = 17) and did not (n = 9) complete the task.

### *Psychological indices*

There was no main effect of day on self-reported morning stress, [F (3,35) = 1.96,  $p = .138$ , Wilks'  $\Lambda = .86$ , partial  $\eta^2 = .14$ ]; happiness, [F (3,35) = .78,  $p = .516$ , Wilks'  $\Lambda = .94$ , partial  $\eta^2 = .06$ ]; mental alertness, [F (3,33) = 1.22,  $p = .317$ , Wilks'  $\Lambda = .90$ , partial  $\eta^2 = .10$ ]; physical tension, [F (3,33) = 1.43,  $p = .251$ , Wilks'  $\Lambda = .89$ , partial  $\eta^2 = .12$ ]; or wellness, [F (3,34) = .85,  $p = .476$ , Wilks'  $\Lambda = .93$ , partial  $\eta^2 = .07$ ].

There was, however, a significant effect of day on the extent to which individuals reported thinking about the study, [F (3,34) = 4.32,  $p = .011$ , Wilks'  $\Lambda = .724$ ], with a large effect size (partial  $\eta^2 = .28$ ). See Figure 8.3. Post hoc revealed a significant difference between the two control days; with greater reports of thinking about the study on control day 1 ( $p = .014$ ). In order to investigate whether these findings were due to the order of sampling days (i.e. whether conducting the task in week 1 or week 2 influenced the results), the data was re-analysed for thinking about the study, comparing participants completing the task in week 1 and week 2. There was no main effect of task week on reports of thinking about the study, [F (1,36) = 3.25,  $p = .080$ , partial  $\eta^2 = .08$ ]. There was additionally no effect of day on reports of worrying about the study, [F (3,33) = .10,  $p = .406$ , Wilks'  $\Lambda = .92$ , partial  $\eta^2 = .08$ ].

Analyses were repeated for all indices to compare participants who did and did not complete the task as instructed, with no effects observed for stress, happiness, mental alertness, physical tension, wellness, thinking about the study, or worrying about the study.



**Figure 8.3** Mean (and SE) scores for ‘thinking about the study’ in the morning, on each of the four sampling days (n = 37).

#### *Cortisol*

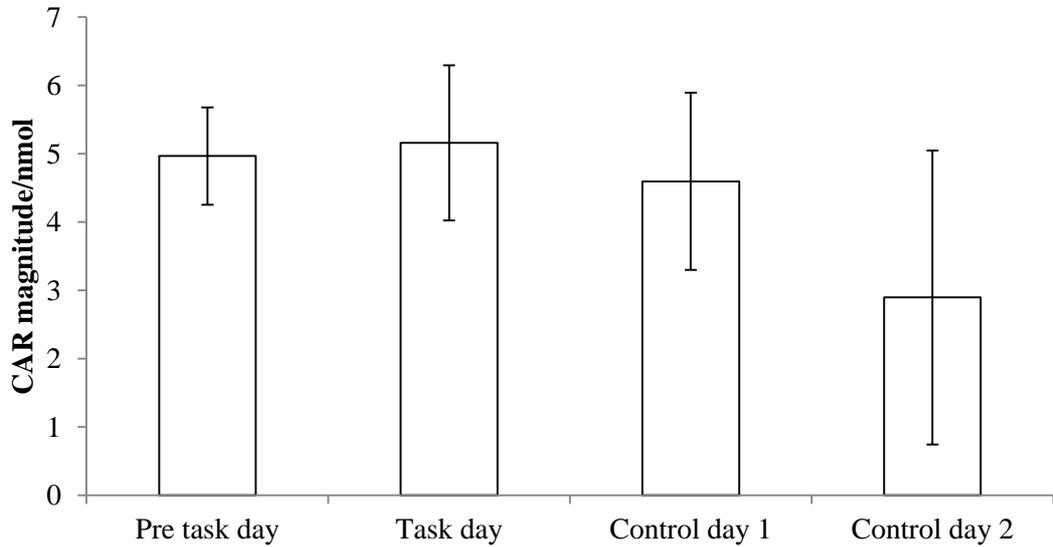
##### *AUC<sub>G</sub>*

There was no main effect of day on AUC<sub>G</sub>, [F (3,22) = .10, p = .959, Wilks’  $\Lambda$  = .99, partial  $\eta^2$  = .01].

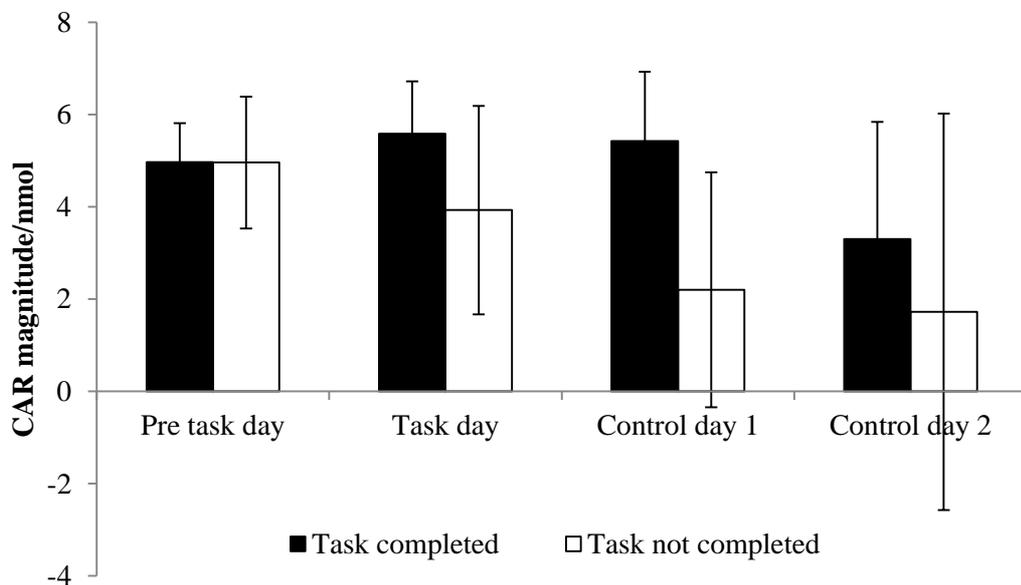
##### *CAR magnitude*

No significant effects of day were observed in relation to CAR magnitude, [F (3,28) = .33, p = .806, Wilks’  $\Lambda$  = .97, partial  $\eta^2$  = .03] (see Figure 8.4). As was the case for the previous psychological indices, participants were compared based on whether or not they had completed the task (for complete cortisol data, those who completed the task n = 23, for those who did not n = 8). This analysis, again, revealed no significant difference between days, [F (3,27) = .37, p = .779, Wilks’  $\Lambda$  = .96, partial  $\eta^2$  = .04]. There was additionally no significant day x

group interaction, [F (3,27) = .35, p = .787, Wilks'  $\Lambda$  = .96, partial  $\eta^2$  = .04] (see Figure 8.5).



**Figure 8.4** Mean (and SE) CAR magnitude for all participants observed across the four sampling days (n = 31).



**Figure 8.5** Mean (and SE) CAR magnitude for participants who did (n = 23) and did not (n = 8) complete the task.

## Discussion

Recent studies have alluded to the possibility that the anticipation of positive stimuli may elicit cortisol responses similar to those observed in response to forthcoming adverse events (e.g., Bäumlér et al., 2014). The present study therefore aimed to investigate this proposition by assessing psychobiological and psychological responses during the anticipatory period preceding a positive reward stimulus. Participants provided saliva samples to assess the CAR and diurnal cortisol secretion over four days: two consecutive days in the first week, and two in the following week. Prior to study commencement, participants were told to expect a text message during the sampling period, requesting them to complete a task the following day, whereby they would be required to send a text to the same number within a specified time frame (30 minutes). They were notified that successful completion of the task would secure an additional £10 shopping voucher and prize draw entry for an iPad mini.

Based on the limited research reporting increased CARs and cortisol responses to positive stimuli, it was hypothesised in the present study that greater CARs may be observed on the morning of the task, compared with the other three sampling days. This hypothesis, however, was not supported by the findings of the present study, with no observed significant effects of sampling day on the CAR or diurnal secretion of cortisol. There were also no significant effects of day on psychological indices assessed in the present study.

Interestingly, whilst not significant, there was a trend suggesting differences between those participants who completed the task and those who did not. Participants who completed the task as instructed demonstrated CARs of

greater magnitude on the day of the task, despite having equal CAR magnitude on the pre task day. This trend was undetectable when analysing the dataset as a whole, and was only visible following separation of participants based on task completion. Participants who completed also had greater CARs on the two control days in the separate week, which could also suggest greater levels of engagement with the study protocol than the non-completers. This is further supported by findings that all four participants who either failed to return samples, or were non-compliant, were in the group who did not complete the task.

Although no statistically significant differences were observed, given the novel nature of this approach, it would be premature to conclude that this response does not feature as part of the day-to-day functioning in healthy adults. Moreover, it is likely that the anticipatory response in the proximal context of positive stimuli may encompass more complex pathways than previously expected. That said, the complexity of the mechanisms underlying the activation and functioning of the HPA axis should not be underestimated, as even in a number of adverse, stressful situations assessed in the laboratory, HPA axis activation is not always observed; and therefore, eliciting the activation of such a response to positive stimuli could be perceived as a somewhat greater challenge. Therefore, as shall be discussed in more detail shortly, it is important to consider that the manipulation utilised in the present study was very subtle in nature, and that the use of a more substantial positive stimulus may be capable of eliciting amplified CAR effects, suggested both by previous studies and by the trend observed here.

This was the first study to directly investigate the anticipatory response preceding a positive reward stimulus in healthy adults. The number of studies that informed the current rationale were limited and were primarily conducted with child participants. There may; therefore, be individual differences pertaining to age-related effects on HPA function influencing the observed results. However, although there is some heterogeneity in the literature, the majority of studies examining differences in sensitivity of the CAR in children and adults have reported no significant effects of age, and therefore any differences relating to age may be present elsewhere in the anticipation process. For example, Christmas, the positive stimuli used in the Flinn et al. (2011) study, is a day which is only experienced once a year, and for children, the day is typically associated with receiving presents, eating special food and playing. Therefore, assessing the anticipation of Christmas in children seems appropriate in order to observe a response to an event which elicits excitement. The expectation of receiving a gift (Bäumler et al., 2014), again, may obtain more excitement inducing properties for a child than receiving an additional £10 shopping voucher may do for adults, as was the reward in the present study.

However, it may well be that the participants were not ‘excited’ by the prospect of receiving a reward, as the incentive was not substantial. In studies on gamblers; for example, where participants are playing blackjack with their own money, significant cardiovascular and cortisol reactivity has been observed (Meyer et al., 2000). Whilst there are a number of possible confounding variables in the relationship between regular gambling and physiology; on a basic level the

findings demonstrate that when the incentive is perceived to be high, situations involving a monetary reward are capable of eliciting a physiological response.

The additional inclusion of the prize draw to win an iPad in the present study aimed to increase the potential incentive to complete the task, as well as the perception of exciting stimuli; however, it is very plausible that this would not induce feelings of excitement of the same magnitude as would be observed in children.

Future research in this area could therefore assess the anticipatory period as conducted here, but use stimuli more robustly perceived as exciting and pleasant in the adult population. For example, individuals preparing to go on holiday report more positive emotions prior to their planned trip compared with controls who were not going on holiday (Nawijn, Marchand, Veenhoven & Vingerhoets, 2010) and therefore an event such as a holiday could provide an appropriate paradigm to enable the assessment of physiological arousal in response to positive stimuli, albeit with some additional methodological challenges.

Age may, however, also be relevant because of potential differences in the way that adults appraise events. That is, if their day to day activities involve multitasking, as is the case for many individuals (Mark, Hausstein & Kloecke, 2008), it is possible that they are able to cope, or may have habituated to the delivery of a number of tasks, and therefore the notion of remembering to send a text message may not be stimulating enough to elicit a response.

This topic is extremely novel and therefore studies such as the present one should be considered as preliminary and exploratory in nature and, as such, the

methodology is therefore open to refinement. The evidence of increased cortisol in anticipation of some of the pleasant stimuli previously discussed also needs to be considered within its limitations. The anticipation of a meal; for example, whilst an interesting finding and one that could suggest cortisol reactivity to positive, as well as negative stimuli, may include additional effects on the brain areas associated with the regulation of energy and glucose homeostasis, and therefore observed cortisol responses may not exclusively be due to anticipation (Ott et al., 2011).

Finally, it should be noted that the majority of participants chose weekdays as their sampling days, with the majority of participants working full time. It is possible that although typical days were chosen to collect the samples, that these days may be characterised by a number of tasks, which could therefore lead to no significant differences being observed between them. It should also be considered that all participants reported using some form of aid to remind them to send the text at the specified time on the day of the task, with the majority of participants reporting that they set an alarm on their phone so they would not forget to send the text message. Whilst setting the alarm indirectly formed part of the task completion, if participants did this the evening before the task day, it is possible that unlike the children in the gift study (Bäumler et al., 2014), participants did not need consciously to remember to send the text, as they knew they would receive a cue to do so at the appropriate time.

Whilst the possible explanations for the null findings have been discussed, these explanations and the findings should be interpreted with caution. The study investigated a very subtle effect, and the findings may, therefore demonstrate the

complexity of mechanisms involved in the pathway between anticipation of positive or neutral stimuli and physiological responses. Alternatively, as speculated earlier in this section, it may also be the case that positive stimuli which are associated with a stronger emotional response than is elicited by gaining a small monetary reward, are required to effectively observe detectable reactivity in anticipation of such situations.

### *Conclusion*

This was the first study to directly assess the anticipatory period prior to a positive reward stimulus in healthy adults. No statistically significant effects of a forthcoming positive task were observed for any of the indices assessed in the present study. However, these findings may indicate the complex processes underlying healthy functioning of the HPA axis, and the preservation of valuable resources. It is likely that the task utilised in the present study did not require physiological arousal, as memory activation may not have been required, perhaps due to the use of aids such as alarms, which were adopted by the participants in the present study. This concept could be further explored with refined protocols involving the control of memory aids and manipulation of more salient positive events. However, although such opportunities do exist (e.g., preparation for holidays), their usefulness in terms of salience and ecological validity must be balanced with the necessity for control and rigour.

## **Chapter 9**

The role of individual differences in anticipatory responses to a forthcoming novel  
event: an exploratory analysis

## Introduction

The concept that individuals often vary in their response to the same experience has been widely accepted in psychobiological research (Pleuss & Belsky, 2013). Not only is there general consensus that individual differences in responses occur, but considerable research has demonstrated that these variations in particular traits are associated with longer term health outcomes, such as burnout (Pruessner et al., 1999) and cardiovascular disease (see Dimsdale, 2008 for review). The diathesis-stress framework asserts that some individuals have a greater vulnerability to the adverse or negative consequences associated with prolonged stress responses than others, due to so-called “vulnerability” characteristics (or a “risk gene”: Belsky & Pleuss, 2009). These vulnerabilities take many different forms: for example, behavioural or temperamental (e.g., highly anxious), physiological (e.g., highly physiologically reactive) or genetic, and individuals with these vulnerabilities are considered disproportionately more susceptible to being adversely affected by environmental stressors (Belsky & Pleuss, 2009).

However, whilst the diathesis-stress framework is a well-investigated and widely accepted model, the main body of literature exploring the relationship between “vulnerability” characteristics and subsequent outcomes has generally focused attention on individual differences in relation to problematic development and poor health outcomes, predominantly investigating vulnerability to adversity.

Although a relatively novel concept, there have been reports of positive experiences (e.g., anticipation of a feast: Ott et al., 2011) also eliciting physiological responses and modifications to the daily functions, such as the CAR

(e.g., in children at Christmas; for short review see Chapter 8). HPA activity has also been associated with positive states such as excitement (Flinn et al., 2011) which conversely, could suggest that if individual differences can influence HPA axis activity in response to unpleasant events, that they may also moderate the relationship between HPA axis activity and those which are appraised as pleasant.

A recent contribution to this theory takes this proposition further, in that it suggests that susceptible individuals are not specifically “vulnerable” to adversity, but more generally “developmentally plastic” (Belsky & Pluess, 2009). Therefore with regards to the diathesis-stress framework, those considered disproportionately likely to be adversely affected by negative experiences may, in addition, be disproportionately likely to benefit from positive ones. This suggestion complies with the concept of investigating not only individual difference variables which may predispose individuals to the negative outcomes associated with some experiences, but also allows for the identification of those which may buffer these effects. It also considers that there may be some individual differences, which in some environments or situations are beneficial, but in others, may be less so. For example, having an avoidant coping style is generally considered a negative vulnerability factor with regards to coping with a stressful event (e.g., Stanton, Danoff-burg & Huggins, 2002); however, in some cases, (e.g., if faced with a predator) an avoidant coping style may be advantageous over a coping strategy which is generally considered to be adaptive, such as a social support seeking coping strategy (Stanton et al., 2002).

Further support for the consideration of novel experiences collectively, rather than focusing exclusively on those which are perceived as threatening,

comes from recent findings regarding the mechanisms influencing the CAR. For example, the CAR appears to be altered by forthcoming demands of the day (Fries et al., 2009), particularly stressful events, and studies attempting to establish the underlying mechanisms in this relationship have speculated that the phenomenon may be associated more generally with the reactivation of prospective memory representations upon waking, rather than specifically adverse events (Fries et al., 2009). That is, that waking is linked with the retrieval of relevant information regarding both self-concept and orientation with time and space, to pre-consciousness, which subsequently activates the HPA axis. The hippocampus has previously been reported to play a key role in the formation of representations of the outside world within the central nervous system (Sweatt, 2004), and this suggests that the hippocampus is also involved with the regulation of the CAR. This theory could explain why greater CARs have been observed on the day of special events as it is possible that the orientation with the upcoming day's activities may itself result in higher peak levels of cortisol during the waking period, as demonstrated on days of both naturalistic (e.g., ballroom competition; Rohleder et al., 2007) and laboratory studies (e.g., Wetherell, Lovell & Smith, 2014). Moreover, the absence of CARs in patients with memory disorders due to frontal lobe and hippocampal region damage (Buchanan et al., 2004; Wolf et al., 2005), despite displaying an otherwise normal diurnal cortisol profile, and regardless of type of damage to the hippocampus (e.g., disease or closed head trauma) also lends support to this theoretical suggestion, demonstrating that prospective memory, in general, influences the function of the CAR.

Studies suggesting that the CAR may be influenced by global activity (i.e. not exclusively by stressful events) could indicate that where previous studies have explored relationships between individual difference factors which may be associated with certain magnitudes or types of arousal in response to stressful events, it may be more meaningful to investigate responses to upcoming novel events collectively. This could be approached by assessing both psychological and physiological reactivity to anticipated non-typical experiences, rather than continuing to focus on negative or unpleasant events exclusively, as this may in turn, shed light on whether there are individual differences in overall functioning of physiological response systems and cognition involved in the appraisal of upcoming events.

Although a number of studies have investigated the relationship between individual differences and HPA function, there is considerable heterogeneity within the literature. A number of individual difference factors have relatively consistently been associated with HPA functioning (e.g., blunted CARs in chronic fatigue), while others have suggested that reactivity is determined to a greater extent by situational rather than trait factors (Hellhammer et al., 2007). It is unclear why there is so little consistency within this literature; however, the heterogeneity could be ascribed to the variety of study protocols and diagnostic criteria utilised in these investigations (Kudielka, Hellhammer & Wüst, 2009). In light of the well-established negative health outcomes associated with dysregulation of the HPA axis (e.g., cardiovascular disease: Dimsdale, 2008), it is important to disentangle these potential relationships, as identification of mechanisms or a phenotype which may serve to either predispose individuals to

these consequences, or buffer against these negative effects is crucial in order to inform and develop potential interventions or strategies which may serve to protect those vulnerable to stress-related ill-health.

The following section provides a brief review of the individual difference factors which have been investigated in relation to physiological and psychological reactivity. Individual difference factors such as sex, memory and health complaints are discussed here, however, the majority of the variables investigated in the extant literature refer to personality characteristics, that is, enduring characteristics which reflect long-term individual differences in emotional style and which influence emotional responses (McCrae & Costa, 1994).

