Citation: Lovell, Brian (2014) Assessing the psychophysiological pathways that link chronic stress with increased vulnerability for ill health. Doctoral thesis, Northumbria University.

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ASSESSING THE PSYCHOPHYSIOLOGICAL PATHWAYS THAT LINK CHRONIC STRESS WITH INCREASED VULNERABILITY FOR ILL HEALTH

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PhD

2013
ASSESSING THE PSYCHOPHYSIOLOGICAL PATHWAYS THAT LINK CHRONIC STRESS WITH INCREASED VULNERABILITY FOR ILL HEALTH

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A thesis submitted in partial fulfilment of the requirements of the University of Northumbria at Newcastle for the degree of

DOCTOR OF PHILOSOPHY

Research undertaken in the Faculty of Health and Life Sciences

August 2013
ASSESSING THE PSYCHOPHYSIOLOGICAL PATHWAYS THAT LINK CHRONIC STRESS WITH INCREASED VULNERABILITY FOR ILL HEALTH

ABSTRACT

This programme of work investigated the psychophysiological pathways that link chronic stress with increased vulnerability for ill health. Data from study one indicated that atypical patterns of cortisol secretion, widely implicated in the aetiologies of severe pathologic conditions, partially mediated the effect of higher perceived levels of stress on greater incidences of the kinds of common health problems that typically affect young otherwise healthy individuals.

As a logical next step in the programme, studies two and three looked more closely at the psychophysiological consequences of informal caregiving, one prototypical model for chronic stress. Data indicated that caring for child with autism/ADHD exacts a considerable psychophysiological toll on the carer. Indeed, relative to controls, caregivers reported increased psychological morbidity, greater incidences of ill health and reduced social support. Dysregulated immunity, manifested by higher concentrations of the inflammatory marker, C-reactive protein (CRP) was also apparent in the caregivers. In fact, caregivers’ mean concentrations of CRP satisfied clinical criterion for moderate risk of cardiovascular pathologies, compared with low risk in the controls. However, psychological morbidity and incidences of ill health were reduced in caregivers who reported greater social support. Socially supported caregivers also displayed a steeper cortisol awakening response (CAR), which is indicative of more adaptive endocrine functioning. Therefore, interventions that enhance social connectivity might be effective for alleviating caregiver related stress. However, logistical challenges such as a lack of alternate and reliable supervision make it difficult for caregivers to access support related interventions, most of which are time consuming and based outside the home.

Expressive writing on the other hand is a simple and time effective intervention that can run in participants’ homes, and as such, might be especially well suited for informal caregivers. Data from study four indicated that writing about the benefits of caring for a child with autism/ADHD can be applied as a home based intervention, and is associated with clinically meaningful improvements in caregivers’ psychological well being.
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List of Abbreviations

ACTH - adrenocorticotrophin hormone
ADHD - attention deficit hyperactivity disorder
ANCOVA - analysis of covariance
ANOVA - analysis of variance
BMI - body mass index
CAR - cortisol awakening response
CBG - corticosteroid-binding-globulin
CBSM - cognitive behavioural stress management
CCC - classic complement pathway
CD - cluster of differentiation
CNS - central nervous system
CRH - corticotrophin releasing hormone
CRP - C reactive protein
CTL - cytotoxic T lymphocytes
EPRR - endocytic pattern recognition receptor
GC - glucocorticoid
HADS - Hospital Anxiety and Depression Scale
HIV - human immunodeficiency virus
HPA - hypothalamic-pituitary-adrenal
IFN - interferon
IL - interleukin
ISEL - Interpersonal Support Evaluation Checklist
LIWC - linguistic inquiry & word count
MBL - mannan binding-lectin
MHC - major histocompatibility complex
MS - multiple sclerosis
NK - natural killer cells
PILL - Pennebaker Inventory of Limbic Languidness
PSS - Perceived Stress Scale
PTSD - post traumatic stress disorder
PVN - paraventricular nucleus
SAM - sympathetic-adrenal-medullary
S-IgA - secretory immunoglobulin A
TCR - T cell receptor
Th - T helper
TNF - tumour necrosis factor
VAS - visual analogue scale
VIG - video interaction guidance
WED - written emotional disclosure
Conference Proceedings Relevant to the Thesis


Lovell, B., Moss, M., Wetherell. M.A. The psychosocial, endocrine and immune consequences of caring for a child with autism and ADHD. Psychobiology Section, Annual Scientific Meeting, Lake District, September, 2011.
Lovell, B., Moss, M., Wetherell. M.A. The psychosocial, endocrine and immune consequences of caring for a child with autism and ADHD. North East Postgraduate Research Conference, Newcastle University, September, 2011.


Gemma & puppy

To my beautiful wife, Gemma, thank you for your love, kindness and support. I love you, now and always. To my puppy, Cuddles, for the stress relief that is a long walk with a bouncy ball.

Mum and Dad

To my mum and dad, I dedicate this achievement to you. I love you.

My Supervisors

Thanks to my principal supervisor, Dr Mark Wetherell, whose door was always open (but light never on) when I needed it. Above all, thank you for being someone I admire and look up to, someone who never compromises on standards and always expects the best. Thanks also to my stats partner, Dr Mark Moss for helping me realise that a study is only as good as its hessian matrix.

Office Girls

To Sarah and Laura, thank you for being a constant source of distraction and stifling my productivity. In all seriousness, thank you for a PhD experience filled with laughter, fun and ‘skemmy’ tea. Not sure I could have made it through without you.

Financial support

Thanks also to Research Autism and the Waterloo Foundation who funded study four of the thesis.
Authors Declaration

I declare that the work presented here is my own and has not been submitted in fulfilment for any other award. Work presented here satisfies the appropriate academic standards, is without plagiarism and fully acknowledges ideas, opinions and contributions from the work of others. Ethical clearance for all research presented in the thesis was sought and approved by the Faculty of Health and Life Sciences Ethics Committee. This thesis has been supplied on condition that anyone who reads it understands that copyright rests solely with the author. No quotation or other information may be published without prior consent from the author.

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BRIAN LOVELL

Lovel

12/12/2013
1. Introduction

This programme of work aimed to assess the psychophysiological pathways that link chronic stress with increased vulnerability for ill health. Dysregulation of the hypothalamic-pituitary-adrenal (HPA) axis, widely observed in the context of chronically stressful life events (Adam & Gunnar, 2001; Cohen et al., 2006; Fries, Dettenborn, & Kirschbaum, 2009; Saxbe, 2008), has also been implicated in the aetiologies of severe pathologic conditions such as cardiovascular disease (Matthews, Schwartz, Cohen, & Seeman, 2006; Seldenrijk, Hamer, Lahiri, Penninx, & Steptoe, 2012), aspects of the metabolic syndrome (Rosmond, 2005), even earlier mortality (Sephton, Sapolsky, Kraemer, & Spiegel, 2000). However, fewer studies have assessed the physiological processes that might underlie stress related disparities in the kinds of common health problems that most of us frequently experience. To date, research involving young otherwise healthy individuals has individually linked higher perceived levels of stress with atypical patterns of cortisol secretion (Abercrombie et al., 2004; O’Connor et al., 2009), as well as increased vulnerability (Cohen, Tyrrell, & Smith, 1991, 1993) and severity (Cohen, Doyle, & Skoner, 1999) of common health problems such as upper respiratory illness. To extend work in this area, study one examined whether the effect of higher perceived levels of stress on greater subjective reports of common health problems might be mediated by perturbations in basal stress hormone activity.

As a logical next step in the programme, study two used the caregiver control model to look more closely at the psychophysiological consequences of chronic stress. Disruptions in endocrine (Bauer et al., 2000; Brummett et al., 2008; Da Roza Davis & Cowen, 2004; Gallagher-Thompson et al., 2006; Vedhara et al., 1999) and immune parameters (Glaser, Kiecolt-Glaser, Malarkey, & Sheridan, 1998; Kiecolt-
Glaser, Glaser, Gravenstein, Malarkey, & Sheridan, 1996; Segerstrom, Schipper, & Greenberg, 2008; von Kanel, Dimsdale, & Mills et al., 2006), indicative of greater vulnerability for ill health, have been widely observed in chronically stressed caregivers, relative to age matched non-caregiver controls. However, most of this work has been conducted with older caregivers, most commonly spousal carers of dementia patients, who are already contending with pre-existing endocrine and immune impairments associated with their advancing age (i.e., senescence). Therefore, researchers have started to examine whether chronic caregiver stress results in endocrine and immune dysfunction in younger caregivers, who are not contending with age associated alterations in these physiological systems. Findings to date have been mixed. Indeed, dysregulated immunity manifested by poorer antibody response to vaccination (Gallagher, Phillips, Drayson, & Carroll, 2009) and elevated markers of inflammation (Miller et al., 2008; Rohleder, Marin, Ma, & Miller, 2009), has been observed in relatively young caregivers of children with additional complex needs such as autism, a condition characterised by the same kinds of distressing cognitive and behavioural impairments that affect dementia patients. However, immune efficacy seems to be preserved in younger caregivers of children with severe physical impairments such as multiple sclerosis (Vedhara et al., 2002). To date, few studies have assessed the physiological consequences of caregiver stress in younger populations per se, and even fewer, the psychosocial antecedents that might underlie these effects (Gallagher et al., 2009; Vitaliano, Zhang, & Scanlan, 2003). Consequently, study two compared relatively young caregivers of children with autism and attention deficit hyperactivity disorder (ADHD), conditions that share many clinical manifestations of dementia (Baron-Cohen, 2004; Johnson & Reader, 2002), with a normative control group on a range of psychophysiological
outcomes. Social support, typically diminished in younger caregivers (Fletcher, Markoulakis, & Bryden, 2012; Gallagher, Phillips, & Carroll, 2010) and often associated with dysregulated immunity (Lutgendorf, Anderson, Sorosky, Buller, & Lubaroff, 2000; Kiecolt-Glaser, Gouin, & Hantsoo, 2009), was also examined as one psychosocial pathway that might link chronic caregiver stress with immune dysfunction.

Research has indicated that socially supported caregivers might be protected against the harmful effects associated with the caregiver experience. Indeed, lower levels of psychological distress (Bozo, Anahar, Ates, & Etel, 2010; Smith, Greenberg, & Seltzer, 2011; Pozo, Sarriá, & Brioso, in press; Weiss, 2002; White & Hastings, 2004) and fewer incidences of adverse health (Lin et al., 2009; Sawyer et al., 2010) have been reported in socially supported caregivers of children with additional complex needs. However, fewer studies have examined whether greater social support might protect relatively young caregivers against maladaptive changes in disease relevant physiological processes. In a recent study, Gallagher and Whiteley (2012a) demonstrated that socially supported caregivers of children with additional complex needs such as autism were protected against higher daytime systolic blood pressure, one well established risk factor for cardiovascular related pathologies (Hamer, Chida, & Stamatakis, 2010). To extend the relatively small research base in this area, study three assessed whether greater social support might ameliorate psychological distress, and in so doing, protect relatively young autism/ADHD caregivers against perturbations in basal HPA activity and the immune response.