#### *Age*

Aging is associated with global and progressive deterioration of several bodily processes. The HPA axis is among the most essential of the body's endocrine systems and the negative health outcomes associated with the dysregulation of this system have been well-investigated (e.g., McEwen, 1998). Several studies have therefore examined whether aging affects the functioning of the HPA axis, with somewhat mixed findings: similar response patterns have been observed in young (aged 20-29 years) and elderly men (aged 60-76 years) in response to acute stressors, demonstrating efficient activation of the HPA axis in both age groups (Kudielka, Schmidt-Reinwald, Hellhammer, Schurmeyer & Kirschbaum, 2000). Furthermore, with regard to basal functioning of the HPA axis, some studies have observed reduced amplitude of basal circadian rhythm in older adults, compared with healthy younger adults, due to either lower levels in

the morning or higher evening levels (Van Cauter, Leproult & Kupfer, 1996). Lower mean cortisol has also been observed in children aged 7-9 years old compared to 10-12 year olds, and children generally have lower median cortisol secretion compared with adults (Törnbage, 2002). Notable, yet non-significant effects of age on cortisol reactivity have also been observed across age groups, where children (aged 7 to 14 years) demonstrated smaller increases in cortisol secretion (5.63/nmol) throughout the CAR period compared with young adults (aged 19 to 37 years: mean increase = 9.26/nmol) and older adults (aged 59 to 82 years: mean increase = 8.46/nmol). Observations of greater ACTH responses in younger males compared to older men when exposed to the TSST have also been reported (Kudielka, Buske-Kirschbaum, Hellhammer & Kirschbaum, 2004), which could be suggestive of possible age-related variation in the HPA cascade. However, there are inconsistencies across the literature with regard to diurnal secretion, with some studies reporting no evidence that age modifies the circadian rhythm of cortisol secretion (e.g., Seeman & Robbins, 1994). Findings regarding the effect of aging on the CAR specifically are less known: the literature examining this relationship is relatively sparse, though the majority of studies report no significant effect of age on the CAR (Wüst et al., 2000). However, one possible explanation for the lack of consistency across the literature regards the variation in methodology of these studies: for example, Törnbage (2002) assessed cortisol secretion between 0800 and 0900, whereas studies such as that of Van Cauter and colleagues (1996) and Wüst (2000) assessed cortisol in relation to waking, as is typically done so in more contemporary studies (e.g., Stalder et al., 2016). The method of sample collection also varies across studies, with some

studies reporting the use of blood withdrawal to measure cortisol (e.g., Törnåge, 2002), which could potentially increase a cortisol response, whilst others have utilised the less invasive method of saliva sampling (e.g., Wüst et al., 2000). Therefore, the findings reported for aging effects on cortisol secretion should be interpreted in the context of their methodology; as some studies may serve as useful comparisons to others investigating this relationship, yet not be applicable to others which have applied considerably different protocols.

Despite the majority of studies reporting no aging effects on the HPA axis, the significant associations reported in some studies requires consideration: the effects of aging that have been observed in previous work can be interpreted by the glucocorticoid cascade hypothesis (Sapolsky et al., 1986). This model, derived predominantly from animal studies, postulates that age-related alterations to HPA axis function occur in response to a decrease in hippocampal neuron function to obtain sufficient negative feedback. In contrast, and in support of the studies reporting aging effects on physiological functioning, the corticosteroid receptor balance theory (De Kloet et al., 1991) counter argues that adaption of glucocorticoid receptors occurs, even in older age, allowing for the maintenance of efficient, though altered, HPA axis functioning (supported by findings of Kudielka et al., 2004).

Whilst the possible aging effects on the HPA axis appear to require further investigation, the present study is limited with regard to shedding light on this variable, due to the application of strict age criteria across the research programme in order to assess exclusively healthy, younger adults (18-40 years).

### *Sex and gender*

Early studies investigating the effect of gender on the stress response generally speculated that each stage of the stress response is influenced by gender effects, from the perception of whether a situation will be appraised as stressful, to the coping responses employed to manage the stressor, and the following health consequences of stress exposure (Barnett et al., 1987). Differences in gonadal steroid hormone secretion between males and females have been associated with basal and stress-induced activation of the HPA axis, with greater reactivity observed in female participants (Kudielka & Kirschbaum, 2005).

The literature surrounding sex differences in the functioning of the HPA axis is also mixed; some studies report that older women appear to demonstrate greater stress reactivity when exposed to stressors, compared with men (Kudielka & Kirschbaum, 2004). Older women also generally demonstrate greater sensitivity to activation of the HPA axis; however, elsewhere it is reported that greater HPA axis reactivity is observed in men (Uhart, Chong, Oswald, Lin & Wand, 2006). By way of possible explanation, it has been reported that women report more stress than men (McDonough & Walters, 2001), and findings such as these have led to speculation that women may differ in the way they appraise stimuli as threatening or neutral (Ptacek, Smith & Zanas, 1992).

In studies examining sex differences in the CAR, there is relative consistency in the finding that gender influences both the magnitude and duration of the waking response, with similar peak values following awakening, but a delayed decrease in women compared to men (Wüst et al., 2000; Schlotz et al., 2004). However, one review concluded that between puberty and the menopause,

women demonstrate lower cortisol responses and autonomic activity than men, again contributing to the debate regarding both sex differences and possible aging affects (Kajantie & Phillips, 2006).

### *Self-esteem*

Self-esteem is a personality trait defined as the degree to which an individual values and accepts themselves (Pruessner et al., 2005). According to social preservation theory (outlined by Dickerson & Kemeny in their 2004 review), humans have a fundamental need to belong and to be accepted by others. Self-esteem is considered to play a key role in this innate need for acceptance (Leary & Baumeister, 2000) as a 'sociometer', allowing the subjective monitoring of one's own value. Individuals who perceive their value as low, often have low self-esteem, which the theory stipulates, should subsequently motivate behaviour which will increase social inclusion of the desired group. High self-esteem has theoretically been suggested to serve as a buffer against anxiety (Crocker & Park, 2004) and stress (Mruk, 1999), and this concept has been supported in experimental research, whereby greater reported stress was reported by individuals with low self-esteem, in a sample of student nurses (Edwards, Burnard, Bennett & Hebden, 2010). Higher self-esteem also appears to aid adaption to the environment and challenging situations: those high in self-esteem are more persistent in the face of failure (Shrauger & Sorman, 1977) but interestingly, are less so than those with low self esteem when persistence is futile (i.e. the task is unsolvable; Di Paula & Campbell, 2002). Anecdotally, in a study of children during transition from primary to middle school, a life event which is considered a naturalistic stressor (Turner-Cobb, Rixon & Jessop, 2008), it was

observed that whilst academic grades significantly declined during the transition period, those with higher academic self-efficacy (one's belief in their capabilities to successfully complete the required activities: Bandura, 1986) obtained higher grades across this period than their peers who were less confident in their academic ability (Gutman & Midgley, 2000), further suggesting an adaptive property of self-esteem in the face of novelty and challenge. The apparent adaptive function of self-esteem to situations is considered a possible explanation as to how self-esteem and psychological reactivity interact (Baumeister, Campbell, Krueger & Vohs, 2003). Whilst the benefits of higher self-esteem are relatively well-established, low self-esteem, is associated with a number of negative health outcomes; including depression, anxiety (Sowislo & Orth, 2013) and poorer immune functioning (Leary & McDonald, 2003), and whilst the biological mechanisms underlying self-esteem are largely unknown, the personality variable has also been associated with increased reactivity to stressful situations (Pruessner et al., 2005). In terms of general HPA functioning, low self-esteem has been associated with high levels of burnout and high levels of stress; it has therefore been suggested that low self-esteem may pose as a risk factor for the development of burnout (Pruessner, 1999).

The possible pathways through which self-esteem may be linked with stress reactivity are indirectly suggested in Dickerson and Kemeny's (2004) review of stressor properties, whereby they define one of the key characteristics demonstrated to robustly elicit cortisol reactivity as social evaluation. The authors speculate that the potency of this stressor characteristic in eliciting HPA reactivity is due to the previously mentioned need for acceptance and to protect a positive

‘social self’ (self-preservation theory: Dickerson, Gruenewald & Kemeny, 2004). A number of studies have supported this concept and have demonstrated that events which threaten the social self lead to lower self-esteem and increases in cortisol secretion (Gruenewald, Kemeny, Aziz & Fahey, 2004). It could therefore be hypothesised that an individual who already has low self-esteem may respond particularly negatively in anticipation of, and in direct exposure to, a stressor which further threatens their social self.

In a study utilising a stressor paradigm involving mental challenge only (no social evaluation), no cortisol responses were observed in participants with high self-esteem, yet subjects with low self-esteem demonstrated consistently increased cortisol responses to the challenge (Kirschbaum et al., 1995). Not only do these findings highlight a relationship between views of the self and physiological responses, but they could also indicate that low self-esteem is associated with unnecessary or inappropriate activation of the HPA axis (and thus, stress responding), which as discussed in Chapter 1, is linked with a plethora of negative health outcomes (McEwen, 2003)

Literature examining associations between self-esteem and the CAR, again, is limited. However, in a study examining aging effects on cortisol diurnal profiles high self-esteem was found to buffer against aging effects that were present in those with low self-esteem (Pruessner, Lord, Meaney & Lupien, 2004). The low self-esteem group demonstrated a flattened diurnal cortisol profile particularly during the CAR period, whereas those with high self-esteem exhibited a typical CAR profile which did not differ from that of healthy young adults. In line with these findings, an earlier study reported higher basal levels of cortisol in

participants with high self-esteem, which the authors suggest could indicate that high self-esteem serves to reduce stress-responsiveness and may protect individuals from stress-induced over exposure to glucocorticoids (Zorrilla, DeRubeis & Redei, 1995).

In summary, the general consensus with regard to the role of self-esteem is that high self-esteem is associated with positive health and wellbeing and is an adaptive characteristic, whilst low self-esteem is typically associated with poorer wellbeing and a greater number of negative responses and health outcomes.

### *Personality factors*

Personality traits are relatively stable patterns of affect, behaviour and cognition (Fleeson, 2001). The Big Five encompasses five broad domains within which the majority of all trait characteristics can be categorised: extraversion, emotional stability, agreeableness, conscientiousness and openness to experience (Goldberg, 1992). Early studies suggest that high introversion and extraversion are associated with a variation of stress responses (Eysenck, 1967), whereby highly introverted individuals demonstrate higher levels of physiological arousal than extraverts. Introversion is associated with burnout (Cano-Garcia, Padilla-Munoz & Carrasco-Ortiz, 2005) and a generally blunted CAR profile (Hauner et al., 2008), whilst high extraversion is associated with greater perceived coping and happiness (Penley & Tomaka, 2002).

It is suggested that the links between extraversion and reduced reactivity are due to the behaviour associated with those high in this trait; for example, extraverts are generally more sociable and spontaneous and engage more in higher risk situations (Eysenck, 1967). The trait is also associated with sensation seeking

and it has been suggested that this type of behaviour occurs because extraverts seek to increase their arousal (Eysenck, 1967). Introverts, on the other hand, demonstrate over-sensitivity in their stress responses and adopt behaviours such as being socially reserved and generally more controlled, in order to lower their arousal (Eysenck, 1967). It is suggested that these behavioural differences may drive observed differences between reactivity, with extraverts perhaps engaging in more effective coping strategies (e.g., social support; Evolahti et al., 2006) than introverts.

The observed links between extraversion and introversion and biomarkers of stress, such as cortisol secretion over the day and responses to socially evaluative stressors (Ditzen & Heinrichs, 2007), indicate that extraversion may serve as a protective moderator of physiological reactivity, whereas individuals low in this trait may not only experience altered reactivity itself, but may also be predisposed to less positive short and long term consequences (e.g., burnout: Cano-Garcia et al., 2005).

Another of the Big Five personality dimensions associated with cortisol reactivity is openness to experience, defined as “motivated cognitive flexibility” or “cognitive exploration” (DeYoung, Peterson & Higgins, 2005). Low scores in this personality characteristic have been associated with smaller cortisol responses to a socially evaluative laboratory stressor (Oswald, Zandi & Nestadt, 2006), whilst high scores reflect healthy psychological functioning, consideration towards inner feelings, intellectual curiosity and independence of judgment. This trait has been observed to indirectly increase positive affect upon exposure to a stressor, and subsequently reduce the perceived threat of the stimulus (Schneider,

Rench, Lyons & Riffle, 2012). Low scores in this personality factor can therefore be seen to reflect less optimal psychological functioning, and the association of this factor with lower than typical cortisol responses is characteristic of blunted responding, which in turn could be indicative of dysregulation of the HPA axis.

Emotional stability refers to the “tendency to be confident, secure and steady” (Judge & Bono, 2001) and is considered a positive personality trait, with highly emotionally stable individuals being perceived as calm, and reporting few personal worries (Hills & Argyle, 2001). The trait is associated with a number of positive outcomes, such as job satisfaction and performance (Judge & Bono, 2001), happiness (Hills & Argyle, 2001; De Neve & Cooper, 1998), and life satisfaction (De Neve & Cooper, 1998). Neuroticism, or low emotional stability, refers to the tendency to experience strong negative emotions such as anxiety or depression (Portella, Harmer, Flint, Cowen & Goodwin, 2005). Neuroticism is also associated with the tendency to perceive and experience a greater number of stressors, respond more dramatically when they are encountered and take longer to recover following exposure (Suls & Martin, 2005). A number of studies have examined possible associations between neuroticism and diurnal cortisol secretion: some studies have observed greater CARs in high neuroticism individuals compared with those low in the trait, in a small sample of healthy adults (Portella et al., 2005). Higher neuroticism has also been associated with greater cortisol secretion across the day in a healthy sample of adults (Nater, Hoppman & Klumb, 2010). However, there have also been reports of flattened diurnal profiles in male adolescents with high neuroticism (Hauner et al., 2008), and this pattern has also previously been associated with sadness and rumination

in young adults (Kuehner, Holzhauser & Huffziger, 2007). Kueher et al. (2007) suggest that the discrepancies in the literature may be due to differences in the measurement of cortisol, as Hauner and colleagues (2008) defined the slope excluding the CAR period, whilst other studies have included samples which may have included the CAR, with some commencing the slope from 8:00, (e.g., Polk, Cohen, Doyle & Skoner, 2005) and others including the waking sample, a 9:00 sample, and so on (Nater et al., 2010). Collectively these findings demonstrate the effect of neuroticism on the secretion of cortisol in the absence of clinical depression and, whilst the mechanisms through which this trait interacts with HPA axis functioning are speculative at this stage, it is plausible that elevated cortisol levels may be a consequence of experiencing greater subjective stress or anxiety. With regard to stress reactivity, high neuroticism is associated with lower cortisol responses to the Dexamethasone-CRH test (a test used for diagnosis of decreased HPA activity: Pruessner, 1999) compared to individuals scoring low for neuroticism, despite no significant differences in baseline values (McCleery & Goodwin, 2001). It is therefore possible that high neuroticism is associated with a reduction in pituitary sensitivity which could lead to the down-regulation of otherwise typical effect of cortisol release, as speculated by the authors of this study (McCleery & Goodwin, 2001).

Conscientiousness, the tendency to be organised and reliable, is associated with positive health outcomes (Kern & Friedman, 2008) and it has been suggested that low conscientious individuals have a greater proneness to perceive a stressor as demanding while experiencing low perceived coping ability (Penley & Tomaka, 2002). With regard to daily stress, conscientiousness has also

demonstrated protective properties through its influence on coping strategy selection and is associated with problem-focused coping and higher positive affect (Bartley & Roesch, 2011). In an exploratory study investigating this trait and retrospective appraisals of stress, greater conscientiousness was associated with greater ratings of significance of daily hassles (Gartland, O'Connor & Lawton, 2012), which may demonstrate that conscientiousness can have a differential, or adaptive impact on stress-related outcomes.

In terms of associations between conscientiousness and cortisol reactivity, it could be speculated that as high-conscientious individuals are more organised, they may, therefore, manage daily tasks more effectively; subsequently, becoming less distressed by everyday challenges, potentially secreting less cortisol than low-conscientious individuals (Nater et al., 2010). In a study testing this hypothesis, conscientiousness was associated with lower cortisol concentrations, but only when accounting for positive affect (Nater et al., 2010).

Agreeableness, a personality trait which measures positive social adaption and low irritability, has not been widely investigated. With regard to psychological state indices, higher ratings of agreeableness have been associated with lower reported stress (Hao & Long, 2003) and higher positive affectivity (De Neve & Cooper, 1998). In terms of its associations with cortisol reactivity, greater agreeableness has been associated with greater morning cortisol levels (Tops et al., 2006).

### *Type D*

Type D or “distressed” personality (Denollet, 2000) is a relatively novel and somewhat controversial construct that comprises two personality dimensions: negative affect (NA) and social inhibition (SI). Negative affect, characterised by negative feelings such as anger, subjective stress, fear and nervousness (Watson, 2000), has consistently been associated with a number of negative physical health outcomes, such as exhaustion (Thoresen, Kaplan, Barsky, Warren & de Chermont, 2003), poorer immune functioning (Cohen, Tyrell & Smith, 1993) and heightened cortisol reactivity to social evaluation in the laboratory (Habra, Linden, Anderson & Weinberg, 2003). Social inhibition, on the other hand, refers to the avoidance of potential threats involved in social interaction such as rejection; and those high in this trait typically tend to expect negative reactions from others and are often socially isolated (Denollet et al., 2005).

As Type D personality is defined as the interaction of both negative affectivity and social inhibition (Denollet, 2000); the personality trait encompasses the joint tendency to experience negative emotions and to inhibit these emotions while avoiding social contact (Habra et al., 2003). Although significant effects of NA and SI have been observed individually, the combined Type D construct is also associated with a range of health outcomes which are not individually associated with NA or SI: for example, poorer quality of life (Schiffer et al., 2005) and a four-fold increased mortality risk 6-10 years following a cardiac event (Habra et al., 2003). Type D is associated with a number of poor health outcomes, particularly related to cardiovascular disorders and is a strong predictor of death in cardiac patients (Denollet, Pedersen, Vrints &

Conraads, 2006). Due to the well-established relationship between Type D and cardiovascular disease, the majority of studies investigating associations between Type D and physiological function (including basal functioning and stress reactivity) have been undertaken with clinical populations, predominantly cardiac patients. However, in healthy populations Type D has been associated with a greater number of physical symptoms, greater perceived stress and avoidant coping strategies (Williams & Wingate, 2011), demonstrating that Type D does not only present negative consequences to those who have an existing diagnosis of cardiac disease, but also in healthy adults.

It terms of stress responding, Type D has been associated with greater cardiovascular reactivity during exposure to a mental arithmetic stressor (Williams, O'Carroll & O'Connor, 2009). However, other studies have reported 'blunted' cardiovascular reactivity to cognitive stressors (e.g., Kupper Denollet, Widdershoven & Kop, 2013; O'Leary et al., 2013; Kelly-Hughes, Wetherell & Smith, 2014). Further, in response to a mental arithmetic stressor, there have been reports of associations between only SI and elevations in systolic and diastolic blood pressure, but not for Type D (Habra et al., 2003). Additionally, a more recent study reported blunted heart rate responses in heart failure patients upon exposure to a public speaking task which elicited typical stress reactivity in non-Type D heart failure patients (Kupper et al., 2013), whilst others have reported no differences in cardiovascular reactivity to a mental arithmetic stressor in females classified as Type D (Williams et al., 2009). Despite mixed findings, there is now a more general consensus that Type D is predominantly associated with blunted cardiovascular activity. This relationship is speculative due to underlying

characteristics of the personality construct, whereby Type D is associated with high levels of perceived stress, which can subsequently lead to symptomology associated with burnout (Pederson & Middle, 2001), which in turn, is related to blunted physiological reactivity.

With regard to general HPA functioning, reports on associations between Type D and basal diurnal activity are also somewhat inconsistent, although the majority of studies have observed no significant effect of Type D on the CAR, for example, in cardiac patients (Molloy, Perkins-Porras, Strike & Steptoe, 2008). However, in another study, also with cardiac patients, greater CARs were observed (Whitehead, Perkins-Porras, Strike, Magid & Steptoe, 2007). Further, in terms of typical diurnal secretion of cortisol, Type D individuals have demonstrated greater overall cortisol secretion across the day (Molloy et al., 2008), however, in anticipation of daily stressors, an earlier study reported elevations in cortisol diurnal secretion in participants high in NA (one of the two scales forming Type D), compared with those low in the characteristic (Smyth et al., 1998).

In summary, Type D is associated with blunted cardiovascular reactivity to stress, whilst the relationship between this trait and HPA functioning seems less clear, with considerable heterogeneity in the literature to date.

### *Trait anxiety*

Trait anxiety is a characteristic that has also been described as negative affect (Grupe & Nitschke, 2013) and in previous studies, has been negatively associated with happiness and wellbeing (Henker, Whalen, Jamner & Delfino, 2002). Trait anxiety can interfere with emotion processing, whereby highly

anxious individuals encompass a strong sensitivity to threatening information (Fox, 2002). Furthermore, there have been reports of a relationship between anxiety and feelings of uncertainty in anticipation of future negative events (Grupe & Nitschke, 2013). This is a relationship which is adaptive under conditions of healthy functioning, as an appropriate level of anxiety preceding an adverse event aids preparation for coping with the experience (as discussed in greater detail in Chapter 1) and can facilitate the assessment of both the likelihood and severity of the threat. However, excessive anxiety can lead to inappropriate levels of arousal, which, as described in Chapter 1, can be maladaptive; this phenomenon has been proposed as a link between anxiety disorders and excessive anticipatory reactivity to uncertainty of a threat (Grupe & Nitschke, 2013), as discussed in Study 3 (see Chapter 7).

Whilst there is agreement in the literature regarding the impact of anxiety in the formation of appraisals regarding threat, in relation to cortisol reactivity to such experiences, there is currently a lack of consensus across research findings. Some studies have reported decreases in endocrine activity during exposure to a social stressor in highly anxious individuals, suggesting potentially inappropriate responding (i.e. no response) and an inability to elicit a typical response (Jezova, Makatsori, Duncko, Moncek & Jacubek, 2004); while others have found no associations between state anxiety and cortisol reactivity (Takai et al., 2004). However, the types of stressor utilised in these studies must be taken into consideration, as they were considerably different in nature, with Jezova and colleagues (2004) exposing participants to a psychosocial stressor based on the TSST (Kirschbaum et al., 1993), whilst Takai and colleagues (2004) played

participants a video of corneal transplant surgery as their stressor stimuli. In individuals with social anxiety, situations which encompass negative evaluation of their qualities are considered to be threatening (Rapee & Heimberg, 1997). It could therefore be argued that the profile of arousal resulting in response to a psychosocial stressor could differ considerably from the response following viewing a gruesome video, in socially anxious individuals. Not only are psychosocial stressors associated with the greatest stress responses in healthy populations (Dickerson & Kemeny, 2004), but it could be argued that whilst some may be uncomfortable watching a graphic video of surgery, the general population are exposed to material of this nature through a number of media outlets and may therefore be relatively desensitised to such stimuli, thus removing the novelty factor from the paradigm. Therefore, it may well be that responses observed following such a video may represent disgust and not stress, which could explain the differing profiles observed in these two studies. The finding that psychosocial stress can evoke blunted reactivity in anxious individuals further supports previous suggestions that over-activation of stress systems can lead to dysfunction and inappropriate (or absent) arousal. Similarly, with regard to basal cortisol secretion in highly anxious individuals, there is evidence of strong associations between greater anxiety and flattened diurnal profiles, with elevated evening cortisol levels and flatter waking levels compared to those lower in the trait (Van den Bergh, Van Calster, Puissant & Van Huffel, 2008).

### *Coping*

In Chapter 1 the appraisal process undertaken by individuals in the context of a potentially challenging situation was discussed. That is, the concept that

when faced with a potential threat, individuals make an appraisal regarding the abilities to cope with the stimuli. If the individual possesses the resources to cope with the threat, homeostasis is maintained; however, if the resources required exceed those the individual possesses, a stress response follows, which involves the engagement of coping strategies. Therefore, the coping strategy or style attributed to an individual is likely to influence the appraisals made in relation to a stressor and can thus impact upon their subsequent response to a potential threat. In support of this proposition, active coping has been reported to predict physical health in medical students, and seemingly serve as a buffer against the impact of novel stressors (Park & Adler, 2003).

Psychological coping is defined as “the thoughts and behaviours used to manage the internal and external demands of situations that are appraised as stressful” (Folkman & Moskowitz, 2004) and is thus the process which unfolds when the demands of the situation are perceived to exceed the individual’s resources for coping (Lazarus & Folkman, 1984). Coping responses to a threatening situation involve the down-regulation of negative emotions resulting from the initial stress exposure and, as discussed in the anticipation literature review (Chapter 5), negative emotions associated with challenging or threatening events can themselves pose as a stressor in their own right (Ennis, Kelly & Lambert, 2001; Schlotz et al., 2004). Due to the strong links between coping and emotion regulation as well as psychological distress experienced during exposure to a stressor, it is highly plausible that the coping style an individual adopts may also be associated with their physiological stress response. Previous studies have suggested this to be the case, although somewhat similar profiles have been

observed across studies, with blunted cortisol reactivity to stress observed in those with an avoidant coping style (Hori, 2010) and lower overall secretion in healthy older adults with an adaptive coping style (O'Donnell, 2008).

Overall, the literature suggests that coping styles which utilise social support seeking strategies are associated with more adaptive physiological responses to challenge and that avoidant coping styles are associated with less adaptive responses. Adopting an active coping strategy, in relation to cancer treatment specifically has also been associated with positive psychological and physical benefits in child cancer patients (Aldridge & Roesch, 2007). Whilst these findings have also been mirrored in studies assessing general health outcomes, it has been considered that coping should be an adaptive and dynamic process; that is, that one coping style may be appropriate for one stressor but may have a negative impact on another challenge or threat situation (Folkman, Chesney, Pollack & Phillips, 1992; Zeidner & Saklofske 1996). For example; avoidant coping, generally considered a negative coping strategy has been effective in reducing stress when faced with an uncontrollable stressor (Altshuler & Ruble, 1989). Furthermore, acceptance, considered a positive coping strategy in the main body of literature, has been associated with lower reports of wellbeing in children undergoing cancer treatment (Aldridge & Roesch, 2007), providing further support for the suggestion that coping is adaptive, and dependent on situational factors (e.g., Folkman, 1992).

Whilst there is evidence that coping strategies adopted by individuals impact the stress response, evidence as to whether coping influences responses elicited in anticipation of such stimuli has currently not been reported. However,

due to the influence of coping strategies on appraisals regarding the ability to cope with stressful events, it could be hypothesised that coping may also play a role in the anticipation of novel experiences, as it may well be that forthcoming events which are appraised to require resources which exceed those obtained by the individual, may elicit some form of response prior to direct exposure to such an activity. Therefore, this is a possibility which will be explored in the present chapter, in relation to a novel task.

### *Perceived stress*

Perceived stress, defined as the extent to which an individual appraises a situation as stressful (Cohen, Kamarck & Mermelstein, 1983) and more specifically, the degree to which individuals perceive daily events as unpredictable, uncontrollable and overwhelming (Pizzagalli, Bogdan, Ratner & Jahn, 2007), is associated with a number of poor health consequences, including anxiety and depression (Spada, Nikčević, Moneta & Wells, 2008), slower wound-healing following a biopsy (Ebrecht et al., 2004), as well as generally reduced immunologic efficacy (for review, see Lovell & Wetherell, 2011). As well as inducing depressive symptoms, perceived stress is also associated with poorer treatment prognosis and greater likelihood to relapse (Tennant, 2002), and is furthermore associated with incident coronary heart disease (Redmond et al., 2013). Individuals reporting greater perceived stress are generally less happy (Piqueras, Kuhne, Vera-Villaruel, Straten & Cuijpers, 2011) and report lower ratings of wellbeing than those reporting lower perceived stress (Skok, Harvey & Reddihough, 2006). Conversely, perceived threat, which could be argued to relate to perceived stress, has been observed to elicit psychological and cardiovascular

stress responses in anticipation of a psychosocial stressor which involved interacting with a prejudice cross-race individual (Sawyer, Major, Casad, Townsend & Mendes, 2012), indicating a role of the appraisal and perception of stress in the anticipation of forthcoming challenge.

With regards to basal physiological activity, higher perceived stress has been associated with flatter diurnal slopes (Abercrombie, 2004; Lovell, Moss & Wetherell, 2011) and enhanced overall cortisol secretion across the day (Lovell et al., 2011). Blunted CARs have been observed in healthy females reporting high stress, compared with those reporting low stress (O'Connor et al., 2009). Conversely, participants reporting high levels of stress have also often reported higher levels of burnout, which is a condition associated with blunted CARs (Pruessner, 1999), suggesting the possible pathways through which greater perceived stress may be linked with observed dysregulation of the HPA axis.

Overall, perceived stress is associated with internalised disorders (such as depression and anxiety-related disorders) and also appears to share some aspects of the physiological profile of such disorders, including blunted or flattened CARs but also seemingly enhanced diurnal secretion across the remainder of the day.

### *Perseverative thinking*

Perseverative thinking refers to “repetitive or sustained activation of cognitive representations of past stressful events or feared events in the future” (Brosschot, Veruil & Thayer, 2010). Perseverative thinking therefore encompasses negative emotions associated with these characteristics including worry and rumination. The concept of perseverative cognition is based on the theory that lengthened anticipatory responses prior to stressor exposure and slow

recovery following the event, are not driven exclusively by physiological mechanisms, but instead suggest a considerable contribution from psychological variables (Brosschot et al., 2010). This proposition is supported by studies demonstrating considerably faster recovery periods following activities shown to elicit HPA activation but which do not involve psychosocial stress (e.g., Linden, Earle, Gerin & Christenfeld, 1997). The perseverative cognition hypothesis posits that sources of stress only lead to prolonged activation of stress mechanisms when individuals persevere about these events for a long duration; that is, that their mental representations of stressors relating to a forthcoming exposure (anticipation) and during the period following exposure (recovery) are appropriate to the stressor (Brosschot, Pieper & Thayer, 2005).

There is a paucity of studies investigating the effects of perseverative thinking on the stress response; however, it has been suggested as a key mediator in the relationship between stress and its (negative) impact on the body (Brosschot et al., 2005). That is, perseverative cognition serves to maintain cognitive representations in relation to a stressor, which may contribute to prolonged activation of the stress response. It has been argued that perseverative thinking may be responsible for a number of the observed responses reported in studies assessing reactivity to psychosocial stressors, as the responses to such stressors are not generally due to the 'threat' involving physical harm but to the psychological integrity of the individual (the social self; Dickerson & Kemeny, 2004). This therefore indicates that it is the cognitive appraisal of the situation that elicits the ensuing stress response (Frijda, 1988) and not the physical risk presented by the experience itself.

Few studies have specifically examined the relationship between perseverative cognition as a global construct and diurnal cortisol secretion, but one which did investigate this relationship reported no significant effects of the trait characteristic (Zoccola, Dickerson & Yim, 2011). However, worry and rumination reported in the evening (i.e. state worry and rumination) were significantly associated with greater CARs the following morning, and the observation of higher evening cortisol secretion, and a flattened CAR in high-ruminators has been reported elsewhere (Cropley, Rydstedt & Devereux, 2013). Similar findings were reported in a study that encouraged rumination by asking participants to write about a stressful or traumatic event they had experienced (O'Connor et al., 2013). Participants who ruminated (i.e. those in the writing condition) demonstrated greater CARs four weeks later, compared with those who did not ruminate. These findings demonstrate the influence of perseverative thinking on the functioning of the HPA axis and that both trait and state perseverative thinking require further investigation, preferably in relation to anticipated challenge or novelty in order to control the manipulation of anticipatory responses.

With regard to the impact of perseverative cognition on recovery to baseline following arousal, rumination has been associated with slower heart rate recovery following a cognitive stress task (Roger & Jamieson, 1988). This demonstration that negative emotion-induced heart rate increase withstands, even following recovery of negative emotions themselves, has been supported by further studies (e.g., Brosschot & Thayer, 2003). Zoccola and colleagues (2008) also reported significant associations between rumination and increased and

prolonged elevations in cortisol following cessation of a laboratory stressor, strengthening the associations between perseverative cognition and inappropriate stress responding.

### *Sleep and the transition to awakening*

A variety of bodily functions follow circadian rhythms which are associated with normal daily functioning and rest during sleep. The CAR is one of these processes; therefore, due to its occurrence in the sleep-wake cycle it has been speculated that the phenomenon is not only a response to awakening (Wilhelm et al., 2007) but that it serves to prepare an individual for the forthcoming demands of the day ahead (e.g., Fries et al., 2009).

As the CAR has been observed to be so closely related to the transition to awakening, with sharp increases in secretion following waking, peaking 30-40 minutes post awakening, coinciding with the gradual re-establishment of alertness (Balknin et al., 2002), a number of studies have since investigated the effect that sleep may have on the CAR. An influence of waking time, for example, has been observed in relation to the CAR. Edwards and colleagues (2001) observed greater CARs in individuals waking up early; compared with those waking up late, and these findings were replicated in a later study (Kudielka & Kirschbaum, 2003). However, other studies have reported no impact of waking time on the CAR (Wüst et al., 2000). Interestingly, however, another study reported greater CARs on days when individuals woke earlier (on early-shift days), yet also found that controlling for stress and sleep disturbance removed this effect (Williams, Magid & Steptoe, 2005), which provides a plausible explanation for inconsistencies in the literature.

Sleep duration is another variable which has been assessed in relation to cortisol responses in the morning. In an earlier study, higher levels of cortisol were observed in individuals with shorter sleep duration (Spath-Schwalbe, Scholler, Kern, Fehm & Born, 1992); however, elsewhere, no effects have been reported (Wüst et al., 2000). There is also evidence that the quality of sleep may also be important: blunted CARs have been associated with poor sleep quality in primary insomnia patients (Backhaus et al., 2004) and in healthy adults reporting subjective lower sleep quality and mood in the morning following sleep disturbance (Waye, Clow, Edwards, Hucklebridge & Ryander, 2003).

Previous findings regarding the influence of sleep on the CAR are mixed; however, what is clearer is that disruption of usual circadian patterns can cause negative consequences in terms of fatigue and subjective mood and, therefore, the effect of sleep is an important consideration in relation to HPA functioning.

### *Overview*

In light of the findings from the previous studies in this thesis (particularly Studies 2 and 3) that observed an anticipatory response preceding exposure to forthcoming demand, combined with previous evidence that individual differences may alter the stress response, this final empirical chapter will investigate the individual differences discussed, in relation to how they may impact upon the anticipatory response to a forthcoming novel event. The final sample sizes in these individual studies did not permit analyses of these factors with sufficient power (e.g., unequal groups in Study 3) per study, and therefore the present analysis combined the data from Studies 2, 3 and 4 in order to explore factors which may be associated with physiological and psychological reactivity

in anticipation of or during recovery from novel events collectively. The chapter will explore associations during the anticipatory period, between these individual difference factors and HPA axis functioning, as well as state mood, a variable that many previous studies examining individual differences and cortisol reactivity have neglected to consider. Furthermore, in light of previous studies examining and observing anticipatory responses preceding both challenging and positive events, this chapter will assess the proposed individual difference factors in relation to anticipatory reactivity to a general non-typical event (a solo skydive; laboratory stressor; or a simple memory task).

## **Aims**

The aim of this cross-sectional assessment was to explore individual differences in state activity (both psychological and physiological) during the anticipatory period preceding a novel task (stressor or positive memory task), on the day of the task, and on a control day (to assess basal functioning). By combining data collected for the previous three studies (Studies 2, 3 and 4) the analyses aimed to broadly investigate the role of sex, personality traits, trait anxiety, self-esteem, perceived stress, prospective and retrospective memory, coping style, perseverative thinking, and sleep, on both state psychological responses (including anxiety, stress, and happiness) and physiological responses (assessed through assessment of the CAR and overall diurnal cortisol secretion across each day). Whilst the qualitative differences in the stimuli across the included studies vary considerably, the aim of this chapter was not to derive conclusions from the findings, but to indicate individual differences which may, in future, be of interest to explore further in the context of anticipation and recovery from novel stimuli.

## Method

Each of the studies included in the present chapter (Studies 2, 3 and 4) involved the anticipation of a forthcoming, novel event, and obtained detailed physiological and psychological profiles for all participants across each sampling day: the day prior to the novel event, the day of the event, and a control day.

### *Participants*

Data from Studies 2, 3 and 4 were combined, resulting in data for 95 participants being included in the final dataset (35 males, 60 premenopausal females), ranging from 18-40 years of age ( $M_{\text{age}} = 24.48$ ,  $SD_{\text{age}} = 6.51$ ). A total of 31 participants provided data in anticipation of a socially evaluative laboratory stressor (Study 2); 23 provided data during the anticipatory period preceding a skydive (Study 3) and 41 provided data in anticipation of a pleasant stimulus, completing a task to receive a ‘reward’ (Study 4). See Table 9.1.

**Table 9.1** Participant characteristics for individual and combined studies.

	<i>Study 2</i>	<i>Study 3</i>	<i>Study 4</i>	<i>Overall</i>
<i>n</i>	31	23	41	95
<i>M<sub>age</sub>(SD)</i>	24.0(5.18)	21.0(2.41)	26.4(8.09)	24.5(6.51)
<i>Males</i>	10	15	10	35
<i>Females</i>	21	8	31	60

### *Materials*

Materials were standardised across the research programme where possible (see Chapter 2). Details regarding the specific procedures for each of the three studies can be found in the corresponding chapters.

### *Procedure*

The sampling procedure for Study 2 and 3 were almost identical, with sampling taking place over four days (day 1 = pre-stressor day, day 2 = day of stressor exposure, day 3 = recovery day, day 4 = control day), and 6 saliva samples being collected on each of these days (upon waking, +30 minutes, +45 minutes, +1 hour, +6 hours and bedtime). Participants in these studies also provided samples immediately prior to, during stress, and following exposure (although only 8 participants from Study 3 provided these samples).

Participants in Study 4 provided samples across 4 days, which were split into two sets of two consecutive days, a week after the other. On the second day of either the first or second week, participants were required to remember to complete a simple task in order to receive a ‘bonus’ monetary reward (an additional £10 high street shopping voucher) and to be entered into a prize draw to win an iPad, and so this study focused on the anticipation of a positive stimulus. Due to the nature of day 3 differing between Studies 2, 3 and 4, with this day representing recovery in Studies 2 and 3, day 3 was not included in the analysis and analyses therefore included only the day prior to the event, (day 1) the day of the event (day 2), and a control day (day 4).

### *Treatment of data*

Pearson correlations were conducted between  $AUC_G$  and CAR magnitude and each of the following variables: health complaints; extraversion; introversion; conscientiousness; agreeableness; openness; coping style; perceived stress; perseverative thinking; Type D (continuous variable); trait anxiety; self esteem; prospective memory and retrospective memory.

As the anticipation of a stressful/demanding task is likely to elicit different patterns of responding to the anticipation of reward, analyses were conducted separately for the combined data from studies assessing anticipation of a stressor exclusively (i.e. Studies 2 and 3). For this analysis, only state anxiety,  $AUC_G$ , CAR magnitude, and memory indices were included, as whilst the exploration of stressor-only studies was deemed appropriate, the focus of this study is on overall anticipation of a novel event.

A mixed ANOVA was conducted for analyses of sex differences, age and the categorised Type D variable, with repeated factors (day: pre task, task and control).

## Results and Discussion

In the present study individual differences which may play a role in both psychological and physiological (the CAR and daily secretion of cortisol) reactivity during anticipation of a novel event, were assessed on the day before a planned, novel event, the day of a planned event, and on a typical day, to assess basal functioning. Comprehensive psychobiological data was combined from Studies 2, 3 and 4, all of which obtained detailed profiles of diurnal cortisol secretion and state psychological indices across the three days in the anticipatory period preceding a novel event. Due to modest sample sizes across the groups in these individual studies, study-specific examination of individual differences in anticipatory reactivity were not appropriate. However, in order to explore this data, the data were combined to explore individual differences in anticipation of a novel event, rather than exclusively to those perceived as stressful or positive. The anticipation of novel events as a more general category has not yet been explored, particularly not with regards to the role of individual differences and, therefore, the majority of previous work which can provide literary context to the present findings is generally limited to studies which have either examined anticipation of an adverse event (the majority of these studies) or positive events. The findings and reported below, and relationships between pertinent individual difference factors and anticipatory and recovery parameters (with pearson correlations exceeding 0.3) are presented in Tables 9.2, 9.3, and 9.4. Tables including full correlations conducted in the present analysis are presented in Appendix F.

## *Age*

With regards to aging, no significant associations were observed between age and either CAR magnitude or basal cortisol secretion. However, the limited age range in the current samples should be considered and will be discussed shortly.

A very small number of significant associations did, however, emerge following analysis of aging and psychological factors: older participants reported lower ratings of state anxiety on both the evening prior to the task and the evening of the task day. Furthermore, older age (within the present sample age range) was also associated with lower reports of thinking about the study on the morning of the event. These findings could indicate that older adults in the sample appeared to find the anticipation of the novel event less psychologically taxing than younger participants, indicated particularly through lower reports of anxiety the night before and thinking less about the forthcoming event on the morning of the task. These findings could be interpreted as supportive of previous studies which have suggested that older adults report significantly less perceived stress than younger adults (Hamarat et al., 2001) and even more interestingly, that this effect appears to follow a consistent, negative trend, with young adults reporting the most stress, middle aged adults reporting significantly less stress, and older adults significantly reporting the least stress of all (Hamarat et al., 2001).

The age range of the sample recruited for the present research programme was limited to healthy adults, aged 18-40 and, therefore the 'older' end of the scale is not by any means considered to be representative of older populations reported in previous work investigating the effect of age on stress responding.