Quality of life for the care recipient is heavily contingent on the health and happiness of the care provider (Edworthy, 2005; Reinhard, Given, Huhtala-Petlick,
& Bemis, 2008; Schor, 2003), and as such, it becomes important to develop interventions that mitigate the harmful effects of caregiver stress with a view to improving care recipients’ quality of life. Cognitive behavioural therapy (Gallagher-Thompson & Steffen, 1994; Losada-Baltar, Trocónizal-Fernandez, Montorio-Cerrato, Marquez-Gonzalez, & Perez-Red, 2004; Vedhara, et al., 2003), formal aid from respite services (Garcés, Carretero, Ródenas, & Alemán, 2010; Zarit, Gaugler, & Jarrot, 1999), psychoeducational skills training (Gallagher-Thompson, Arean, Rivera, & Thompson, 2001; Sörensen, Pinquart, & Duberstein, 2002) and socially supportive interventions such as community support groups (Chien et al., 2011), while potentially effective for coping with chronic caregiver stress, are often expensive, time consuming and typically based outside the home. Therefore, these varied interventions are often inaccessible to informal caregivers, many of who experience severe financial hardship (Donelan et al., 2002; Kogan et al., 2008; Parish & Cloud, 2006), and owing to logistical/practical challenges such as difficulties arranging alternative and reliable supervision (Bank, Arguelles, Rubert, Eisdorfer, & Czaja, 2006), are often confined to the home (Yantzi, Rosenberg, & McKeever, 2006).

Written emotional disclosure (WED) on the other hand is a time and cost effective intervention that requires participants to write expressively about any stressful/traumatic life event for 15-20 minutes on three-four consecutive days (Pennebaker, Kiecolt-Glaser, & Glaser, 1988). The WED paradigm, typically applied in a controlled lab setting, has been modified to run in participants’ homes (Wetherell et al., 2005), and as such, might be especially well suited for informal caregivers. To date, WED has been linked with positive psychophysiological adjustments in the context of chronically stressful life events such as bereavement.
(Kovac & Range, 2000) and breast cancer (Rosenberg et al., 2002; Stanton et al., 2002), but not informal caregiving (Barton & Jackson, 2008; Mackenzie, Wiprzycka, Hasher, & Goldstein, 2007; Schwartz & Drotar, 2004). However, in a modification to the traditional WED paradigm, reduced anxiety and depression scores were recently reported in caregivers lower in alexithymia (and therefore, better able to express their emotions linguistically), who wrote about positive life experiences (Ashley, O’Connor, & Jones, 2011). Neatly dovetailing with these findings, caregivers have often reported finding positive consequences (i.e., benefits) amidst the stress of the caregiver experience such as greater appreciation for life/loved ones and improved personal relationships (Kayfitz, Gragg, & Orr, 2010; Fletcher, Markoulakis, & Bryden, 2012; Samios, Pakenham, & Sofronoff, 2009). Interestingly, writing about the benefits associated with chronically stressful life events has been linked with adaptational outcomes including reduced psychological morbidity and fewer physical health complaints (Henry, Schlegel, Talley, Molix, & Bettencourt, 2010; Stanton et al., 2002). To date, only one study has applied a positive written disclosure intervention in the context of chronic caregiver stress (Ashley et al., 2011). To address the paucity of research in this area, study four assessed the feasibility and efficacy of at-home written benefit finding intervention on psychophysiological outcomes in caregivers of children with autism/ADHD.
1.0 Literature Review

1.1 Chapter Overview

This chapter provides a review of current literature that links chronic stress with increased vulnerability for ill health via interactions between the hypothalamic-pituitary-adrenal (HPA) axis and immune system. Sources have been carefully selected as the most appropriate for establishing necessary context for the programme of work. Particular emphasis has been given to informal caregiving, one well established model for chronic stress. Limitations of existing literature are discussed as a rationale for the series of studies presented throughout the thesis.