## *Sex*

With regard to sex differences in cortisol reactivity, no effect was observed on CAR magnitude or basal cortisol secretion. There were also no main effects of sex on state anxiety or stress, thus failing to support the suggestion that women may perceive greater stress than men (Barnett et al., 1987). There was, however, a significant day x sex interaction for mental alertness at sleep onset, with females reporting experiencing more mental alertness at sleep onset the night before their first day of sampling and males reporting more mental alertness the night before the forthcoming event. This may indicate sex differences in appraisals of the upcoming task, supporting suggestions by Ptacek and colleagues (1992) which indicate that females may be sensitive to subtle novelty (i.e. a subtle change in routine on the first day of taking part in the study including the collection of saliva samples), whilst men may respond more to stronger novel stimuli (the knowledge they will take part in an event the next day).

Despite suggestions that women may perceive more stress than men (e.g., McDonough & Walters, 2001), this concept was not supported in the present analyses, which observed no differences between men and women with regard to perceived stress or anxiety. These findings are consistent with the overall conclusions to emerge from the main body of literature in this area, demonstrating that the influence of sex on cortisol reactivity is small (Fries et al., 2009), with the majority of studies observing significant effects of sex, reporting very small effect sizes (e.g., Wüst et al., 2000).

### *Self-esteem*

In the present analysis, higher self-esteem was associated with lower state anxiety and stress at almost all time points, with the exception of the morning of the day prior to the novel event and the evening of the control day. It was also associated with greater reports of wellness on the day prior to the event and the day of the event, as well as with greater happiness on the day of the event. Higher self-esteem was also associated with positive reactivity at sleep onset, correlating with lower physical tension the night before the day of the task and lower mental alertness the night before the control day. Whilst caution must be employed when making interpretations of such large correlational analyses, these findings collectively imply and replicate associations between higher self-esteem and lower anxiety, and stress, previously reported in the literature (e.g., Crocker & Park, 2004); they generally indicate support for the positive benefits of higher self-esteem over low self-esteem (e.g., Leary & McDonald, 2003). Secondly, the present findings indicate that self-esteem does, as suggested in previous work, play an adaptive role in response to the environment and activities (e.g., Di Paula & Campbell, 2002), evidenced here through associations between higher self-esteem and greater wellness on the day prior to the event, and both greater wellness and happiness on the morning of the activity. Furthermore, these findings could tentatively suggest that self-esteem is involved in the anticipation of novel activities, whereby those with lower self-esteem perhaps, as a result of personal lower confidence and value in their abilities (e.g., Leary & Baumeister, 2000), are less happy or comfortable when anticipating engagement with a novel activity, whilst those with higher self-esteem may feel less phased by the novel

activity due to higher confidence in their abilities to complete the task. Overall, in addition to supporting the positive effects of higher self-esteem reported in previous literature, these findings concur with previous suggestions that self-esteem is adaptive and may contribute to healthy appraisals with regards to novelty and challenge.

### *Personality*

A number of significant associations were observed between personality characteristics and state reactivity; higher extraversion was associated with greater reports of wellness on the day of the task and on the control day and was associated with lower evening state anxiety. These findings tentatively support previous literature advocating positive effects of extraversion, whereby the trait has been associated with greater happiness and perceived ability to cope (Penley & Tomaka, 2002). The results from the present analyses could indirectly suggest support for these findings, as one might predict that someone with a greater perceived ability to cope may as a result, also experience lower levels of anxiety, as reported here.

Similarly, emotional stability also had a positive impact on psychological indices; higher emotional stability was associated with lower reported state anxiety for almost every assessed time point, with the exception of the morning of the day prior to the task and the morning of the task. Higher scores for this variable were also associated with greater reported happiness on the day prior to the task, lower self-reported stress and greater reported happiness on both the day of the event and on the control day; there were additional associations between higher emotional stability and lower reported stress, greater happiness, lower

mental alertness and physical tension reported at sleep onset the night before the control day. These findings, as with those of extraversion, support the general consensus that emotional stability is a positive personality characteristic, associated with greater levels of happiness and reporting of fewer worries (Hills & Argyle, 2001), but do not necessarily suggest a role of emotional stability in the anticipation of a novel event. However, it could be argued that the absence of a significant association between emotional stability and lower anxiety exclusively on both mornings of the anticipatory days (i.e. the day prior and day of the task), could still indicate that emotional stability may serve as a protective or ‘buffer’ trait in normal daily functioning, but that the relationship may be more complex in the context of anticipation of a novel event.

With regard to general daily functioning, greater agreeableness was associated with lower state anxiety across almost all time points, with the exception of morning state anxiety on the day prior to the event and the day of the event, and with lower reported stress on the control day. Furthermore, agreeableness was associated with greater happiness, and wellness, on the day of the event exclusively, as well as with lower mental alertness at sleep onset on the same day. These findings are in line with the pattern of associations previously reported between agreeableness and state mood, with significant associations with lower reported stress and higher positive affectivity (De Neve & Cooper, 1998), both of which support positive psychological wellbeing. Whilst the precise mood states reported in the present study differ from those previously reported, it is plausible to speculate that happiness and wellness are linked with positive affect and that reports of lower stress may be linked with previous reports of lower

reported anxiety. Moreover, the associations observed only on the morning of the task indicate that agreeableness may also be involved in the anticipatory response, particularly with regards to greater positive mood preceding a novel activity (observed here as greater happiness and wellness). As suggested following similar state anxiety findings for emotional stability, the observations that greater agreeableness was associated with lower state anxiety at every time point except the morning of the day prior and the day of the event, could further indicate a role of the trait in the anticipatory response, although at this stage this observation is merely speculation and would need further investigation.

Whilst the previously discussed Big Five traits appeared to be involved in the anticipation of a novel event, to varying degrees, greater openness to experience was associated with lower reports of morning state anxiety and greater reported wellness, yet only on the control day, assessing typical daily functioning. These findings not only support previous observations that openness is reflective of healthy psychological functioning but could tentatively indicate that previous suggestions that openness reduces the threat experienced by an individual in the face of a stressor (Schneider et al., 2012) may apply specifically to adverse threats (i.e. a psychosocial stressor), rather than to a novel activity in general. As discussed earlier in the present chapter, openness to experience is not a widely studied trait; however, the presence of these significant associations could suggest that broadening knowledge of the trait could be beneficial in increasing our understanding of the specific influences or roles of personality characteristics in the anticipation of a novel activity.

Most interestingly, greater conscientiousness was associated with greater diurnal cortisol secretion, exclusively on the day of the task. Whilst it was speculated earlier in this chapter that conscientiousness could be associated with lower cortisol secretion due to greater organisation and preparation in these individuals (thus lower stress-related arousal) there are other possibilities that could provide an explanation for the greater cortisol secretion observed in the present study. Conversely, it is possible, for example, that highly conscientious individuals, due to their tendency to be highly organised, and to be task and goal-directed (John & Srivastava, 1999), may be more likely to engage with a task which is novel to them and, therefore, it is possible that elevated diurnal cortisol secretion on the day of a planned event may be reflective of increased arousal in response to novelty. This finding differs from the results observed in the only known study examining the association between this personality characteristic and diurnal cortisol secretion, where high-conscientiousness was associated with lower overall daily secretion (when accounting for positive affect); however, the previous study exclusively assessed typical daily functioning and not anticipation of a novel event, as investigated here. It is therefore plausible to consider that conscientiousness may in fact have a greater influence over physiological functioning than previously comprehended and that whilst it may be associated with lower diurnal secretion of cortisol in the context of typical daily functioning (Nater et al., 2010), this profile of reactivity may be sensitive to alterations in activities, or anticipated activities, which deviate from those typical to the individual. More specifically, the findings suggest that conscientiousness is involved in the anticipation of a novel event and, that as suggested earlier, this

observed greater cortisol secretion on the day of a planned novel activity could serve to prepare the individual for anticipated engagement with a forthcoming demand. Whilst some literature has suggested increased cortisol secretion to be indicative of stress, or other forms of negative affect, the additional positive associations observed between conscientiousness and state indices lend support to the interpretation of greater cortisol reflecting a positive response in this instance: an association between greater conscientiousness and greater happiness on the day of the task and between greater conscientiousness and lower stress, greater wellness and lower state anxiety both on the morning and evening of the control day. It can therefore be considered that the observed association between greater conscientiousness and greater cortisol secretion on the day of a planned, novel event represents healthy functioning and increased arousal to meet the demands of the activity. Whilst there have been a small number of reports of less positive associations between conscientiousness and psychological wellbeing; for example, an association between high conscientiousness and greater appraisals of daily hassles (Gartland, O'Connor & Lawton, 2012), the associations reported here, combined with the plethora of studies demonstrating the positive influence of the construct, withhold the general consensus that conscientiousness is associated with a more positive health status (Goodwin & Friedman, 2006). Furthermore, the present findings regarding anticipation suggest that conscientiousness is a complex construct which interacts with environmental demands, which may mediate the relationship between appraisals of anticipated activity and subsequent response. However, as this analysis was correlational, there is, at this stage, no means of interpreting cause or consequence in this

relationship. Further work is recommended to establish the robustness of this association and to further uncover the causal direction in this very interesting, suggested relationship.

#### *Type D*

When assessed as a continuous variable Type D was associated with state anxiety at all but one of the time points, the morning of the control day. Greater Type D was also associated with greater reported stress and lower reported happiness on each of the sampling days, as well as with lower reported wellness on the day prior to, and of the event. Higher scores for Type D were also associated with greater reports of thinking about the event on the morning of the task and with lower physical tension at sleep onset the night before the event, as well as with lower mental alertness at sleep onset the night before the control day. When Type D was calculated as a dichotomous variable, a near significant trend emerged between Type D and smaller CAR magnitude across all three sampling days. With regards to psychological state indices, there was a main effect of Type D on both state anxiety and stress, with greater anxiety and stress reported by Type D individuals at all time points. There was also a main effect of Type D on reported happiness and wellness, with lower scores for both indices reported by those meeting the Type D criteria.

The overall pattern of results observed for Type D supports previous observations that Type D is associated with greater general distress, including anxiety, and stress. These findings could further indicate the pathways through which Type D is associated with negative health outcomes (e.g., burnout: Pederson & Middle, 2001) as greater perceived stress may lead to inappropriate

responding (i.e. through recurrent or unnecessary activation, or failure to recover following the removal of threat).

The trend between Type D and smaller CAR magnitude was, although not significant, a notable finding, particularly as previous studies have reported mixed results, with greater CARs being observed in Type D cardiac patients previously (Whitehead, Perkins-Porras, Strike, Magid & Steptoe, 2007), while other studies have reported no effects (Molloy, Perkins-Porras, Strike & Steptoe, 2008). Furthermore, the present findings could allude to a possible anticipatory effect of Type D in response to a novel event, with lower reported wellness on both anticipatory days (i.e. the morning of the day prior to the task, and the morning of the event) but not on the control day, as well as greater reports of thinking about the activity on the morning of the event. These exploratory findings should be interpreted as indicative of a potential role of Type D in the anticipation of novelty and challenge. However, further work is required in order to fully examine the potential influence of Type D under such circumstances.

#### *Trait anxiety*

As might be expected, greater trait anxiety was associated with higher levels of state anxiety at all reported time points. It was also associated with lower reported happiness on the task day and control day, as well as lower reported wellness on the day prior to, and the day of the task. On each of the three days, higher levels of trait anxiety were also associated with greater levels of stress, and on the control day, higher trait anxiety was additionally associated with greater physical tension and mental alertness when attempting to sleep the night before.

With regard to general functioning, the relationships between greater anxiety and greater stress reported on each of the sampling days, and lower happiness reported on the task and control day support previous findings in the identification of high anxiety as having a general negative impact of psychological health and wellbeing (Henker, Whalen, Jamner & Delfino, 2002).

In relation to anticipation-specific findings, individuals with greater anxiety generally reported less wellness on the morning of a novel event, which again, indicates that anxiety negatively effects psychological wellbeing. Furthermore, as this was the only significant association observed exclusively in anticipation of the novel event, it may well be that anxious individuals are more chronically anxious and stressed, as suggested through the consistent findings for these measures across all three sampling days.

The absence of a significant association between trait anxiety and cortisol reactivity is consistent with previous studies and can, in light of the consistently greater stress and anxiety being reported by these individuals, suggest that either a) these individuals are demonstrating dysregulation of the HPA axis through an apparent absence of response or b) that this may be an example of a lack of concordance between psychological and physiological reactivity (e.g., in experienced skydivers; Hare et al., 2013).

The findings reported here concur with those of previous studies examining the role of anxiety in reactivity to stress, although interestingly, the present analysis included general events (i.e. not only stressful events) and, therefore the findings that responses to stress and neutral novel stimuli do not

appear to differ, supports the concept that anxious individuals may be chronically stressed, irrespective of the nature of daily activities.

### *Coping*

On the day prior to the task, greater engagement in behavioural disengagement was associated with lower diurnal cortisol secretion, as was greater acceptance. Greater engagement of positive reframing was associated with greater diurnal cortisol secretion on the day of the task, whilst a greater report of substance use was associated with lower diurnal cortisol secretion, as was greater emotional support on this day. Higher ratings of acceptance as a coping strategy were associated with greater diurnal secretion across the day of the event, as was the case for engagement in greater emotional support seeking on this day. On the control day, however, greater reported emotional support seeking was associated with greater CAR magnitude and furthermore, active coping was associated with greater diurnal secretion.

The observation of an association between acceptance and greater diurnal cortisol secretion only on the day prior to and the day of the task and not the control day, could indicate that acceptance, considered a positive coping style (e.g., Park & Adler, 2003) and one which has been implemented in interventions to reduce avoidant coping behaviour with regards to improving health and wellbeing (Forman, Butryn, Hoffman & Herbert, 2009), may increase arousal by addressing the forthcoming novel event, that is, to aid appropriate preparation for the experience. These findings could therefore be indicative of a positive role of acceptance in anticipation of a novel event. However, these findings are not consistent with previous reports of associations between cortisol and active

coping styles, with previous work observing lower diurnal secretion in those with a greater sense of control (Vedhara, Miles, Sandermand & Ranchor, 2006). Greater levels of cortisol secretion, on the other hand, have typically been associated with avoidant coping strategies (e.g., Rosenberger et al., 2004). The present finding may seem inconsistent with the notion that acceptance is a positive coping strategy, especially as lower cortisol output across the day has been associated with greater psychological wellbeing (Lindfors & Lundberg, 2002). However, whilst excessively high levels of cortisol secretion have been associated with physiological dysfunction, low or blunted cortisol has also reliably been associated with a plethora of clinical conditions (e.g., depression: PTSD: Gill, Vythilingam & Page, 2008). At this stage, optimal salivary cortisol levels have not yet been established and, as suggested earlier in this thesis, under acute conditions cortisol serves an adaptive function and facilitates arousal and focused attention. It may therefore be prudent to consider that the association between acceptance (and other positive, active coping strategies) and greater cortisol secretion on the day of an anticipated novel event may be due to increased engagement with acceptance of the forthcoming task.

The observation that greater substance use and emotional support seeking were associated with lower diurnal cortisol secretion on the day of a novel task is somewhat puzzling; based on the positive affect associated with emotional support seeking and less positive avoidant/distraction behaviour of substance use, one might expect there to be differences in the patterns observed in response of these coping strategies. The association observed between higher emotional support seeking and lower cortisol secretion supports the findings from O'Donnell

and colleagues (2008), where adaptive coping styles were associated with lower overall secretion in healthy older adults but does not extend to support the findings reported for substance use. However, as alluded to earlier in this chapter, there is a paucity of studies thoroughly investigating cortisol reactivity and diurnal secretion in relation to coping styles and, therefore, establishing whether these findings support previous literature is complex. That said, the apparent inconsistencies observed in the present analyses have been reported in previous literature; for example, avoidant coping strategies have been associated with psychological dysfunction (e.g., Higgins & Endler, 1995) and with blunted cortisol reactivity to stress (Hori et al., 2010), whilst lower overall secretion has been observed in healthy older adults with an adaptive coping style (O'Donnell et al., 2008), considered to be more positive with regards to health outcomes. It could therefore be suggested that in anticipation of a novel event, avoidant coping strategies may be associated with blunted reactivity in anticipation of a novel event (as observed in profiles of reactivity to stress: Hori et al., 2010) due to dysfunction of physiological systems involved in arousal. However, a more simplistic, yet possible explanation for this association could alternatively be that avoidant coping, such as substance use (a distraction technique), may merely be associated with lower diurnal secretion due to these individuals failing to engage with the forthcoming task (or, as the name of the coping style suggests, that they avoid such engagement), therefore eliciting no response. Although not recorded in the current analyses, the type of substance use may also be a consideration. That is, chronic use of ecstasy / MDMA has been associated with dysregulated diurnal cortisol secretion (Wetherell & Montgomery, 2014) and cannabis and

MDMA use has been associated with increased levels of anxiety and dysregulated psychobiological responding to stress (Wetherell et al., 2012).

The observation of more exaggerated CARs on the day of an event has been speculated to serve to prepare the individual for the forthcoming demands of the day, with cortisol facilitating a state of enhanced arousal (Schulz, Kirschbaum, Pruessner & Hellhammer, 1998). In the present analyses, with regards to coping styles, only one significant correlation was observed relating to CAR magnitude, which was observed on the control day only: emotional support seeking (with greater emotional support seeking being associated with a greater CAR). Whilst increased CARs have, in some instances, been associated with burden, such as on work days compared with weekends (Schlotz et al., 2004) and work overload (Steptoe et al., 2000), the results reported here support the concept that (appropriate) reactivity itself is not associated with negative health outcomes and, on the contrary, is linked with positive mood states. Conversely, however, emotional support seeking was also associated with lower reported wellness, which could suggest less optimal effects of this form of coping, and could potentially indicate that the greater CARs may reflect burden rather than appropriate or healthy functioning. The heterogeneity in these findings again demonstrates the complexity of the processes underpinning HPA axis functioning, however, in some cases where increased CARs have been associated with negative or chronic experiences, such as work overload, it is possible that the exaggerated CARs are associated more with the anticipation of many responsibilities and tasks to complete, rather than 'stress' itself (McEwen, 2011). Of course, repeated burden of excessive workload may still result in the repeated

activation of the HPA axis, or failure to recover, and it is under these circumstances that the moderators of this adaptive response malfunction, and subsequently can lead to stress-related ill-health (e.g., McEwen, 1998). These findings demonstrate that both psychological and physiological processes may be involved in appraisal and the subsequent coping strategies that individuals engage with. Therefore, whilst the differing associations between state indices and these appraisal-related variables observed across days are somewhat difficult to interpret, the findings support the notion that coping styles vary depending on the context of the situation. That is, coping style is not a global construct, as different situations require altered coping strategies: for example, problem-focused strategies (e.g., managing the issue by seeking information) are often engaged with when coping with job-related stress, whilst emotion-focused strategies (such as seeking others company) are typically adopted for family stressors (O'Brien & DeLongis, 1996).

With regard to psychological state reactivity, coping style was also associated with a number of the state indices. Active coping, a coping strategy associated with positive health outcomes (Kopp et al., 2003), was associated with lower anxiety on the evening before the task and lower stress on the morning of the event. Planning and acceptance were associated with lower state anxiety the night prior to the event. Acceptance was also associated with lower reported stress on the morning of the control day. The majority of the significant findings, however, refer to less optimal coping strategies; denial and self-distraction were associated with greater state anxiety, stress, and lower reported happiness on the day of the event. Self-distraction was also associated with lower reported wellness

on the morning of the novel event and was associated with greater anxiety, stress, and lower happiness on the control day. Furthermore, greater substance use as a coping strategy was associated with greater anxiety on both the morning and evening of control day, as well as with greater reported stress and lower reported happiness. This is in concordance with previously reported links between substance use and increased levels of psychological morbidity (e.g., Wetherell et al., 2011; Wetherell & Montgomery, 2014).

Greater behavioural disengagement was associated with greater reported anxiety at all time points with the exception of the morning of the day prior to the task, it was also associated with greater reported stress on the morning of the event, and lower reported wellness on the morning of the task. In addition, greater behavioural disengagement and self-blame were also associated with greater reports of mental alertness at sleep onset the night before the control day, indicating interference with sleep. Substance use and self-blame were also associated with greater anxiety the night before the event and on the morning of the control day. Greater venting and self-blame were associated with lower wellness on the day of the event. Self-blame was also associated with lower reported happiness and with greater anxiety and stress reported on the day of the event. The use of humour as a coping strategy was associated with lower reported happiness and wellness on the morning of the event and with greater reported stress on the morning of the control day. Emotional support seeking was also associated with lower reported wellness on the control day.

The association between coping styles and state indices are more easily interpreted than those of the cortisol findings, with active coping styles being

associated with lower state anxiety, and greater happiness in the present analysis. These findings support those of previous work, indicating that active or task-focused coping styles are the most beneficial for optimal functioning (e.g., Aldridge & Roesch, 2007), whilst avoidant coping styles are associated with greater psychological dysfunction (Higgins & Endler, 1995; Proulx, Koverola, Fedorowicz & Kral, 1995). Furthermore, the associations between active coping strategies on the day prior and the day of the event could not only be indicative of a role of coping in the anticipation of novel activity but may also be argued to support previous suggestions that coping is adaptive, with different coping styles becoming useful under various environmental conditions (e.g., Braverman, 1992; Folkman, 1992; Zeidner & Saklofske, 1996).

These findings are generally consistent with those reported in previous studies, observing less positive and more adaptive responses in those who engage with an avoidant coping style such as behavioural disengagement or self-distraction. The absence of evidence for positive effects of more adaptive coping styles may suggest that the less positive coping styles are stronger ‘risk factors’ than the positive styles are ‘buffers’ in the context of anticipation of a novel event.

#### *Perceived stress*

Perceived stress was associated with greater state anxiety, greater stress and lower reports of morning happiness and wellness, at all time points. Higher perceived stress was also associated with greater physical tension and mental alertness at sleep onset the night before the control day. These findings are consistent with previous reports of associations between perceived stress and anxiety (Spada et al., 2008), as are the findings that higher perceived stress is

associated with lower levels of wellbeing (Skok, Harvey & Reddihough, 2006; Raina et al., 2005) and happiness (Piqueras, Kuhne, Vera-Villarroel, Straten & Cuijpers, 2011). The association between physical tension the night before each sampling day suggests sleep disturbance in those who perceive their lives to be more stressful; these findings indirectly support those of previous studies demonstrating associations between disturbed and shortened sleep (Åkerstedt, 2006) and insomnia in individuals reporting more life stress (Healey et al., 1981).

Collectively, whilst the findings are in line with previous evidence indicating that high perceived stress is associated with poorer general physical and psychological health and wellbeing, as was the case with the overall findings reported for Type D, the anticipatory period was not unique in being more taxing than the typical day. That is, those higher in perceived stress reported greater anxiety, greater stress, lower happiness and wellbeing, regardless of the day's activities. Therefore, it is likely that individuals reporting greater levels of stress in their lives are likely to be more anxious and less happy on a daily basis; this could indicate pathways underlying the association between stress and negative health consequences, such as poorer immune functioning (Ebrecht et al., 2004; Lovell & Wetherell, 2011) and depression (Spada et al., 2008). High perceived stress may inappropriately prolong arousal, which in turn, may lead to dysfunction of physiological stress systems, such as the HPA axis (McEwen, 1998) which may be responsible for the association between stress and the negative health factors mentioned above.

### *Perseverative thinking*

Although it was not possible to examine the period immediately following the event in the present analysis, perseverative thinking was examined in relation to anticipation of a forthcoming novel event. Greater core perseverative thinking was associated with greater cortisol secretion across the control day. In addition, greater core perseverative thinking was also associated with greater state anxiety at the majority of the time points, with the exception of the morning of the day prior to the task and the morning of the control day, and was also associated with lower reports of wellness on both the day prior and the day of the event. Greater core perseverative thinking was also associated with lower happiness reported on the morning of the event. Unproductive thoughts were associated with greater anxiety at all but one time point, the morning of the control day, and with lower happiness and wellness on the morning of the day prior to the task and the day of the task. Perseverative thoughts associated with difficulty to disengage were also associated with state anxiety at the majority of the assessed time points, with the exception of the evening prior to the task and the morning of the control day. There was also an association between greater difficulty to disengage and lower reported wellness on the day prior to the task. Perseverative thinking additionally interfered with sleep onset, with unproductive thoughts and difficulty to disengage being associated with greater physical tension on the night prior to the task.

Although previous studies have focused predominantly on the effect of perseverative thinking on prolonged stress responding, the findings of the present study can be seen to indicate additionally that perseverative thinking may contribute to general daily functioning. This suggestion emerges from findings

that core perseverative thinking, unproductive thoughts and difficulty to disengage were all associated with greater state anxiety at almost every assessed time point. Furthermore, core perseverative thinking was associated with greater basal diurnal cortisol secretion on the control day (i.e. on the day of ‘typical’ activity) but not during the preparatory period preceding the novel task, or on the day of the task itself. A previous study (O’Connor et al., 2013) observed greater CARs in those individuals who had recalled a stressful event four weeks following and demonstrates that a period of rumination may have extended effects on aspects of diurnal cortisol secretion. A link between rumination and cortisol may therefore be expected following the current manipulated stressors; however, the follow-up period in the current studies were considerably shorter and therefore, the impact of rumination is likely to be less significant. Based on the notion of delayed effects, the observed relationships between rumination and cortisol secretion on a neutral day may therefore reflect the effects of prior rumination.

In addition, the findings also suggest that the relationship between perseverative thinking and state anxiety is not based on a specific activity but may be associated with other variables suggestive of an anticipation process: core perseverative thinking, for example, was associated with lower wellness on both anticipatory days (i.e. the day prior to and the morning of the event), as well as lower happiness exclusively on the morning of the event. Similarly, unproductive thoughts were associated with lower happiness and wellness on both the morning of the day prior to and the morning of the task; difficulty to disengage was associated with lower wellness on the day prior to the task. Together, these

findings indicate that whilst typical days are characterised by elevated anxiety (a form of negative affect), in greater perseverative thinkers, days involving anticipation of a novel event are instead associated with lower positive affect, including happiness and wellness. These associations are therefore worthy of further exploration beyond cross-section analyses.

### *Sleep*

Sleep duration was associated with a variety of individual differences: fewer prospective and retrospective memory errors were associated with longer sleep duration on the day of the task; unproductive thoughts were associated with shorter sleep duration the night before the day prior to the event; greater emotional stability was associated with shorter sleep on the day of the event; active coping was associated with shorter sleep duration; and denial (coping style) was associated with shorter sleep duration.

There was no association between sleep duration and cortisol secretion, with regards to either CAR magnitude or overall diurnal secretion. Interestingly, however, longer sleep duration was observed on the control day, compared with the day prior to the event and the day of the event, suggesting that anticipation of a novel event may be related shorter sleep on the two anticipatory days. This finding is consistent with previous reports of longer sleep duration the night preceding non-eventful days compared with those which are perceived as more demanding (e.g., Åkerstedt, 2006), suggestive of an anticipatory influence. Furthermore, the finding that anticipation of forthcoming challenge is associated with shorter sleep duration concurs with those reported in a study of military personnel awaiting deployment to Iraq and Afghanistan, where these participants

experienced significantly shorter sleep duration compared to personnel who were not awaiting deployment (Seelig et al., 2010).

### *Health complaints*

Greater reports of health complaints were associated with Type D, trait anxiety, core perseverative thinking, unproductive thoughts (associated with perseverative thinking), difficulty to disengage (perseverative thinking) perceived stress, some coping strategies (substance use, and denial), lower emotional stability and poorer self-esteem. Interestingly, each of these trait variables have previously been ‘flagged’ as those linked with poor health outcomes to varying degrees and, therefore, the significant associations reported above are consistent with previous reports; for example, both Type D and low self-esteem have been associated with poorer immune functioning (Type D: Leary & McDonald, 2003; self-esteem: Cohen, Tyrell & Smith, 1993) and perseverative thinking, through its influence over chronic, prolonged physiological activation, is a risk factor for cardiovascular disease and suppression of the immune system (Brosschot, Gerrin & Thayer, 2006).

With regards to state variables, greater numbers of health complaints were associated with greater state anxiety at all time points (across all three days), as well as greater stress on the morning of the event. Health complaints were also associated with lower morning happiness on the day prior to and of the task, and wellness on the day prior to the event, as well as physical tension at sleep onset the night before the task. It is possible that other variables are involved in the associations reported here, for example, individuals reporting state anxiety at every time point may also score highly for trait anxiety and perceived stress. As

the previously reported observations indicate, trait anxiety and perceived stress were also associated with lower happiness. It is therefore possible that the greater number of health complaints are better explained by trait variables, which subsequently influence state indices, reported here.

### *Overview*

The associations reported in these analyses are consistent with the general profiles found in anxious, distressed and stressed individuals and conform to the characteristics which may expose individuals to more stress and have undesirable effects on health and wellbeing (e.g., wound healing: Ebrecht et al., 2004). Moreover, these findings further highlight the physical impact negative characteristics may have on individuals with these traits. It is important to consider that these relationships are correlational and do not permit inference of causality but are, however, worthy of further exploration.

Although the primary research aim of this chapter was to assess individual differences in relation to anticipation of a general, novel event, the events included in the present analysis were qualitatively different. That is, despite suggestions that these responses may be observed in response to general novelty (Fries et al, 2009), it is possible that the anticipatory response profile elicited in response to a stressful event may differ from that observed in response to anticipation of a reward. In order to acknowledge and assess whether this was the case, further analyses were run only on those data which exclusively assessed anticipation of a stressful event (i.e. data from Studies 2 and 3 only). This analysis revealed considerably fewer significant associations: prospective memory failure was positively correlated with CAR magnitude on both the day of the stressor and

on the control day, demonstrating that poorer prospective memory (memory for carrying out planned forthcoming tasks) was associated with greater CAR magnitude on the day of the planned stressor, and on the control day. The emergence of this association in the analysis of stressor data exclusively, is intriguing as it has been previously suggested that the CAR may, in part, contribute to prospective memory functioning (e.g., Fries et al., 2009). Whilst at first glance this finding could suggest that poorer prospective memory is only relevant when a stressful event is anticipated, the association was also observed on the independent control (typical) day. Despite assertions in the literature relating to the involvement of prospective memory activation in the CAR, very little research has directly investigated memory and the CAR, making interpretation of the present findings somewhat speculative. However, in a study measuring the CAR in patients with global amnesia, no CAR was observed, despite an otherwise typical diurnal cortisol profile (Wolf, Fujiwara, Luwinski, Kirschbaum & Markowitsch, 2005). Whilst these observations are not directly comparable with those of the present study, together, they support theoretical suggestions that memory is associated with the CAR, they also illustrate the complex mechanisms underlying this relationship and could indicate that the relationship may be stronger when faced with a stressor, compared with a novel task (which could be pleasant or neutral). Furthermore, the report of only two significant associations following analysis of the data exclusively focusing on stressor anticipation provides additional support for this suggestion, as a number of significant associations were observed when examining the role of individual differences in anticipation of a general novel event.

Overall, the findings of the present analyses demonstrate that individual differences that have previously been associated with psychopathology and generally less adaptive psychological functioning are associated with less positive state reactivity in anticipation of a novel task and, for some variables, also to general daily functioning, whereby little or no discrimination is demonstrated, regardless of daily activities (e.g., as is the case for trait perceived stress and state anxiety).

The functioning of the HPA axis and how it interacts with the external environment and behaviour are, however, complex. The observation of limited associations between cortisol reactivity on the day of the event and personality characteristics in the present study demonstrate that these variables may play more of a key role in adaption to challenge or novel experience than previously considered. However, this was the first assessment of comprehensive psychobiological data assessing the more global anticipatory stress response to a variety of stimuli, in light of studies indicating a more generalised response than previously thought. Therefore, it is possible that, just as there appear to be individual differences which appear to affect how individuals respond to stressful events, there may be characteristics which withstand differences in the nature of the stimuli itself, and influence the general anticipatory response to novel experiences as a whole.

Furthermore, in some respects, the lack of significant correlations between the majority of the individual differences assessed in the present study and the CAR, or diurnal cortisol secretion, supports suggestions that the phenomenon is predominantly determined by situational factors, with trait variables only

contributing to a small extent (Hellhammer et al., 2007). Moreover, it should be considered that whilst the present study examined the period preceding a novel task (an experience which may, in itself, alter glucocorticoid secretion: e.g., Schlotz et al., 2004), the protocol did not assess stress reactivity in direct relation to the task for all studies, therefore making interpretation and comparison between these studies challenging in that respect.

Whilst in some instances it may be tempting to speculate on the effects of individual differences on state activity, the majority of results reported for the present analyses are correlational (with the exception of age, sex and categorised Type D effects) and therefore preclude any conclusions regarding the direction of causality. Previous studies have, however, suggested that a number of individual difference characteristics may have deleterious effects on general wellbeing, although these relationships are likely more complex than a simple one-way process. For example, health complaints may lead to increased anxiety and stress, or the relationship may instead indicate poorer immune functioning in these individuals as a result of being highly stressed and anxious, which may potentially have caused dysregulation or over-activation of the HPA axis (thus, potential reduced immune functioning, hence, health complaints). Whilst establishing cause and effect in these relationships is problematic, future studies could seek to assess these variables longitudinally to observe the rigidity of these relationships and perhaps attempt to develop interventions which may encourage the development of characteristics and thought processes which have been consistently demonstrated to promote healthy functioning and psychological wellbeing.

The current chapter included a large number of analyses and the risk of Type 1 error should therefore be considered when interpreting the findings. However, these analyses were exploratory in nature, aiming to shed light on possible associations between a range of theoretically relevant individual difference factors and the anticipatory response preceding a special/non-typical event. These relationships should therefore be followed up to assess direct pathways that link individual differences factors to anticipatory response to novel stimuli.

### *Conclusion*

This was the first assessment of collated data from novel events to investigate anticipation of global novel activity, regardless of the nature of the stimuli. Each of the studies included in the analysis provided data for the days surrounding the anticipatory period preceding the event (the day prior and the day of the event) and a control day, to assess basal activity.

The findings of this exploratory work support the concept that some individual characteristics play a role in both basal and anticipatory psychological (and physiological) functioning. Moreover, the findings presented here may shed light on relationships between certain individual difference factors and physiological functioning triggered in anticipation of a novel experience, particularly in relation to the influence of coping styles, and of traits such as conscientiousness. Finally, the present analyses identify characteristics which may pose as risk factors for greater appraisals of stress, which may impact upon subsequent psychological and physiological reactivity to a novel event. The identification of these characteristics can inform future research on areas which

require the development of interventions which may encourage engagement with techniques or behaviours which may in turn serve to moderate the relationship between stress and poor health outcomes.

**Table 9.2** Pearson correlations greater than 0.3, between individual differences factors and state indices for day 1 (day prior to task).

	<i>CAR magnitude /nmol</i>	<i>AUC<sub>G</sub></i>	<i>State anxiety AM</i>	<i>State anxiety Bed time</i>	<i>Stress AM</i>	<i>Happiness AM</i>	<i>Physical tension</i>	<i>Wellness</i>
<i>Health complaints</i>	-	-	-	r=.345*	-	r=-.331*	-	r=-.310*
<i>Type D</i>	-	-	-	r=.308*	-	-	-	r=-.426*
<i>Trait anxiety</i>	-	-	-	r=.446*	-	-	-	r=-.431*
<i>Self esteem</i>	-	-	-	r=-.305*	-	-	-	r=.337*
<i>Perceived stress</i>	-	-	-	r=.457*	-	r=-.311*	-	-
<i>Perseverative thinking: Core</i>	-	-	-	r=.309*	-	-	-	r=-.326*
<i>Unproductive thoughts</i>	-	-	-	-	-	-	-	r=-.349*
<i>Difficulty to disengage</i>	-	-	-	-	-	-	r=.369*	-
<i>Substance use</i>	-	-	-	r=.317*	-	-	-	-
<i>Self blame</i>	-	-	-	r=.317*	-	-	-	-

**Table 9.3** Pearson correlations greater than 0.3, between individual differences factors and state indices for day 2 (day of the task).

	AUC <sub>G</sub>	State anxiety AM	State anxiety Bed time	Stress AM	Happiness AM	Wellness
<i>Health complaints</i>	-	r=.321*	r=.409*	-	-	-
<i>Conscientiousness</i>	r=.337*	-	-	-	-	-
<i>Emotional stability</i>	-	-	r=-.348*	-	r=.324*	-
<i>Type D</i>	-	r=.523*	r=.381*	r=.505*	r=-.321*	r=-.390*
<i>Trait anxiety</i>	-	r=.535*	r=.488*	r=-.487*	r=-.409*	r=-.386*
<i>Self esteem</i>	-	r=-.394*	r=-.378*	r=-.418*	-	r=.385*
<i>Perceived stress</i>	-	r=.428*	r=.424*	r=.339*	r=-.339*	-
<i>Perseverative thinking: Core</i>	-	r=.316*	r=.452*	-	r=-.358*	r=-.313*
<i>Unproductive</i>	-	r=.329*	r=.496*	-	r=-.351*	r=-.326*
<i>Difficulty to disengage</i>	-	-	r=.418*	-	-	-

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<i>Denial</i>	-	r=.317*	r=.391*	r=.303*	-	-
<i>Emotional support seeking</i>	r=-.340*	-	-	-	-	-
<i>Behavioural disengagement</i>	-	r=.316*	r=.318*	-	-	-
<i>Self blame</i>	-	r=.355*	r=.314*	r=.328*	-	-

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**Table 9.4** Pearson correlations greater than 0.3, between individual differences factors and state indices for the control day.

	<i>AUC<sub>G</sub></i>	<i>State anxiety AM</i>	<i>State anxiety Bed time</i>	<i>Stress AM</i>	<i>Mental alertness</i>	<i>Wellness</i>
<i>Agreeableness</i>	-	-	-	r=-.318*	-	-
<i>Emotional stability</i>	-	-	r=-.315*	-	-	-
<i>Openness</i>	-	-	-	-	-	r=.312*
<i>Type D</i>	-	-	-	-	r=.301*	-
<i>Trait anxiety</i>	-	r=.334*	r=.361*	r=.322*	-	-
<i>Self esteem</i>	-	r=-.306*	-	-	r=-.309*	-
<i>Perceived stress</i>	-	r=.335*	r=.340*	r=.311*	-	-
<i>Perseverative thinking: Core</i>	r=-.343*	-	r=.302*	-	-	-
<i>Unproductive</i>	r=.326	-	r=.342*	-	-	-
<i>Difficulty to disengage</i>	-	-	r=.319*	-	-	-

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<i>Coping: Self distraction</i>	-	r=.400*	r=.324*	r=.397*	-	-
<i>Active coping</i>	r=.305*	-	-	-	-	-
<i>Substance use</i>	-	r=.425*	-	r=.410*	-	-
<i>Self blame</i>	-	-	-	-	r=.305*	-

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## **Chapter 10**

General discussion

The primary aim of this thesis was to investigate psychological and physiological reactivity during the anticipatory period preceding, and the recovery period following, exposure to novel, adverse and positive stimuli, both under controlled laboratory conditions and in naturalistic settings. This aim was based on previous literature reporting greater physiological and psychological reactivity on days which are considered more challenging than typical days (e.g., work days compared with weekends: Kunz-Ebrecht et al., 2004; the day of examinations compared with a typical day: Weekes et al., 2008; and on the day of a competition, compared with non-competition days: Rohleder et al., 2007). These findings are suggestive of anticipation of forthcoming demand and contribute to previous suggestions that the HPA axis plays a role in preparing individuals for coping with forthcoming stressful events (e.g., Wetherell et al., 2014).

The aim of the first study, therefore, was to develop and evaluate an ecologically valid laboratory stressor paradigm, as a tool to enable a) the assessment of psychobiological response to a novel socially evaluative, ecologically valid, and economical stressor and b) the subsequent assessment of the anticipatory and recovery period following this stressor, in Study 2. Studies 2 and 3 investigated the anticipatory response to a planned stressor: the stressor paradigm (developed in Study 1) in Study 2, and a naturalistic stressor (a solo skydive) in Study 3.

Following a recent study in children (Flinn et al., 2011), a further aim of the thesis was to assess whether, in addition to activation preceding challenging activities, anticipatory responses are observed in response to positive stimuli. This was investigated by examining physiological and psychological reactivity during

the anticipatory period in proximal context of a positive novel event. Finally, the thesis investigated the role of individual differences in the psychobiological anticipatory response to novel activity.

### *Summary of key findings*

With regard to the aims of the first study, a novel stressor paradigm was developed and evaluated in terms of its physiological and psychological stress-inducing properties. The stressor paradigm, involving multitasking under direct critical social evaluation led to increases in self-reported anxiety and acute cardiovascular reactivity. Furthermore, and of great importance for subsequent uses of the paradigm, those individuals who expected to participate in the condition involving direct critical social evaluation, reported greater levels of distress on the morning prior to participation compared with those expecting to undertake multitasking with indirect evaluation.