1.2 Stress, the Stress System and Allostasis

In their transactional model, Lazarus and Folkman (1987) describe a two level appraisal process that determines our psychophysiological reactions to environmental challenges, or stressors. At level one (primary appraisal), the host appraises external events for their threat value, and at level two (secondary appraisal), options for coping with perceived threats (identified at level one) are evaluated. The transactional model stipulates that a psychological stress response, composed of negative cognitive and emotional states, is elicited when perceived threats in the environment exceed the coping resources of the host. Indeed, changes in psychological symptoms reliably quantify what happens to the host during stressful experiences. For example, subjective reports of anxiety (Wetherell et al., 2006), jitteriness (Childs, Vicini, & De Wit, 2006), depression (al’Absi et al., 1997), tension (von Dawans, Kirschbaum, & Heinrichs, 2011), perceived stress and emotional insecurity (Hellhammer & Schubert, 2012) are markedly elevated in response to acute (i.e., short term) stress paradigms in laboratory settings. Acute
reactivity studies have often been criticised for using stress paradigms such as mental arithmetic (Phillips, Der, & Carroll, 2009), mirror tracing (Sawada, Nagano, & Tanaka, 2002), cold pressor (McRae et al., 2006) and the stroop task (Bachen et al., 1992; Gerra et al., 2001) that fail to approximate the kinds of momentary stressors encountered in everyday life. Indeed, acute lab based paradigms should provide a psychophysiological snapshot of how a person would respond to stress in the real world. To enhance the ecological validity of lab based reactivity studies, researchers have begun to develop acutely stressful tasks that are more socially salient. For example, Wetherell and Sidgreaves (2005) reported a significant increase in perceived levels of stress and frustration in response to a computer based multitasking stressor designed to approximate the cumulative stress of working in a busy office environment. Elevations in psychological symptoms such as anxiety and tension have also been reported in response to acute socially evaluative performance stressors such as simulated public speaking (Childs, Vinci, & De Wit, 2006; Kirschbaum, Klauer, Filipp, & Hellhammer, 1995; von Dawans, Kirschbaum, & Heinrichs, 2011), and the stress of interviews (Dimsdale, Stern, & Dillon, 1988). Indeed, researchers have demonstrated that acute lab based paradigms that incorporate elements of social evaluative threat elicit enhanced psychopsychological responsivity (Dickerson & Kemeny, 2004). Psychological adjustments such as increased feelings of anger, vigilance and perceived levels of stress have also been reported in response to momentary socially evaluative stressors in the real world such as delivering a public academic lecture (Dimsdale, 1984), and in competitive ballroom dancers on the day of a competition (Rohleder, Beulen, Chen, Wolf, & Kirschbaum, 2007).
Negative cognitive and emotional states represent the proximal pathway by which stress gets under the skin to modify the physiology of the body (Cohen et al., 1993; Polk, Cohen, Doyle, Skoner, & Kirschbaum, 2005). The paraventricular nucleus (PVN) of the hypothalamus and sympathetic neurons of the brainstem autonomic nuclei represent the central (nervous system) components of the stress system, and respectively regulate the peripheral activities of the sympathetic adrenal medullary (SAM) and hypothalamic-pituitary-adrenal (HPA) axes. When threats in the environment exceed the coping resources of the host, parvocellular neurons of the dorsal and ventral regions of the PVN of the hypothalamus release corticotrophin releasing hormone (CRH) and activate sympathetic nerve fibres in the brainstem. Preganglionic sympathetic fibres leave the central nervous system (CNS) via thoracic and lumbar spinal nerves and synapse with postganglionic fibres in bilateral chains of autonomic ganglia. By releasing noradrenaline at the neuroeffector junction (the point at which a nerve impulse is translated into motor action), postganglionic fibres innervate target tissues of the vasculature, heart, skeletal muscles and kidneys. As an exception to this rule, motor axon terminals of preganglionic sympathetic fibres secrete acetylcholine and synapse with chromaffin cells of the adrenal medulla (that are anatomically homologous to cells of autonomic ganglia) to release adrenaline, and to a lesser extent, noradrenaline, into the bloodstream as endocrine messengers (Chrousos, 2000, 2009; Lovallo, 2005). In addition, parvocellular neurons of the medial division of the PVN of the hypothalamus secrete CRH and vasopressin into the hypophyseal portal venous plexus, a network of capillaries and the primary way station to the anterior pituitary. Here, CRH initiates the release of adrenocorticotrophin hormone (ACTH) by acting on basophilic cells. ACTH is carried by the peripheral circulation to the zona fasciculata of the adrenal cortex.
where it triggers the secretion of glucocorticoid (GC) hormone, cortisol.

Concentrations of cortisol are regulated by a negative feedback circuit, such that GCs inhibit CRH secretion at the level of the hypothalamus, and in so doing, prevent the subsequent release of ACTH from the anterior pituitary (Sapolsky, 1998). A simplified presentation of central and peripheral components of the stress system is displayed in Figure 1.1.

*Figure 1.1* Simplified representation of the central and peripheral components of the stress system (adapted from Lovallo, 2005). **ACET**, acetylcholine; **ACTH**, adrenocorticotrophin hormone; **AD**, adrenaline; **CORT**, cortisol; **CRH**,
corticotrophin releasing hormone; **NA**, noradrenaline; **PVN**, paraventricular nucleus; **VAS**, vasopressin.