This finding supports previous research which has observed greater anticipatory stress reactivity preceding a stressful task than during actual exposure to a stressor itself (e.g., Aschbacher et al., 2013) but also demonstrates that the paradigm is capable of eliciting an anticipatory response. The developed stressor could therefore be appropriately used as a tool to manipulate anticipatory demands in laboratory conditions (Study 2)

Anticipation of the novel stressor developed in Study 1 was assessed in Study 2, where psychobiological indices were recorded across four days: the day prior to stressor exposure, the day of stressor exposure, the day following exposure, and on a typical day. This intensive protocol enabled thorough investigation of changes in cortisol and state psychological measures such as

mood and state anxiety in relation to forthcoming demand. No significant effects were observed for cortisol indices; however, participants did experience other anticipatory responses prior to exposure to the planned stressor and, furthermore, these responses were detectable on the morning prior to stressor exposure, with participants reporting greater stress and lower happiness on this day. On the morning of the planned stressor, participants also reported being more anxious than the following two sampling days, again, indicative of an effect of the stressor itself on anticipatory mood. Concurrent with the findings alluding to an anticipatory response, which emerged in Study 1, the findings of Study 2 demonstrate that the developed paradigm is capable of eliciting anticipatory responses for psychological indices.

A similar sampling protocol was applied to investigate the anticipatory period preceding a solo skydive; a naturalistic stressor characterised by considerably greater threat than laboratory stress. Whilst the actual procedure that unfolded during the data collection period differed from the planned protocol in terms of the variation in duration of anticipation, the findings provided novel insight into anticipation of a skydive and supported previous literature regarding the psychological burden of uncertainty (e.g., Greco & Roger, 2003). The observed acute reactivity to the skydive was consistent with that of previous studies and further supports the use of a skydiving paradigm in the assessment of the acute stress response under naturalistic conditions. In relation to the aim to assess anticipation of the skydive, the present findings demonstrated promising results, with greater reports of thinking about the skydive on the morning of the jump revealing particularly interesting results for those who anticipated skydiving

with uncertainty: the group who went to bed on the day prior to the planned skydive with uncertainty regarding whether they would jump the following day reported a psychological and physiological profile characteristic of greater burden on the morning of the planned event, compared with both those who knew they would be skydiving the next day, and those who knew, with certainty, that they would not.

In further support of the use of a skydiving paradigm to assess anticipation of a stressor, on the morning of the planned skydive, greater CARs were observed in the group who knew they would jump and those who anticipated with doubt, compared with those who awoke knowing that they would not be skydiving that day. This trend, although underpowered and therefore not statistically significant, supports previous studies demonstrating the association between forthcoming demand and cortisol secretion, particularly in relation to the CAR; it suggests adaption of the response (in healthy participants), whereby arousal is only triggered under circumstances which require such reactivity, and resources are spared upon cessation of a threat, or it fails to present itself.

These findings suggest that participants who a) knew they would jump, or b) anticipated with doubt demonstrate responses which indicate that they were thinking about, and anticipating a forthcoming demand. Furthermore uncertainty of whether a forthcoming event will or will not present itself is perceived to be more onerous than the certainty of such an event, a finding which has been reported in previous literature. These findings provide preliminary knowledge of prolonged activation associated with anticipation of stressful stimuli; however,

given the small sample sizes that are typical of naturalistic studies, future work should attempt to replicate these comprehensive assessments in larger samples.

Evidence, albeit limited and restricted to children, has suggested that in addition to anticipatory responses preceding adverse events, similar responses may also be evident for stimuli perceived as pleasant. Study 4 therefore assessed responses during the anticipatory period preceding a pleasant, reward stimulus. The findings did not support the proposition that anticipation of a pleasant activity evokes psychological or physiological responses; however, it is suggested that an absence of effects was due to the salience of stimulus. That is, the manipulation of the pleasant stimulus was not sufficiently exciting to encourage the level of engagement that may be required to detect psychobiological reactivity to a subtle manipulation.

An emerging theme across the research programme relates to the impact of appraisal of the stressor or novel forthcoming event. In Study 1, participants with the knowledge that they would be standing in front of the researcher, who would be providing critical feedback on their performance whilst completing the task, reported thinking and worrying about the forthcoming task more than those who expected to be seated with their back to the researcher. Furthermore, in Study 3, psychobiological indices were activated in those who expected to skydive compared with those for whom the threat had been removed. The influence of psychosocial factors on the stress response is well-established within the literature, as is the effect on basal indices of physiological functioning: days perceived as 'good' are associated with lower diurnal cortisol secretion compared with those reported as 'bad' (Lundberg, 1989) and greater CARs have been

observed on days containing daily stressors, such as arguments (Stawski et al., 2013).

With regards to internalised characteristics, greater cortisol secretion has been observed in highly anxious individuals (Young et al., 2001) and those reporting social stress and worry (Wüst et al., 2000), all of which collectively involve perception/appraisal of environmental factors (e.g., Skinner & Brewer, 2002). These findings support speculations that humans have the ability to cognitively control their response to stress, and the findings of this thesis lend further support to this concept. The findings can collectively indicate that individuals who appraise situations in such a way as to reduce the emotional impact of these events demonstrate more adaptive psychological and physiological responses (Jamieson, Nock & Mendes, 2012) to both stressful situations and to generalised novel events. For example, findings from Study 5 revealed associations between perceived coping in relation to stress and anxiety across all sampling days, regardless of activity. Perceived stress was also associated with greater state stress and lower happiness on each of the sampling days. Perseverative thinking, another variable encompassing the process of appraisal, was also associated with state anxiety across all three of the sampling days and was associated with lower reports of both happiness and wellness on the day of the task. The importance of coping strategies were also identified: on the day prior to the task, acceptance, behavioural disengagement and positive reframing were all associated with greater diurnal cortisol secretion and, on the day of the task, acceptance was, again, associated with greater diurnal cortisol

secretion, whilst substance use and emotional support seeking were associated with lower diurnal secretion.

Acceptance is a coping strategy that is deemed positive and, as such has been incorporated in interventions aiming to reduce avoidant behaviour to improve quality of life, with promising success (Lillis, Hayes, Bunting & Masuda, 2009). The finding that acceptance is related to greater cortisol secretion only on the day prior to and the day of a novel event could indicate that those who consciously accepted the novel activity were more engaged in their approach to the task and that this arousal may have subsequently influenced diurnal cortisol. Similarly, conscientiousness, a trait associated with organisation and diligence, was also associated with greater cortisol secretion, but only on the day of the event. As discussed in Chapter 9, a previous study examining daily physiological functioning and conscientiousness observed associations between the trait and lower cortisol secretion (Nater et al., 2010). It is therefore possible that conscientiousness is an adaptive characteristic, typically associated with lower daily secretion of cortisol but which may serve to aid in the preparation for forthcoming demand on the day of novel events.

Conversely, a range of negative appraisal-related individual difference factors were associated with the number of health complaints reported. Greater numbers of health complaints were associated with trait anxiety, perceived stress, all forms of perseverative thinking, denial and substance use (both avoidant coping strategies). Furthermore, positive appraisal-related factors (self-esteem and emotional stability) were associated with fewer reported health complaints. The influence of appraisals over physiological reactivity has been reported elsewhere

(Jamieson, Nock & Mendes, 2012), where participants encouraged to reappraise arousal as functional and positive prior to stressor exposure elicited smaller cardiovascular responses than those who did not receive such intervention. These findings also fit with the associations observed in Study 5 between positive and negative coping strategies and a plethora of state reactivity indices. Interestingly, there have also been reports that the appraisal of stress as being negative for one's health is more harmful than stress itself: in a large sample Keller and colleagues (2012) observed that the combination of greater reported stress and a greater perception that stress is bad for one's health was associated with worse physical and mental health outcomes, even compared with people who reported more stress in their lives but lower perception of stress as negative for their health.

In a real-world context, the present findings support the concept that the tendency to make positive or negative appraisals can alter physiological and psychological responses to both stressful and positive novel stimuli and that these responses can either be adaptive or have negative effects on physiological function, that is, that perceiving more threat than is warranted may elicit inappropriate levels of arousal (McEwen, 1998). Whilst the present research programme focused on anticipation of acute stress reactivity (Studies 1, 2 and 3), these findings may present implications for our understanding of chronic stress, as it is likely that individuals who perceive greater stress (and poorer coping resources) may elicit inappropriate responses to activities on a regular basis. Therefore this repeated response might lead to chronic activation of the stress response, which as discussed earlier in this thesis, is widely associated with a number of poor health outcomes (McEwen, 2005; Schulkin, 2011). However, as

these findings are cross-sectional and exploratory in nature, further examination is warranted in order to ascertain the robustness of this relationship and to explore further, the possible underlying mechanisms which may contribute to the potential mediating effect of appraisal in the association between stress and health.

Furthermore, whilst the causal direction of correlations cannot be ascertained, this thesis has presented exploratory findings identifying individual difference factors which may serve as potential buffers in the relationship between stress and psychological functioning. Although some of these relationships have been previously observed, this thesis has identified their importance in relation to anticipation of novel events collectively. The identification of these relationships supports previous suggestions that the activation of memory representations for upcoming activity in general may impact on anticipation, rather than the expectance of a stressor specifically (Fries et al., 2009). Furthermore, they suggest potentially protective individual difference factors which could be encouraged through intervention and those which should be 'flagged' as potential risk factors for dysregulation of physiological processes.

### *Limitations*

The findings of this thesis should be viewed in light of a number of limitations that are typical of studies that incorporate comprehensive assessments of psychobiological indices and naturalistic designs.

Firstly, a key issue for consideration is the predicament of adherence to protocol, a common complication in psychobiological research heavily reliant on the collection of samples in the domestic setting. As previously discussed (see Chapters 1 and 2), adherence to strict timings of saliva sampling is crucial when

seeking to obtain accurate measures of diurnal profiles of cortisol secretion (e.g., Stalder et al., 2016). This is especially crucial when assessing the CAR, whereby samples are collected in relation to awakening, as even a delay as short as 15 minutes can produce inaccurate readings, such as smaller CARs (Okun et al., 2010). Inaccurate observations such as this can lead to the interpretation of abnormal profiles, suggestive of dysregulation of the HPA axis, when actually such observations may simply be due to sampling error. Following other studies of this nature (e.g., Wetherell, Lovell & Smith, 2014) and considering recent recommendations (e.g., Stalder et al., 2016), adherence was monitored through the recording of self-reported sleep and waking diaries, and sampling times. The collection of this information provides positive indicators of adherence and has been observed to be equally as reliable as forms of objective measures (Dockray et al., 2008; Kraemer et al., 2006; Okun et al., 2010). Furthermore, adherence was encouraged by clearly stating to participants on a number of occasions throughout the study, that their compliance with sampling times was monitored, a tactic which has considerably improved adherence in previous work (Broderick et al., 2004), to the point where subjective and objective adherence are congruent. In the present research programme, this data enabled the successful cross-examination of sampling times with times of waking and allowed the exclusion of participants who provided waking samples more than 15 minutes after their reported time of waking. Although attempts were made to provide objective monitoring in a subset of participants through assessments of actigraphy, the subsequent data were not comprehensive enough for reliable interpretation.

Despite the absence of objective data, the successful removal of a small number of individuals based on self-reported information provides confidence that rigour was applied in the assessment of adherence and that non-adherent data was identifiable and could subsequently be excluded from analysis. Although the use of objective, electronic monitoring does, as previously recommended (e.g., Stalder et al., 2016) offer greater objectivity in adherence monitoring, these methods are also reliant on participant engagement and can only be used retrospectively to remove the influence of non-adherence. In order to overcome fully the issue of participant compliance, absolute control must be possible over the participants and their environment. This opportunity can be provided within a highly controlled experimental environment, such as a sleep laboratory, which can be designed to maximise ecological validity (e.g., Elder, Wetherell, Barclay & Ellis, 2014). However, the maximal control this procedure would apply would be offset by a considerable reduction in external validity and could not be successively implemented in relation to a unique naturalistic anticipation paradigm, such as skydiving. Moreover, one of the strengths of this thesis is the application of ecologically valid protocols, as far as could be achieved. The aim was to observe responses of individuals in their natural environment and, therefore, the inclusion of a residential protocol would have been inappropriate in the present research programme, particularly for indices of cortisol secretion, which, as identified in previous literature, is a biomarker sensitive to novelty (e.g., Biondi & Picardi, 1999).

A second limitation related to the salience of manipulations. In Study 2, the stressor may not have been perceived as sufficiently threatening to the social

self to elicit an anticipatory stress response (Dickerson & Kemeny, 2004), as evidenced by the finding that the direct critical social evaluation group found the stressor to be more stressful than those exposed to considerably less social evaluation. This was also evident when attempting to manipulate a positive, rewarding stimulus in Study 4, where although there were trends towards greater CARs in participants who completed the task, compared with those who failed to do so, the lack of salience of the manipulation may have precluded more convincing effects. That is, the incentive to complete the reward task may not have been substantial enough to elicit a physiological or psychological response to a pleasant stimulus, as observed in children during anticipation of Christmas (Flinn et al., 2011). This suggestion is based on previous findings where reward has elicited significant physiological responses in gamblers (e.g., Meyer et al., 2000), indicating that the need to complete a task for a monetary reward does elicit reactivity when the reward is perhaps perceived to be more substantial or of greater importance.

As anticipatory responses are adaptive and sophisticated processes capable of preparing individuals for forthcoming demand, they should not be triggered in situations where there is no perceived threat and therefore no requirement for arousal. Skydiving represents a significant life-threatening event and is therefore a useful tool for the assessment of anticipatory processes. As a naturalistic stressor, its use is, however, limited. Attempts were made to develop paradigms to assess anticipatory processes to aversive (Study 2) and positive (Study 4) stimuli. Both paradigms have been successfully applied in this programme; however, ethical, physical and logistical restraints prevent the manipulation of extreme negative and

positive events of the salience required to elicit robust HPA activation, especially in paradigms where the event is embedded in everyday activities that, despite efforts at control, will influence appraisal and responding.

Issues related to the representativeness of the sample should also be considered. Whilst the target sample group was healthy adults aged 18-40 years, in line with the aim to assess stress reactivity in healthy younger adults (and to avoid possible confounding variables associated with physiology alterations in younger and older groups), the majority of participants actually recruited were at the lower end of the target age bracket, with mean ages across the studies ranging between 21-26 years. Furthermore, the majority of participants were undergraduate university students and, whilst this is in common with the majority of previous literature, it does preclude application to wider samples. Furthermore, the sample sizes, although similar to those of previous studies assessing stress responses in naturalistic designs, in some cases were not sufficient for the level of analyses required. This issue was further compounded in cases of insufficient provision of samples, or deviations from the planned protocol (e.g., Study 3). Small samples are often sufficient for the detection of effects where there are very high expected incidence rates (for example, in the subgroup of participants who completed the planned skydive, which reliably elicits a significant physiological response, even in a small group,  $n = 8$ ), but may not enable the observation of more subtle effects. Although the sample sizes for some of the studies were small, the sampling protocols were more intensive than many larger scale studies that have collected considerably fewer samples per participant, some with just 4 data points on a single day (e.g., van Santen et al., 2010), and many only assessing

responses over one (Whitehead, Perkins-Porras, Strike, Magid & Steptoe, 2007) or two days (e.g., Oosterholt, Maes, Van Der Linden, Verbraak, & Kompier, 2015). Therefore, whilst larger samples may have allowed greater detection of subtle relationships in the data, the comprehensive protocols utilised for the present research programme enabled the collection of high-quality data, allowing more meaningful (although sometimes tentative) interpretations. Future work should therefore attempt to replicate similarly thorough procedures with larger and more generalisable samples to establish the replicability of findings.

A further limitation related to relatively modest samples sizes across study groups in the present thesis is that this prevented meaningful exploration of associations between individual difference factors and anticipatory and acute reactivity to novel in the individual studies. However, in order to proceed with exploration of this data, the data from studies assessing the anticipatory and recovery periods surrounding novel stimuli (i.e. a laboratory stressor, solo skydive, and a positive memory task) were combined to tentatively investigate individual difference factors which may play a role in the anticipation or recovery period in proximal context of novel stimuli. Whilst due to the nature of the analysis, conclusions could not be drawn, these correlations provided interesting insight into individual difference factors which may warrant further investigation in future empirical work.

#### *Future research directions*

This programme of work has explored the role of anticipation of demanding and positive events with regards to both psychological and physiological parameters. Furthermore, the thesis sought to uncover associations

between individual differences and these responses. The key findings of this thesis demonstrate physiological and psychological reactivity to planned, ‘stressful’ events, and could therefore hold implications for real-world situations fitting this criteria (e.g., in a medical context, such as in patients awaiting surgery: e.g., Johnston, 1980). Investigation of both the anticipatory period preceding, and the recovery window following, medical procedures in future work could inform further as to the physiological impact of differing anticipation-recovery patterns, as dysregulated or delayed psychophysiological recovery may also delay surgical recovery. Exploration of this phenomena may not only enable the investigation of underlying mechanisms involved in the anticipation-recovery process, but may, in turn indicate ways in which less positive reactivity (i.e. resulting in delayed recovery) may be improved, and through which pathways this may be achieved.

With regard to reactivity to positive events, future work should focus attention on investigating anticipation of neutral and positive novel stimuli; this should then be assessed under both ecologically valid experimental settings (as was attempted in Study 4) and under natural conditions. For the experimental research, as attempted in the design of a laboratory stressor in Study 1, the paradigm would need to allow for the manipulation of anticipation towards the pleasant event and the incentive would need to be greater than that of the reward offered in the present thesis. With regard to the naturalistic investigation of anticipatory responses, a positive event, such as going on holiday, may be a suitable candidate, as this is a familiar occurrence which is associated with positive emotions (Nawijn et al., 2010).

Additional future directions include closer examination of the relationship between appraisal and acute events, through replication of present findings with larger samples. The identification of robust associations between both protective and risk factors and psychobiological functioning in healthy populations could aid the prevention of illness prior to the onset of symptoms. Furthermore, investigating these relationships in clinical populations at risk of stress-related illness (e.g., cardiac patients) may inform intervention delivery to help reduce the risk of chronic ill-health, for example, by enhancing our understanding of therapies such as Cognitive Behavioural Therapy (Jamieson et al., 2013).

#### *Summary and conclusions*

This thesis sought to investigate and expand upon knowledge within the relatively novel research area of anticipation of forthcoming events. The findings support the proposition that anticipation of forthcoming demand may prolong activation of stressor systems (e.g., Engert et al., 2013) and that individual characteristics associated with greater sensitivity to stress are also associated with poorer physical health (e.g., perceived stress: Ebrecht et al., 2004; Spada et al., 2008). The present thesis expanded on previous research by exploring whether responses observed in anticipation of stress are exclusive to adverse stimuli, or whether the anticipation of a novel or non-typical positive experience may also elicit some form of physiological or psychological arousal. The findings indicate that the role of anticipation is highly complex and adaptive. By exploring the process of anticipation of a planned naturalistic stressor, these findings also contribute to knowledge regarding the uncertainty of exposure to a stressor,

specifically, this is the first study to have observed such findings in the proximal context of a solo skydive.

A final interesting finding to emerge from this research programme, is the role of appraisal in the response to both stressful and general novel events. The findings presented here demonstrate that the anticipation of any novel event, regardless of the positive or negative nature of the activity, is dependent on how it is interpreted by the individual, thus revealing appraisal to be considerably involved in reactivity.

In light of the plethora of evidence demonstrating the considerable impact of inappropriate stress responding on health outcomes and the highly influential role of appraisal in this relationship, further work could usefully incorporate samples identified as 'high-risk' (e.g., those with high trait stress and anxiety). In order to assist these individuals and to maintain healthy functioning in currently-well individuals, the development and assessment of interventions which may increase engagement with characteristics associated with more positive and adaptive appraisals (as demonstrated with positive effects in small samples) could be implemented to facilitate adaptive anticipatory and direct responses with regards to both adverse and general novel events.

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## Appendices

### Appendix A

Mean (and SD) physiological responses for both direct and indirect critical social evaluation.

	<i>Direct critical social evaluation (n = 20)</i>	<i>Indirect critical social evaluation (n = 19)</i>
<i>HR</i>		
Arrival at laboratory	80.35(15.43)	75.68(9.59)
Post- demonstration	85.30(17.16)	78.00(9.18)
Stressor cessation	96.55(17.54)	76.58(9.82)
Relaxation +10 minutes	76.45(11.20)	74.63(8.75)
End of study	77.25(11.19)	74.68(7.78)
<i>SBP</i>		
Arrival at laboratory	116.15(13.95)	114.26(12.01)
Post- demonstration	115.90(17.31)	113.84(13.02)
Stressor cessation	126.05(12.84)	116.00(13.94)

Relaxation +10 minutes	116.80(12.31)	112.21(11.84)
End of study	115.70(13.77)	114.00(10.24)
<i>DBP</i>		
Arrival at laboratory	67.70(8.46)	64.58(7.71)
Post- demonstration	68.10(8.89)	66.74(8.31)
Stressor cessation	77.35(9.21)	66.16(6.96)
Relaxation +10 minutes	67.95(7.96)	66.37(7.63)
End of study	66.50(9.32)	65.74(9.38)
<i>Cortisol/nmol</i>		
Arrival at laboratory	3.99(1.90)	4.93(2.84)
Post- demonstration	4.09(2.112)	4.84(2.96)
Stressor cessation	3.35(1.421)	3.89(2.52)
Relaxation +10 minutes	3.02(1.342)	3.43(2.16)
End of study	2.88(1.154)	2.91(1.81)

## Appendix B

Mean (and SD) psychological responses to the laboratory stressor for both direct and indirect critical social evaluation.

	<i>Direct critical social evaluation (n=20)</i>	<i>Indirect critical evaluation (n= 19)</i>
<i>State Anxiety</i>		
Arrival at laboratory	180.00(83.28)	144.37(106.71)
Post-demonstration	197.85(73.80)	166.42(112.02)
Stressor cessation	255.95(100.34)	255.89(121.55)
Relaxation +10 minutes	144.05(76.65)	129.26(99.63)
End of study	122.20(76.79)	107.84(101.21)

Appendix C

Mean stress reactivity assessed through the following indices: cortisol (nmol), state anxiety, heart rate (bpm), systolic and diastolic blood pressure (mmHg).

	<i>Arrival at laboratory</i>	<i>Post demo</i>	<i>Stressor begin</i>	<i>Stressor +5mins</i>	<i>Stressor +10mins</i>	<i>Stressor +15mins</i>	<i>Stressor cessation</i>	<i>Cessation +10mins</i>	<i>End of laboratory session</i>
<i>Heart rate/bpm (n=28)</i>	76.11 (14.32)	81.14 (14.47)	89.96 (16.72)	91.82 (14.63)	93.18 (15.14)	94.57 (14.211)	92.36 (17.80)	76.54 (12.28)	76.14 (12.17)
<i>SBP/bpm (n=28)</i>	118.64 (13.67)	117.57 (11.26)	130.18 (9.44)	125.54 (8.75)	128.29 (10.277)	127.11 (10.08)	127.32 (12.49)	117.79 (12.03)	114.36 (11.45)
<i>DBP/bpm (n=28)</i>	67.89 (6.70)	68.75 (7.90)	77.96 (9.89)	75.07 (9.17)	77.11 (9.75)	75.75 (8.68)	72.71 (8.91)	68.96 (9.23)	67.79 (8.88)
<i>Cortisol/nmol (n=26)</i>	6.54 (7.97)	5.16 (3.69)	-	-	-	-	4.78 (3.16)	5.01 (3.80)	4.42 (3.03)
<i>State anxiety (n=27)</i>	173.26 (81.53)	205.70 (87.09)	-	-	-	-	273.44 (107.27)	136.59 (67.93)	100.41 (66.85)

Appendix D

Means (SDs) for diurnal variables assessed over the course of the two sampling days: pre skydive day (day 1) and the day of the planned skydive (day 2).

	<i>Jumped as planned</i> ( <i>n</i> = 6)	<i>Anticipated with doubt</i> ( <i>n</i> = 10)	<i>Knew not jumping</i> ( <i>n</i> = 7)
<b><i>CAR magnitude</i> (<i>n</i>=21)</b>			
<i>Pre skydive day</i>	6.56(6.85) <i>n</i> =5	6.06(4.87) <i>n</i> =9	5.394(3.66) <i>n</i> =7
<i>Day of planned skydive</i>	8.14(7.20) <i>n</i> =5	8.03(9.19) <i>n</i> =9	3.17(3.26) <i>n</i> =7
<b><i>AUC<sub>G</sub></i> (<i>n</i>=20)</b>			
<i>Pre skydive day</i>	7349.55(3087.42) <i>n</i> =4	4930.05(2452.15) <i>n</i> =10	3982.85(1107.27) <i>n</i> =6
<i>Day of planned skydive</i>	9770.40(3767.15) <i>n</i> =4	5484.51(2244.69) <i>n</i> =10	4214.00(906.60) <i>n</i> =6
<b><i>State anxiety</i> (<i>n</i>= 20)</b>			
<i>Pre skydive day: AM</i>	109.33 (87.92) <i>n</i> =6	156.44(95.97) <i>n</i> =9	120.80(111.69) <i>n</i> =5
<i>Pre skydive day: wake +</i> <i>6hrs</i>	106.50(91.54) <i>n</i> =6	179.22(98.17) <i>n</i> =9	140.20(92.64) <i>n</i> =5

<i>Pre skydive day: bed time</i>	79.33(83.26) n=6	158.56(106.78) n=9	92.80(75.83) n=5
<i>Planned skydive day: AM</i>	120.17(76.77) n=6	179.89(116.15) n=9	114.00(88.29) n=5
<i>Planned skydive day: wake + 6hrs</i>	118.17(84.55) n=6	174.00(85.64) n=9	163.00(113.33) n=5
<i>Planned skydive day: bed time</i>	93.00(100.64) n=6	164.00(97.79) n=9	149.20(189.31) n=5
<b><i>Stressed (n=22)</i></b>			
<i>Pre-skydive day: AM</i>	21.67(26.90) n=6	29.60(25.95) n=10	24.67(24.83) n=6
<i>Planned skydive day: AM</i>	13.67(16.97) n=6	36.20 (32.83) n=10	15.83(10.11) n=6
<b><i>Happiness (n=22)</i></b>			
<i>Pre-skydive day: AM</i>	72.33(15.41) n=6	68.00(22.98) n=10	77.33(13.62) n=6
<i>Planned skydive day: AM</i>	80.00(14.13) n=6	74.70(18.54) n=10	82.00(11.10) n=6
<b><i>Mental demand (n=22)</i></b>			
<i>Pre-skydive day: AM</i>	1.83(1.33) n=6	1.80(1.14) n=10	2.50(1.38) n=6
<i>Planned skydive day: AM</i>	1.00(.89) n=6	2.30(1.16) n=10	2.33(1.37) n=6

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<b><i>Physically tense (n=22)</i></b>			
<i>Pre-skydive day: AM</i>	.33(.52) n=6	1.40(1.07) n=10	1.00(1.26) n=6
<i>Planned skydive day: AM</i>	.33(.52) n=6	1.80(1.48) n=10	1.33(1.37) n=6
<b><i>Wellness (n=22)</i></b>			
<i>Pre-skydive day: AM</i>	2.83(1.17) n=6	2.40(.70) n=10	3.00(.89) n=6
<i>Planned skydive day: AM</i>	3.00(.63) n=6	2.50(.85) n=10	2.33(1.21) n=6
<b><i>Thinking about the skydive (n=22)</i></b>			
<i>Pre-skydive day: AM</i>	37.33(25.92) n=6	56.30(26.81) n=10	26.17(19.44) n=6
<i>Planned skydive day: AM</i>	55.67(13.91) n=6	66.80(24.32) n=10	45.83(23.57) n=6
<b><i>Worrying about the skydive (n=22)</i></b>			
<i>Pre-skydive day: AM</i>	17.00(23.49) n=6	27.40(16.08) n=10	16.83(15.51) n=6
<i>Planned skydive day: AM</i>	17.833(13.42) n=6	38.70(26.61) n=10	30.00(30.76) n=6

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## Appendix E

Mean (SD) diurnal cortisol patterns, and psychological indices (n = 41).

	<i>Day prior to task</i>	<i>Day of task</i>	<i>Control day 1</i>	<i>Control day 2</i>
<i>Cortisol</i>				
<i>CAR/nmol</i>	4.97(3.97)	5.16(6.32)	4.60(7.22)	2.90(11.98)
<i>n= 31</i>				
<i>AUC<sub>G</sub></i>	3.49(.23)	3.47(.16)	3.49(.21)	3.47(.25)
<i>n= 25</i>				
<i>Stressed</i>	25.45(23.20)	20.95(16.48)	30.37(23.12)	26.21(22.09)
<i>n=38</i>				
<i>Happy</i>	67.63(21.70)	69.47(22.60)	63.76(23.07)	67.74(22.35)
<i>n=38</i>				
<i>Mentally alert</i>	1.56(1.11)	1.28(1.09)	1.67(1.07)	1.61(1.10)
<i>n= 36</i>				

Physically tense	.86(.90)			.72(.66)			1.06(1.19)			1.03(1.03)		
n=36												
Well	2.65(.79)			2.68(.88)			2.45(.93)			2.38(.92)		
n=37												
<i>Thinking about study</i>	32.49(28.49)			26.78(26.00)			37.54(25.59)			25.84(24.20)		
n=37												
<i>State anxiety</i>	AM	PM	Bedtime	AM	PM	Bedtime	AM	PM	Bedtime	AM	PM	Bedtime
n= 26												
	164.31	185.00	148.19	132.96	180.31	130.23	159/04	151.31	115.42	152.69	192.04	168.00
	(105.36)	(98.29)	(110.38)	(83.08)	(96.80)	(106.97)	(100.60)	(101.61)	(91.18)	(100.74)	(113.51)	(141.78)

Appendix F

Pearson correlations between individual differences factors and state indices for day 1 (day prior to task).

	<i>CAR</i>	<i>AUC<sub>G</sub></i>	<i>State</i>	<i>State</i>	<i>Stress</i>	<i>Happiness</i>	<i>Thinking</i>	<i>Mental</i>	<i>Physical</i>	<i>Wellness</i>
	<i>magnitude</i>		<i>anxiety</i>	<i>anxiety</i>	<i>AM</i>	<i>AM</i>	<i>about task</i>	<i>alertness</i>	<i>tension</i>	
	<i>/nmol</i>		<i>AM</i>	<i>Bed time</i>				<i>night</i>	<i>night</i>	
								<i>before</i>	<i>before</i>	
<i>Health complaints</i>	r=.008	r=-.050	r=.237*	r=.345*	r=.109	r=-.331*	r=-.031	r=.052	r=.299*	r=-.310*
	p=.947	p=.711	p=.029	p=.001	p=.319	p=.002	p=.780	p=.636	p=.006	p=.004
	n=81	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85
<i>Extraversion</i>	r=.017	r=.047	r=-.005	r=-.100	r=-.104	r=-.034	r=-.034	r=.095	r=.037	r=.130
	p=.879	p=.723	p=.963	p=.362	p=.345	p=.758	p=.760	p=.391	p=.740	p=.234
	n=81	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85
<i>Agreeableness</i>	r=-.163	r=-.019	r=-.032	r=-.218*	r=-.159	r=.136	r=-.154	r=-.044	r=.054	r=.217*
	p=.146	p=.890	p=.771	p=.045	p=.145	p=.212	p=.159	p=.694	p=.631	p=.047
	n=81	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85

<i>Conscientiousness</i>	r=.097	r=.181	r=-.014	r=-.069	r=-.076	r=-.049	r=-.126	r=-.081	r=.147	r=.176
	p=.391	p=.173	p=.895	p=.533	p=.487	p=.656	p=.252	p=.465	p=.185	p=.107
	n=81	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85
<i>Emotional stability</i>	r=.024	r=.122	r=-.162	r=-.282*	r=-.081	r=.192	r=.018	r=-.002	r=-.125	r=.273*
	p=.833	p=.360	p=.139	p=.009	p=.461	p=.077	p=.870	p=.986	p=.261	p=.011
	n=81	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85
<i>Openness</i>	r=.156	r=-.006	r=.019	r=-.049	r=-.039	r=.016	r=.049	r=.097	r=.105	r=.056
	p=.165	p=.965	p=.866	p=.659	p=.725	p=.884	p=.656	p=.379	p=.344	p=.612
	n=81	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85
<i>Type D</i>	r=-.065	r=-.126	r=.286*	r=.308*	r=.293*	r=-.229*	r=.065	r=.104	r=.149	r=-.426*
	p=.572	p=.351	p=.009	p=.005	p=.008	p=.037	p=.562	p=.356	p=.186	p<.001
	n=78	n=57	n=82	n=82	n=82	n=83	n=82	n=81	n=80	n=82
<i>Trait anxiety</i>	r=.025	r=-.053	r=.245*	r=.446*	r=.214*	r=-.245*	r=.089	r=.004	r=.118	r=-.431*
	p=.824	p=.695	p=.024	p<.001	p=.049	p=.023	p=.419	p=.972	n=.287	p<.001
	n=81	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85

<i>Self esteem</i>	r=.099	r=.174	r=-.154	r=-.305*	r=-.132	r=.166	r=-.010	r=-.014	r=-.077	r=.337*
	p=.384	p=.195	p=.162	p=.005	p=.230	p=.130	p=.926	p=.899	p=.493	p=.002
	n=79	n=57	n=84	n=84	n=84	n=85	n=84	n=83	n=82	n=84
<i>Perceived stress</i>	r=-.071	r=.046	r=.242*	r=.457*	r=.223*	r=-.311*	r=-.025	r=-.009	r=.202	r=-.234*
	p=.534	p=.733	p=.026	p<.001	p=.042	p=.004	p=.821	p=.932	p=.069	p=.032
	n=79	n=57	n=84	n=84	n=84	n=85	n=84	n=83	n=82	n=84
<i>Prospective memory</i>	r=.007	r=-.115	r=.121	r=.038	r=.098	r=-.162	r=-.025	r=-.222	r=.009	r=-.229
	p=.956	p=.470	p=.339	p=.762	p=.439	p=.196	p=.843	p=.081	p=.945	p=.069
	n=60	n=42	n=65	n=65	n=65	n=65	n=64	n=63	n=63	p=64
<i>Retrospective memory</i>	r=.061	r=-.106	r=-.021	r=-.028	r=.109	r=-.013	r=-.108	r=-.091	r=.059	r=-.163
	p=.643	p=.506	p=.871	p=.826	p=.388	p=.918	p=.395	p=.476	p=.648	p=.198
	n=60	n=42	n=65	n=65	n=65	n=65	n=64	n=63	n=63	n=64
<i>Perseverative thinking:</i>	r=.124	r=-.097	r=.199	r=.309*	r=.107	r=-.229	r=.047	r=-.010	r=.224	r=-.326*
<i>Core</i>	p=.350	p=.550	p=.118	p=.014	p=.405	p=.069	p=.716	p=.940	p=.080	p=.009
	n=59	n=40	n=63	n=63	n=63	n=64	n=63	n=62	n=62	n=63

<i>Unproductive</i>	r=.044	r=.069	r=.255*	r=.287	r=.074	r=-.293*	r=.141	r=.017	r=.267*	r=-.349*
	p=.739	p=.671	p=.044	p=.023	p=.567	p=.019	p=.270	p=.898	p=.036	p=.005
	n=59	n=40	n=63	n=63	n=63	n=64	n=63	n=62	n=62	n=63
<i>Difficulty to disengage</i>	r=.034	r=.058	r=.266*	r=.209	r=.210	r=-.193	r=-.040	r=.111	r=.369*	r=-.278*
	p=.797	p=.722	p=.035	p=.101	p=.098	p=.126	p=.756	p=.390	p=.003	p=.028
	n=59	n=40	n=63	n=63	n=63	n=64	n=63	n=62	n=62	n=63
<i>Coping:</i>	r=.088	r=-.037	r=.092	r=.152	r=.141	r=-.140	r=.082	r=.227*	r=-.034	r=-.158
<i>Self distraction</i>	p=.437	p=.783	p=.400	p=.164	p=.198	p=.200	p=.455	p=.031	p=.758	p=.147
	n=80	n=58	n=85	n=85	n=85	n=86	n=85	n=90	p=83	n=85
<i>Active coping</i>	r=.072	r=.089	r=-.151	r=-.213*	r=-.166	r=.154	r=-.026	r=-.145	r=-.133	r=.018
	p=.527	p=.507	p=.167	p=.050	p=.129	p=.158	p=.810	p=.187	p=.232	p=.871
	n=80	n=58	n=85	n=85	n=85	n=86	m=85	n=84	n=83	n=85
<i>Denial</i>	r=.047	r=.066	r=.245*	r=.262*	r=.175	r=-.117	r=.137	r=.000	r=.293*	r=-.146
	p=.682	p=.623	p=.024	p=.015	p=.109	p=.285	p=.212	p=1.00	p=.007	p=.184
	n=80	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85

<i>Substance use</i>	r=.023	r=-.075	r=.104	r=.317*	r=.076	r=-.137	r=.149	r=-.083	r=.095	r=-.149
	p=.841	p=.575	p=.343	p=.003	p=.492	p=.209	p=.173	p=.452	p=.394	p=.173
	n=80	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85
<i>Emotional support</i>	r=-.020	r=-.105	r=.041	r=.018	r=-.011	r=-.038	r=-.110	r=.071	r=-.066	r=-.033
<i>seeking</i>	p=.861	p=.428	p=.707	p=.871	p=.924	p=.728	p=.315	p=.520	p=.551	r=.768
	n=80	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85
<i>Instrumental support</i>	r=-.051	r=-.028	r=-.039	r=-.123	r=-.119	r=.159	r=-.059	r=.125	r=-.145	r=-.016
<i>seeking</i>	p=.652	p=.835	p=.721	p=.263	p=.276	p=.150	p=.593	p=.259	p=.192	p=.885
	n=80	n=58	n=85	n=85	n=86	n=86	n=85	n=84	n=83	n=85
<i>Behavioural</i>	r=-.010	r=-.293*	r=.180	r=.286*	r=.004	r=-.093	r=.068	r=.025	r=.085	r=-.146
<i>disengagement</i>	p=.931	p=.026	p=.099	p=.008	p=.970	p=.392	p=.539	p=.818	p=.443	p=.182
	n=80	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85
<i>Venting</i>	r=.001	r=.081	r=.112	r=-.019	r=-.014	r=-.158	r=-.049	r=-.083	r=-.093	r=-.090
	p=.992	p=.546	p=.309	p=.863	p=.896	p=.145	p=.655	p=.454	p=.403	p=.411
	n=80	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85

<i>Positive reframing</i>	r=-.024	r=.284*	r=-.095	r=-.103	r=-.020	r=.163	r=-.104	r=.190	r=.028	r=.046
	p=.830	p=.031	p=.386	p=.347	p=.858	p=.134	p=.343	p=.083	p=.800	p=.676
	n=80	n=58	n=85	n=85	n=85	n=86	p=85	n=84	n=83	n=85
<i>Planning</i>	r=.137	r=.163	r=-.141	r=-.229*	r=-.123	r=.122	r=-.089	r=.012	r=-.067	r=.086
	p=.227	p=.223	p=.199	p=.035	p=.262	p=.262	p=.420	p=.912	p=.549	p=.432
	n=80	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85
<i>Humour</i>	r=.150	r=.136	r=.029	r=.074	r=-.094	r=-.079	r=.027	r=.075	r=-.025	r=-.135
	p=.184	p=.310	p=.790	p=.500	p=.390	p=.468	p=.808	p=.500	p=.825	p=.217
	n=80	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85
<i>Acceptance</i>	r=.135	r=.260*	r=-.138	r=-.282*	r=-.080	r=.061	r=-.098	r=.096	r=-.154	r=.039
	p=.233	p=.049	p=.208	p=.009	p=.467	p=.578	p=.373	p=.384	p=.165	p=.722
	n=80	n=58	n=85	n=85	n=85	n=86	n=85	n=84	n=83	n=85
<i>Religion</i>	r=-.093	r=.066	r=.115	r=-.052	r=.162	r=.073	r=-.011	r=.104	r=.207	r=-.027
	p=.414	p=.620	p=.296	p=.634	p=.139	p=.505	p=-.103	p=.348	p=.061	p=.807
	n=80	n=58	n=85	n=85	n=85	n=86	n=83	n=84	n=83	p=85

<i>Self blame</i>	r=.151	r=-.130	r=.095	r=.317*	r=.021	r=-.018	r=.177	r=-.077	r=.091	r=-.216*
	p=.181	p=.329	p=.388	p=.003	p=.846	p=.873	p=.109	p=.485	p=.414	p=.047
	n=80	n=58	n=85	n=85	n=85	n=86	n=83	n=84	n=83	n=85

\* indicates  $p < .05$

Pearson correlations between individual differences factors and state indices for day 2 (day of the task).