Unsurprisingly, elevations in outputs of the SAM (e.g., adrenaline, noradrenaline) and HPA (e.g., cortisol) axes have been widely evidenced in response to acutely stressful paradigms in laboratory settings (Cacioppo et al., 1998; Childs et al., 2006; Foley & Kirschbaum, 2010; Gerra et al., 2001; Schwabe, Haddad, & Schachinger, 2008; Wetherell et al., 2006). In response to acutely stressful emergencies, outputs of the stress system act on receptors in target tissues and organs to produce adaptive physiological and behavioural changes referred to as allostasis (McEwen et al., 1997). Catecholamines, the collective term for outputs of the SAM system, are responsible for the immediate response of the body to stress. That is, enhanced secretion of noradrenaline from postganglionic sympathetic nerve terminals and adrenaline from the adrenal medulla increase cardiovascular activity (i.e., heart rate and blood pressure), and by counteracting the anabolic effects of insulin, mobilise metabolic support for vigorous physical activities such as fight or flight behaviours. Cortisol on the other hand acts at a slower rate than the immediate effects of catecholamines, such that concentrations typically peak between 20-30 minutes post stressor onset (Kirschbaum, Klauer, Filipp, & Hellhammer, 1995). Cortisol, by synergising with outputs of the SAM system, permissively augments cardiovascular arousal and works to increase the release of stored glucose and fat (Juster, McEwen, & Lupien, 2010; Sapolsky, Romero, & Munck, 2000). Moreover, by shutting down less immediately essential activities such as digestive function, reproduction and growth, cortisol conserves valuable energy for more critical needs (e.g., energy mobilisation to exercising muscle) in the face of immediate challenges.
(Chrousos, 2009; Sapolsky, 2000). Research has indicated that, in the same way adrenocortical hormones gear up the cardiovascular and metabolic systems, they also prepare the immune system for impending challenges. Indeed, in the face of an acutely stressful emergency, adrenocortical hormones mediate the adaptive redeployment of leukocytes from the blood to primary defence barriers (i.e., skin) in preparation for potential antigenic invasion (Dhabhar & McEwen, 1997). Indeed, the adaptive effects of stress hormones on immune function under acutely stressful conditions have been widely evidenced (Ackerman, Martino, Heyman, Moyna, & Rabin, 1998; Altemus, Rao, Dhabhar, Ding, & Granstein, 2001; Dhabhar et al., 2010).

Unlike the acutely physical fight or flight emergencies faced by our cousins in the animal kingdom, human experience is replete with chronic (i.e., enduring) psychological stressors such as unemployment (Steptoe, Brydon, & Kunz-Ebrecht, 2005), relationship problems (Adam & Gunnar, 2001) and financial hardship (Cohen et al., 2006) that are impossible to fight off or flee from, and tend to resurface time and time again. While adaptive in the short term, physiological mediators (e.g., cortisol) can, if over secreted by repeated stress, lead to cumulative wear and tear on target cells and organs in ways that foster increased vulnerability for a range of pathophysiological outcomes referred to as allostatic load. For example, by increasing blood pressure, outputs of the stress system serve the body well in the short term by providing physiological support for fight or flight. However, sustained changes in blood pressure as a result of repeated stress can damage the interior lining of blood vessels, making them perfect docking points for fatty acids mobilised as part of the metabolic stress response, and the subsequent formation of atherosclerotic plaques (Sapolsky, 2000).
If overused by repeated stress, the normal operating levels, or regulatory set points of stress responding systems can shift in ways that predispose a person to illness (Koolhaas et al., 2011). For example, cortisol, the final effector hormone of the HPA axis, displays a robust basal diurnal pattern. That is, concentrations typically reach an acrophase between 30-45 minutes post waking (i.e., cortisol awakening response), decline steadily throughout the day (i.e., diurnal cortisol slope) and reach a nadir at around midnight (Smyth et al., 1997). Measures that summarise aspects of the diurnal cortisol pattern are presented in Figure 1.2. Dysregulation of the HPA axis has been implicated as one physiological marker for accumulated psychosocial stress, and diagnostic indicator for a broad catalogue of severe pathologic conditions. For example, flattening of the diurnal cortisol slope, typically characterised by lower waking and higher evening values, has been observed in the context of chronically stressful life events such as lower socioeconomic status (Cohen et al., 2006), metastatic breast cancer (Abercrombie et al., 2004; Bower et al., 2005), and in children managing chronic stress in the home (Wolf, Nicholls, & Chen, 2008). In addition, flattening of the diurnal cortisol slope has been implicated in the aetiologies of disease states such as chronic fatigue syndrome (Nater et al., 2008), subclinical atherosclerosis (Bhattacharyya, Molloy, & Steptoe, 2008; Matthews et al., 2006; Seldenrijk et al., 2012), even earlier mortality in breast cancer survivors (Sephton et al., 2000).
Researchers have linked alterations in the cortisol awakening response (CAR) with stress related factors such as informal caregiving (De Vugt et al., 2005), higher reported life stress (O’Connor et al., 2009), chronic work overload (Schlotz, Hellhammer, Schulz, & Stone, 2004), and lower socioeconomic status (Wright & Steptoe, 2005). Aberrations in the CAR would also appear to have predictive value for health. Indeed, accelerated progression of the carotid intima-media thickness, one marker of subclinical atherosclerosis (Eller, Netterstrøm, & Allerup, 2005), and clinically verified coronary artery disease (Bhattacharyya, Molloy, & Steptoe, 2008) has been observed in patients who displayed a steeper CAR. On the other hand, attenuation, or blunting of the CAR has been linked with pain and fatigue related disorders such as burnout (Sonnenschein et al., 2007), chronic fatigue syndrome (Roberts, Wessely, Chalder, Papadopoulos, & Cleare, 2004) and vital exhaustion (Chida & Steptoe, 2009).

*Figure 1.2* Measures that summarise aspects of the diurnal cortisol pattern: the cortisol awakening response (CAR) and diurnal cortisol slope.
Dysregulation of the HPA axis, manifested by cortisol hypersecretion across the day, has been observed in the context of chronically stressful life events such as informal caregiving (Vedhara et al., 1999), and in patients enduring the emotional burden associated with metastatic breast cancer (Sephton et al., 2009). Cortisol hypersecretion has also been linked with reduced immune efficacy (Bauer, 2005) and with pathologic conditions such as adverse cardiovascular events (Dekker et al., 2008) and aspects of the metabolic syndrome (Rosmond, 2005). Evidence from a meta analysis indicated that sustained elevated levels of cortisol (as a result of repeated stress) rebound to normal and below normal levels over time (Miller, Chen, & Zhou, 2007). Indeed, reduced bioavailability of cortisol (i.e., cortisol hyposecretion) has been implicated as one physiological mechanism that translates chronically stressful life events into burnout, fibromyalgia and vital exhaustion, a collection of functional somatic syndromes characterised by enhanced pain and fatigue (Fries, Hesse, Hellhammer, & Hellhammer, 2005; Gunnar, & Vasquez, 2001; Heim, Ehlert, & Hellhammer, 2000).

Atypical patterns of cortisol secretion, widely implicated in the aetiologies of severe pathologic conditions (Matthews et al., 2006; Seldenrijk et al., 2012), might also underlie the effects of stress on greater incidences of kinds of health problems (e.g., coughs, colds, running noses, aches and pains etc) that most of us frequently experience. For example, atypical cortisol secretion patterns (Abercrombie et al., 2004; O’Connor et al., 2009), as well as greater susceptibility (Cohen et al., 1991) and severity (Cohen et al., 1999) for common health problems such as upper respiratory illness, have been observed in young otherwise healthy individuals who reported higher perceived levels of stress. However, dysregulated HPA activity was not responsible for increased susceptibility to the common cold in participants who
reported a greater number of stressful life events (Cohen et al., 1998). More recently, abnormal patterns of cortisol secretion have been linked with increased severity of upper respiratory symptoms (Edwards, Hucklebridge, Clow, & Evans, 2003), poorer self reported physical health status (Hagger-Johnson, Whiteman, Wawrzyniak, & Holroyd, 2010; Sjögren, Leanderson, & Kristenson, 2006) and greater incidences of commonly occurring ailments (Hellhammer, Schlotz, Stone, Pirke, & Hellhammer, 2004).

Consequently, **study one** assessed whether differential patterns of cortisol secretion might underlie perceived stress related disparities in the kinds of common health problems that typically affect young otherwise healthy individuals.
1.3 Overview of the Immune System and HPA-Immune Interactions

Foreign invaders (i.e., antigens) that breach primary defence barriers (i.e., the skin) are attacked non-discriminately by cells of the *innate immune response*. Indeed, phagocytic cells (i.e., macrophages) display endocytic pattern recognition receptors (EPRR) that recognise and bind broad molecular patterns on invading antigens. Released by activated macrophages, proinflammatory cytokines such as interleukin-1 (IL-1), interleukin-6 (IL-6) and tumour necrosis factor-alpha (TNF-α) operate via leaky regions of the blood brain barrier and cytokine specific transport molecules to trigger the release of prostaglandins from the anterior hypothalamus (Brydon et al., 2009). Prostaglandins slow the growth and multiplication of the foreign invader by increasing body temperature (i.e., inducing fever); however, the metabolic costs associated with inducing fever leaves little room for behaviours other than those that facilitate heat production such as shivering (Dantzer, 2001). Therefore, proinflammatory cytokines also signal the brain to induce the sickness syndrome, a range of non specific sickness behaviours such as social withdrawal, fatigue, reduced sexual interest and depressive affect that conserve valuable energy during an ongoing infection (Dantzer, Connor, Freund, Johnson, & Kelley, 2008). Proinflammatory cytokines also initiate synthesis of the acute phase reactants, C-reactive protein (CRP) and mannan binding-lectin (MBL) from the liver, and by stimulating opsonisation (i.e., the enhanced sticking of phagocytes to microorganisms), these soluble mediators represent an important aspect of the innate immune response to antigenic invasion.

The HPA axis is involved in a long bidirectional feedback loop with the immune system, such that proinflammatory cytokines arising from an ongoing immune response operate via afferents of the vagus nerve to stimulate central
components of the stress system. Indeed, CRH and ACTH decrease the transcription rate and stability of messenger ribonucleic acid, and in so doing, increase concentrations of inflammatory markers such as IL-6 (Dhabhar, 2003; Miller, 1998). Cortisol on the other hand, by inhibiting proinflammatory transcriptional control pathways, helps prevent the immune system reaching potentially damaging levels (Evans, Hucklebridge, & Clow, 2000). Feedback disturbances between the HPA axis and immune system such as reduced GC mediated signalling, and concomitant disinhibition of proinflammatory activity have been implicated in the pathophysiology of stress related bodily disorders such as fibromyalgia (Heim, Ehlert, & Hellhammer, 2000), burnout (Gunnar & Vasquez, 2001), post traumatic stress disorder (Rohleder, Joksimovic, Wolf, & Kirschbaum, 2004) and chronic fatigue syndrome (van Houdenhove, van den Eede, & Luyten, 2009). Figure 1.3 displays the bidirectional feedback loop between the HPA axis and immune system.
Figure 1.3 Bidirectional feedback loop between the HPA axis and immune system. (adapted from Chrousos, 2000). Stimulation is represented by solid lines and inhibition by dashed lines. **ACTH**, adrenocorticotrophin factor; **CRH**, corticotrophin releasing hormone; **IL**, interleukin; **TNF-α**, tumour necrosis factor-alpha; **PVN**, paraventricular nucleus.