	CAR magnitude/ nmol	AUC <sub>G</sub>	State anxiety AM	State anxiety Bed time	Stress AM	Happiness AM	Thinking about task	Mental alertness night before	Physical tension night before	Wellness
<i>Health complaints</i>	r=.106 p=.345 n=78	r=-.034 p=.799 n=58	r=.321* p=.003 n=83	r=.409* p<.001 n=78	r=.256* p=.020 n=83	r=-.243* p=.027 n=83	r=-.105 p=.346 n=83	r=.148 p=.181 n=83	r=.117 p=.291 n=86	r=-.146 p=.190 n=82
<i>Extraversion</i>	r=.090 p=.432 n=78	r=.121 p=.367 n=58	r=-.067 p=.545 n=83	r=-.181 p=.112 n=78	r=-.139 p=.209 n=83	r=.145 p=.191 n=83	r=-.164 p=.138 n=83	r=-.118 p=.287 n=83	r=-.102 p=.358 n=83	r=.222* p=.045 n=82
<i>Agreeableness</i>	r=-.075 p=.514 n=78	r=.002 p=.987 n=58	r=-.063 p=.572 n=83	r=-.241* p=.034 n=78	r=-.164 p=.140 n=83	r=.260* p=.017 n=83	r=-.115 p=.301 n=83	r=.236* p=.032 n=83	r=.145 p=.191 n=83	r=.188 p=.091 n=82

<i>Conscientiousness</i>	r = -.043	r = .337*	r = -.075	r = -.121	r = -.090	r = .262*	r = -.117	r = .034	r = -.083	r = .145
	p = .706	p = .010	p = .502	p = .289	p = .416	p = .017	p = .291	p = .758	p = .454	p = .194
	n = 78	n = 58	n = 83	n = 78	n = 83	n = 83	n = 83	n = 83	n = 83	n = 82
<i>Emotional stability</i>	r = -.062	r = .252	r = -.201	r = -.348*	r = -.239*	r = .324*	r = .097	r = -.067	r = -.039	r = .118
	p = .592	p = .056	p = .069	p = .002	p = .029	p = .003	p = .383	p = .546	p = .725	p = .292
	n = 78	n = 58	n = 83	n = 78	n = 83	n = 83	n = 83	n = 83	n = 83	n = 82
<i>Openness</i>	r = -.120	r = .132	r = .169	r = -.035	r = .006	r = .073	r = .015	r = .045	r = .147	r = -.013
	p = .297	p = .324	p = .659	p = .764	p = .954	p = .512	p = .893	p = .689	p = .184	p = .909
	n = 78	n = 58	n = 85	n = 78	n = 83	n = 83	n = 83	n = 89	n = 83	n = 82
<i>Type D</i>	r = -.175	r = -.119	r = .523*	r = .381*	r = .505*	r = -.321*	r = .258*	r = .169	r = .215	r = -.390*
	p = .131	p = .377	p < .001	p = .001	p < .001	p = .004	p = .021	p = .133	p = .055	p < .001
	n = 76	n = 57	n = 80	n = 75	n = 80	n = 80	n = 80	n = 80	n = 80	n = 79
<i>Trait anxiety</i>	r = .016	r = .009	r = .535*	r = .488*	r = -.487*	r = -.409*	r = .145	r = .072	r = .118	r = -.386*
	p = .890	p = .946	p < .001	p < .001	p < .001	p < .001	p = .191	p = .519	p = .287	p < .001
	n = 78	n = 58	n = 83	n = 78	n = 83	n = 83	n = 83	n = 83	n = 83	n = 82

<i>Self esteem</i>	r=-.083	r=.143	r=-.394*	r=-.378*	r=-.418*	r=.284*	r=-.181	r=-.201	r=-.226*	r=.385*
	p=.474	p=.288	p<.001	p=.001	p<.001	p=.010	p=.104	p=.070	p=.041	p<.001
	n=76	n=57	n=82	n=77	n=82	n=82	n=82	n=82	n=82	n=81
<i>Perceived stress</i>	r=-.119	r=-.063	r=.428*	r=.424*	r=.339*	r=-.339*	r=.073	r=.032	r=.149	r=-.276*
	p=.308	p=.642	p<.001	p<.001	p=.002	p=.002	p=.513	p=.777	p=.182	p=.013
	n=76	n=57	n=82	n=77	n=82	n=82	n=82	n=82	n=82	n=81
<i>Prospective memory</i>	r=.134	r=-.150	r=.067	r=.195	r=.062	r=-.175	r=.080	r=-.062	r=-.052	r=-.225
	p=.315	p=.343	p=.602	p=.139	p=.631	p=.173	p=.535	p=.630	p=.689	p=.082
	n=58	n=42	n=62	n=59	n=62	n=62	n=62	n=62	n=.62	n=61
<i>Retrospective memory</i>	r=-.083	r=.061	r=.086	r=.148	r=-.031	r=-.049	r=.189	r=.037	r=.056	r=-.111
	p=.537	p=.701	p=.505	p=.263	p=.810	p=.705	p=.141	p=.775	p=.663	p=.394
	n=58	n=42	n=62	n=59	n=62	n=62	n=62	n=62	n=62	n=61

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<i>Perseverative thinking:</i>	r=.040	r=-.120	r=.316*	r=.452*	r=.107	r=-.358*	r=.054	r=.075	r=.194	r=-.313*
<i>Core</i>	p=.768	p=.460	p=.013	p<.001	p=.405	p=.005	p=.681	p=.563	p=.134	p=.014
	n=56	n=40	n=61	n=57	n=63	n=61	n=61	n=61	n=61	n=61
<i>Unproductive</i>	r=.051	r=.097	r=.329*	r=.496*	r=.074	r=-.351*	r=.067	r=.035	r=.171	r=-.326*
	p=.707	p=.552	p=.010	p<.001	p=.567	p=.006	p=.606	p=.791	p=.187	p=.010
	n=56	n=40	n=61	n=57	n=63	n=61	n=61	n=61	n=61	n=61
<i>Difficulty to disengage</i>	r=-.041	r=.206	r=.259*	r=.418*	r=.210	r=-.209	r=-.058	r=-.046	r=-.037	r=-.138
	p=.762	p=.202	p=.044	p=.001	p=.098	p=.105	p=.658	p=.727	p=.776	p=.290
	n=56	n=40	n=61	n=57	n=63	n=61	n=61	n=61	n=61	n=61
<i>Coping:</i>	r=-.068	r=-.134	r=.110	r=.259*	r=-.028	r=-.233*	r=.000	r=.014	r=.108	r=-.067
<i>Self distraction</i>	p=.554	p=.317	p=.320	p=.022	p=.803	p=.034	p=.997	p=.899	p=.333	p=.551
	n=77	n=58	n=85	n=78	n=83	n=83	n=91	n=83	n=83	n=82

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<i>Active coping</i>	r=.047	r=.037	r=-.151	r=-.136	r=-.230*	r=.041	r=-.159	r=-.023	r=.048	r=.202
	p=.620	p=.781	p=.173	p=.234	p=.036	p=.715	p=.151	p=.836	p=.668	p=.066
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82
<i>Denial</i>	r=-.032	r=.111	r=.317*	r=.391*	r=.303*	r=-.081	r=.122	r=.117	r=.175	r=-.071
	p=.786	p=.406	p=.004	p<.001	p=.005	p=.469	p=.274	p=.292	p=.113	p=.525
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82
<i>Substance use</i>	r=.170	r=-.289*	r=.042	r=.129	r=.075	r=-.152	r=.152	r=-.022	r=-.003	r=-.055
	p=.140	p=.028	p=.708	p=.259	p=.502	p=.171	p=.169	p=.844	p=.978	p=.623
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82
<i>Emotional support seeking</i>	r=.073	r=-.340*	r=-.024	r=.018	r=-.039	r=-.190	r=-.135	r=.015	r=-.156	r=-.095
	p=.527	p=.009	p=.830	p=.875	p=.726	p=.085	p=.223	p=.895	p=.159	p=.396
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82
<i>Instrumental support seeking</i>	r=.091	r=-.176	r=-.055	r=-.151	r=-.175	r=-.094	r=-.101	r=-.033	r=-.180	r=.013
	p=.430	p=.186	p=.623	p=.188	p=.114	p=.396	p=.366	p=.768	p=.104	p=.906
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82

<i>Behavioural</i>	r=-.150	r=-.245	r=.316*	r=.318*	r=.285*	r=-.112	r=.124	r=.069	r=.085	r=-.220*
<i>disengagement</i>	p=.895	p=.064	p=.004	p=.005	p=.009	p=.305	p=.262	p=.532	p=.443	p=.047
	n=77	n=58	n=83	n=78	n=83	n=86	n=83	n=83	n=83	n=82
<i>Venting</i>	r=.096	r=-.031	r=.087	r=.053	r=.043	r=-.215	r=.098	r=.097	r=.002	r=-.220*
	p=.407	p=.820	p=.433	p=.646	p=.698	p=.051	p=.376	p=.381	p=.983	p=.047
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82
<i>Positive reframing</i>	r=-.106	r=.185	r=-.051	r=-.088	r=-.113	r=-.068	r=-.196	r=-.178	r=-.091	r=.118
	p=.360	p=.164	p=.645	p=.445	p=.307	p=.542	p=.076	p=.106	p=.413	p=.290
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82
<i>Planning</i>	r=-.064	r=.181	r=-.149	r=-.140	r=-.176	r=-.122	r=-.074	r=-.046	r=-.076	r=.027
	p=.583	p=.175	p=.179	p=.220	p=.112	p=.273	p=.507	p=.678	p=.497	p=.811
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82
<i>Humour</i>	r=-.004	r=.216	r=.088	r=.075	r=.041	r=-.218*	r=.019	r=-.066	r=-.011	r=-.225*
	p=.970	p=.103	p=.429	p=.514	p=.714	p=.046	p=.863	p=.552	p=.921	p=.042
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82

<i>Acceptance</i>	r=.062	r=.282*	r=-.083	r=-.065	r=-.229*	r=-.094	r=.080	r=.104	r=-.021	r=-.006
	p=.595	p=.032	p=.457	p=.571	p=.037	p=.397	p=.472	p=.348	p=.848	p=.958
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82
<i>Religion</i>	r=-.188	r=.087	r=.047	r=-.078	r=-.090	r=.119	r=-.103	r=.012	r=.086	r=.125
	p=.102	p=.516	p=.674	p=.496	p=.417	p=.285	p=.352	p=.915	p=.439	p=.262
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82
<i>Self blame</i>	r=.057	r=-.121	r=.355*	r=.314*	r=.328*	r=-.258*	r=.177	r=.106	r=.199	r=-.239*
	p=.622	p=.367	p=.001	p=.005	p=.002	p=.019	p=.109	p=.339	p=.071	p=.031
	n=77	n=58	n=83	n=78	n=83	n=83	n=83	n=83	n=83	n=82

\* indicates  $p < .05$

Pearson correlations between individual differences factors and state indices for day 3 (control day).

	<i>CAR</i>	<i>AUC<sub>G</sub></i>	<i>State</i>	<i>State</i>	<i>Stress</i>	<i>Happiness</i>	<i>Thinking</i>	<i>Mental</i>	<i>Physical</i>	<i>Wellness</i>
	<i>magnitude/</i>		<i>anxiety</i>	<i>anxiety</i>	<i>AM</i>	<i>AM</i>	<i>about task</i>	<i>alertness</i>	<i>tension</i>	
	<i>nmol</i>		<i>AM</i>	<i>Bed time</i>				<i>night</i>	<i>night</i>	
								<i>before</i>	<i>before</i>	
<i>Health complaints</i>	r=-.112	r=.208	r=.249*	r=.255*	r=.121	r=-.161	r=.059	r=.167	r=.137	r=-.044
	p=.322	p=.151	p=.021	p=.019	p=.270	p=.139	p=.587	p=.126	p=.210	p=.690
	n= 81	n=49	n=86	n=85	n=85	n=86	n=87	n=85	n=85	n=85
<i>Extraversion</i>	r=-.022	r=.092	r=-.175	r=-.276*	r=-.117	r=.083	r=-.052	r=-.047	r=.011	r=.102
	p=.843	p=.529	p=.108	p=.011	p=.287	p=.449	p=.637	p=.667	p=.923	p=.352
	n=81	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	p=85
<i>Agreeableness</i>	r=.143	r=-.086	r=-.238*	r=-.257*	r=-.318*	r=.092	r=.051	r=-.082	r=-.137	r=-.071
	p=.203	p=.557	p=.028	p=.017	p=.003	p=.399	p=.643	p=.455	p=.212	p=.519
	n=81	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85

<i>Conscientiousness</i>	r=.096	r=.224	r=-.263*	r=-.208	r=-.290*	r=-.022	r=-.108	r=-.194	r=-.086	r=.224*
	p=.396	p=.122	p=.014	p=.057	p=.007	p=.839	p=.322	p=.075	p=.434	p=.040
	n=81	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85
<i>Emotional stability</i>	r=-.055	r=.156	r=-.256*	r=-.315*	r=-.214*	r=.221*	r=-.123	r=-.296*	r=-.225*	r=.102
	p=.623	p=.285	p=.017	p=.003	p=.049	p=.041	p=.260	p=.006	p=.039	p=.355
	n=81	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85
<i>Openness</i>	r=-.006	r=.067	r=-.271*	r=-.119	r=-.204	r=.012	r=.053	r=-.207	r=-.121	r=.312*
	p=.960	p=.648	p=.012	p=.279	p=.061	p=.909	p=.630	p=.057	p=.271	p=.004
	n=81	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85
<i>Type D</i>	r=-.063	r=-.037	r=.159	r=.295*	r=.192	r=-.111	r=.129	r=.301*	r=.088	r=-.027
	p=.585	p=.805	p=.152	p=.007	p=.083	p=.320	p=.245	p=.006	p=.430	p=.807
	n=78	n=48	n=83	n=82	n=82	n=83	n=83	n=82	n=82	n=82
<i>Trait anxiety</i>	r=-.105	r=.034	r=.334*	r=.361*	r=.322*	r=-.226*	r=.043	r=.280*	r=.214*	r=-.134
	p=.350	p=.818	p=.002	p=.001	p=.003	p=.037	p=.692	p=.009	p=.049	p=.221
	n=81	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85

<i>Self esteem</i>	r=.028	r=.008	r=-.306*	r=-.198	r=-.214*	r=.201	r=.052	r=-.309*	r=-.154	r=.177
	p=.809	p=.955	p=.004	p=.071	p=.050	p=.065	p=.633	p=.004	p=.161	p=.107
	n=79	n=48	n=85	n=84	n=84	n=85	n=85	n=84	n=84	n=84
<i>Perceived stress</i>	r=-.094	r=.007	r=.335*	r=.340*	r=.311*	r=-.287*	r=-.001	r=.193	r=.220*	r=-.245*
	p=.408	p=.965	p=.002	p=.002	p=.004	p=.008	p=.996	p=.079	p=.045	p=.025
	n=79	n=48	n=85	n=84	n=84	n=85	n=85	n=84	n=84	n=84
<i>Prospective memory</i>	r=.188	r=-.097	r=.205	r=.241	r=.183	r=-.249*	r=.152	r=.007	r=-.008	r=-.181
	p=.150	p=.581	p=.101	p=.054	p=.147	p=.045	p=.227	p=.955	p=.950	p=.152
	n=60	n=35	n=65	n=65	n=64	n=65	n=65	n=64	n=64	n=64
<i>Retrospective memory</i>	r=.080	r=.083	r=.017	r=.107	r=-.050	r=.014	r=.222	r=-.047	r=-.045	r=-.055
	p=.544	p=.635	p=.892	p=.396	p=.697	p=.911	p=.076	p=.710	p=.723	p=.667
	n=60	n=35	n=65	n=65	n=64	n=65	n=65	n=64	n=64	n=64

<i>Perseverative</i>	r=-.077	r=-.343*	r=.089	r=.302*	r=.064	r=-.168	r=-.024	r=.165	r=.143	r=-.050
<i>thinking:</i>	p=.568	p=.044	p=.485	p=.016	p=.617	p=.185	p=.852	p=.193	p=.259	p=.693
<i>Core</i>	n= 58	n=35	n=64	n=63	n=63	n=64	n=64	n=64	n=64	n=64
<i>Unproductive</i>	r=-.076	r=.326	r=.066	r=.342*	r=.101	r=-.080	r=-.052	r=.071	r=.158	r=.028
	p=.570	p=.056	p=.604	p=.006	p=.429	p=.531	p=.683	p=.575	p=.212	p=.828
	n=58	n=35	n=64	n=63	n=63	n=64	n=64	n=64	n=64	n=64
<i>Difficulty to</i>	r=.039	r=.173	r=-.014	r=.319*	r=.062	r=-.075	r=.138	r=.005	r=.036	r=-.087
<i>disengage</i>	p=.770	p=.320	p=.912	p=.011	p=.628	p=.555	p=.278	p=.967	p=.779	p=.494
	n=58	n=35	n=64	n=63	n=63	n=64	n=64	n=64	n=64	n=64
<i>Coping:</i>	r=-.096	r=.051	r=.400*	r=.324*	r=.397*	r=-.256*	r=.097	r=.142	r=.178	r=-.074
<i>Self distraction</i>	p=.397	p=.729	p<.001	p=.003	p<.001	p=.017	p=.372	p=.194	p=.103	p=.486
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=91
<i>Active coping</i>	r=-.077	r=.305*	r=-.001	r=-.082	r=-.075	r=-.096	r=.068	r=-.096	r=-.050	r=-.165
	p=.496	p=.033	p=.996	p=.456	p=.497	p=.378	p=.535	p=.380	p=.648	p=.132
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85

<i>Denial</i>	r=.059	r=.000	r=.099	r=.136	r=.148	r=-.057	r=-.009	r=-.013	r=.207	r=-.041
	p=.603	p=.999	p=.365	p=.215	p=.178	p=.602	p=.934	p=.904	p=.057	p=.712
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85
<i>Substance use</i>	r=-.217	r-.045	r=.425*	r=.292*	r=.410*	r=-.291*	r=-.002	r=.145	r=.173	r=-.198
	p=.053	p=.759	p<.001	p=.007	p=.001	p=.007	p=.986	p=.187	p=.113	p=.069
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85
<i>Emotional support seeking</i>	r=-.232*	r=.076	r=.089	r=.089	r=.006	r=-.158	r=-.073	r=.168	r=-.085	r=-.217*
	p=.038	p=.605	p=.414	p=.414	p=.953	p=.145	p=.503	p=.125	p=.439	p=.048
	n=80	n=49	n=86	n=86	n=85	n=86	n=86	n=85	n=85	n=85
<i>Instrumental support seeking</i>	r=-.135	r=.149	r=.034	r=.078	r=-.058	r=-.023	r=.028	r=.148	r=-.159	r=-.140
	p=.232	p=.307	p=.759	p=.477	p=.597	p=.834	p=.795	p=.176	p=.147	p=.201
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85
<i>Behavioural disengagement</i>	r=-.209	r=.114	r=.223*	r=.264*	r=.177	r=-.112	r=.016	r=.271*	r=.081	r=-.207
	p=.063	p=.436	p=.039	p=.015	p=.106	p=.305	p=.885	p=.012	p=.460	p=.057
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85

<i>Venting</i>	r=-.103	r=.040	r=.149	r=-.002	r=.086	r=-.173	r=.110	r=.168	r=.066	r=-.202
	p=.362	p=.786	p=.170	p=.982	p=.436	p=.111	p=.315	p=.125	p=.550	p=.063
	n= 80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85
<i>Positive reframing</i>	r=.030	r=.081	r=-.058	r=.019	r=.066	r=-.005	r=.100	r=.048	r=-.011	r=.015
	p=.792	p=.579	p=.599	p=.865	p=.551	p=.964	p=.359	p=.665	p=.923	p=.889
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85
<i>Planning</i>	r=.028	r=.278	r=-.077	r=-.047	r=-.038	r=-.026	r=.070	r=.024	r=-.062	r=.067
	p=.804	p=.053	p=.482	p=.671	p=.728	p=.812	p=.519	p=.826	p=.571	p=.540
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85
<i>Humour</i>	r=.112	r=-.063	r=.110	r=.128	r=.239*	r=-.129	r=.024	r=-.120	r=-.066	r=.079
	p=.321	p=.665	p=.315	p=.244	p=.028	p=.238	p=.829	p=.272	p=.546	p=.472
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85
<i>Acceptance</i>	r=.017	r=.256	r=-.119	r=-.084	r=-.080	r=.037	r=.077	r=.005	r=-.131	r=.107
	p=.883	p=.076	p=.274	p=.447	p=.465	p=.736	p=.484	p=.964	p=.234	p=.328
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85

<i>Religion</i>	r=-.025	r=-.021	r=-.083	r=.108	r=-.004	r=.065	r=.296*	r=-.057	r=-.049	r=.105
	p=.827	p=.886	p=.446	p=.327	p=.971	p=.555	p=.006	p=.603	p=.656	p=.337
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85
<i>Self blame</i>	r=-.031	r=.090	r=.215*	r=.163	r=.198	r=-.063	r=-.112	r=.305*	r=.178	r=-.083
	p=.788	p=.537	p=.047	p=.135	p=.070	p=.566	p=.303	p=.005	p=.103	p=.451
	n=80	n=49	n=86	n=85	n=85	n=86	n=86	n=85	n=85	n=85

\* indicates  $p < .05$

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