While cells of the innate immune response use primitive non specific recognition systems to detect the foreign invader, cells of the *specific immune response* such as T4 lymphocytes are genetically pre-programmed to express a T-cell receptor (TCR) with a unique specificity. That is, T4 receptors can only match one specific antigen. In addition, T4 cells cannot recognise antigen in their natural (i.e., free) form, only antigen that have been degraded into smaller peptide fragments (called epitopes) and presented in conjunction with major histocompatibility complex II (MHC II), otherwise known as ‘self molecules’, on the surface of phagocytic cells (Miller, 1998). Indeed, cytokines released from the phagocytic cell activate transcription factors within the naive T4 cell that bind to and activate genes responsible for its proliferation (i.e., create clones that share the same unique receptor specificity as the parent cell), and differentiation into effector T4, otherwise known as T-helper (Th) cells. Subpopulations of effector T4 cells can be classified according to the cytokines they produce. Indeed, cytokines of the Th1 phenotype such as IL-12, IL-2, interferon-γ (IFN-γ) and TNF-β promote *cell mediated immunity*. That is, these soluble mediators activate transcription factors that turn on genes responsible for the proliferation and maturation of naive T8 lymphocytes into effector T8 cells, otherwise known as cytotoxic T-lymphocytes (CTL). Perforin, released by effector CTLs, polymerizes to form channels in the membranes of
infected cells, and it is via these channels that proteolytic enzymes (e.g., granzymes) can enter and destroy colonised host cells. Cytokines released from Th1 cells also promote macrophage mediated inflammation (i.e., the release of proinflammatory cytokines from phagocytic cells), as well as the activation of natural killer (NK) cells which are responsible for eradicating infected cells from the body.

Like T-cells, B-lymphocytes are genetically pre-programmed through a series of gene splicing reactions to produce an antibody molecule, otherwise known as an immunoglobulin (i.e., B-cell receptor) with a unique specificity. Once bound, the foreign invader is engulfed by the B-cell, degraded into smaller peptide epitopes, bound to self (i.e., MHC II) molecules and transported to the surface of the B cell for recognition by effector T4 cells. Cytokines released by Th2 cells such as IL-4, IL-5, IL-10 and IL-13 enable the activated B-cell to proliferate (i.e., create clones) and differentiate into effector antibody producing plasma cells. Antibodies, secreted by the plasma offspring of primed B-cells, coat infected host cells, thus tagging them for destruction by NK cells, or lysis by phagocytic cells. Collectively, these immunodefence mechanisms are referred to as humoral immunity. By blocking the maturation of the opposing cell or its receptor function, cytokines released by Th1 and Th2 cells are counter regulatory. Indeed, cytokines of the Th1 phenotype inhibit the actions of Th2 cells, and vice versa (Elenkov, 2004).

The immunosuppressive properties of cortisol have been well documented in the stress literature (Elenkov & Chrousos, 2002; Desantis et al., 2012; Evans, Hucklebridge, & Clow, 2000; Sapolsky, Romero, & Munck, 2000); therefore, a prevailing assumption has been that over activation of the HPA axis and associated immune down regulation is responsible for the effect of chronic stress on negative health outcomes. While this assumption can explain stress induced negative health
characterised by compromised host resistance, it fails to explain studies that have linked enduring periods of stress with the development and progression of diseases characterised by immune disinhibition (Raison & Miller, 2003; Rohleder et al., 2009). Indeed, over activation of the HPA axis (as a result of repeated stress) should, by down regulating immune activity, help to alleviate inflammatory and autoimmune related pathologies. To reconcile these inconsistencies, the GC resistance hypothesis states that immune cells compensate for over activation of the HPA axis by down regulating the expression of receptors responsible for binding GC hormones. Cortisol, therefore, is less able to suppress proinflammatory transcriptional control pathways in cells such as monocytes that have acquired functional resistance to GC mediated signalling (Miller et al., 2008; Rohleder, 2011). As an alternative explanation, the HPA rebound effect states that sustained elevated levels of cortisol rebound to normal (and below normal) over time, as a function of increased negative feedback sensitivity (Gunnar & Vasquez, 2001) or desensitisation of CRH receptors at the level of the anterior pituitary (Raison & Miller, 2003). Indeed, findings from a meta analysis revealed a robust inverse relationship between time since stressor onset and HPA output (Miller et al., 2007).

Insufficient GC mediated signalling characterised by diminished immune system GC sensitivity, or reduced bioavailability of cortisol has been widely observed in the context of chronically stressful life events such as informal caregiving (Miller et al., 2008; Rohleder et al., 2009), PTSD (Rohleder et al., 2004), and in participants experiencing higher levels of general life stress (O’Connor et al., 2009). Insufficient GC mediated signalling and concomitant disinhibition of proinflammatory activity has also been implicated in the aetiologies of stress related bodily disorders such as burnout (Heim et al., 2000), chronic fatigue syndrome (van
Houdenhove, van den Eede, & Luyten, P, 2009) and fibromyalgia (Gunnar & Vasquez, 2000). Researchers have also observed that, rather than being exclusively immunosuppressive, cortisol modulates the immune phenotype by shifting the bias in favour of a Th2 cytokine pattern (Elenkov, 2004). Th2 domination has been linked with inflammatory pathologies such as Graves’ disease (Elenkov & Chrousos, 2002), as well as allergic reactions and asthma (Rook & Lightman, 1997). The GC resistance, HPA rebound and cytokine shift models help reconcile paradoxical findings that have linked chronic stress with increased vulnerability for both infectious and inflammatory related conditions.
1.4 The Caregiver Control Model of Chronic Stress

The study of informal caregivers has become one well established model for assessing the psychophysiological consequences of chronic stress. Indeed, informal caregivers are those who, without payment, provide longstanding care for a significant other (e.g., spouse, parent or child) who, because of disability or illness, is unable to effectively care for themselves (Kuster & Merkle, 2004). Statistics have indicated that there are currently around six million informal caregivers in the United Kingdom, a number expected to increase by 30% over the next 25 years (Edworthy, 2005). To date, the vast majority of research has been conducted with older caregivers, most commonly spousal carers of partners with dementia. The physical, financial and emotional demands of caring for a partner with dementing illness are expected to last between 3-15 years (Vitaliano et al., 2003), and currently over 80% of dementia patients are cared for in the community by close relatives (Bandeira et al., 2007; Haley, 1997). Unsurprisingly, increased psychological morbidity, as evidenced by higher levels of anxiety and hopelessness (Bandeira et al., 2007; Bauer et al., 2000; Vedhara et al., 1999), symptoms of depression (Damjanovic et al., 2007; Da Roza Davis & Cowen, 2001; Kiecolt-Glaser, Dura, Speicher, Trask, & Glaser, 1991; Vitaliano, Persson, Kiyak, Saini, & Echeverria, 2005), and perceived levels of stress (Glaser, Sheridan, Malarkey, MacCallum, & Kiecolt-Glaser, 2000; Kiecolt-Glaser, Marucha, Malarkey, Mercado, & Glaser, 1995) has been widely evidenced in older dementia caregivers, relative to their age matched non caregiving counterparts. Research has indicated that social connectivity and access to social resources are also significantly diminished in spousal care providers (Esterling, Kiecolt-Glaser, & Glaser, 1996; Glaser et al., 2000; Kiecolt-Glaser et al., 1991).
As a proxy measure of the in vivo immune response to antigenic challenge, the vaccination paradigm has been widely applied to model the causal link between caregiver stress and increased vulnerability for infectious disease. To date, the stress of caring for a partner with dementia has been linked with diminished antibody response to influenza (Glaser et al., 1998; Kiecolt-Glaser et al., 1996; Segerstrom et al., 2008; Vedhara et al., 1999), pneumococcal (Glaser, Sheridan, Malarkey, MacCallum, & Kiecolt-Glaser, 2000) and tetanus vaccinations (Li et al., 2007). As the primary defence against mucosal infections, secretory immunoglobulin-A (S-IgA) antibodies are also reduced in older dementia caregivers, relative to age matched normative controls (Gallagher, Phillips, Evans, Der, Hunt, & Carroll, 2008). As well as dysregulated humoral immunity, older dementia caregivers have also displayed perturbations in components of cell mediated immunity such as proliferative response to mitogenic stimulation (Bauer et al., 2000; Castle, Wilkins, Heck, Tanzy, & Fahey, 1995; Damjanovic et al., 2007; Kiecolt-Glaser et al., 1991), virus specific production of Th1 cytokines (Kiecolt-Glaser et al., 1996), mitogen stimulated production of proinflammatory cytokines (Kiecolt-Glaser et al., 1996), Th1 induced secretion of enriched NK cells (Esterling, Kiecolt-Glaser, & Glaser, 1996) and mitogen induced secretion of Th1 cytokines (Bauer et al., 2000). Atypical patterns of cortisol secretion characterised by increased mean output across the day has been implicated as one physiological determinant for dysregulated cellular (Bauer et al., 2000) and humoral immunity (Vedhara et al., 1999) in older dementia caregivers. Indeed, dysregulation of the HPA axis has been widely evidenced in the context of caring for a partner with dementing illness (Brummett et al., 2008; Da Roza Davis & Cowen, 2004; De Vugt et al., 2005; Gallagher-Thompson et al., 2006; Kim & Knight, 2008). Stress induced immune impairments could place older
dementia caregivers at greater risk for deleterious health outcomes above and beyond those associated with a normal age related decline in immune efficacy. Indeed, statistics indicate that mortality is 63% more likely in older dementia caregivers, relative to their age matched non caregiving counterparts (Schulz & Beach, 1999).

Markers of inflammation such as IL-6 and CRP naturally increase with age and provide reliable physiological accompaniments for frailty, and other age related diseases (von Kanel, Dimsdale, & Mills et al., 2006). For example, over production of CRP has been implicated in the aetiologies of atherosclerosis (Pearson et al., 2003; Ridker, Buring, Cook, & Rifai, 2003; Spence, Kennedy, Belch, Hill, & Khan, 2008) and all cause mortality (Hamer et al., 2010), while elevated concentrations of IL-6 have been prospectively associated with increased risk for incident type II diabetes (Hu, Meigs, Li, Rifai, & Manson, 2004) and myocardial infarction (Ridker, Rifai, Stampfer, & Hennekens, 2000). Research has indicated that chronic caregiver stress might exacerbate normal age related elevations in proinflammatory biomarkers. For example, findings from a six year longitudinal study revealed the average rate of IL-6 increase was approximately four times greater in spousal caregivers compared with age matched non caregiving controls (Kiecolt-Glaser et al., 2003). Indeed, elevated concentrations of proinflammatory biomarkers such as IL-6 and CRP have been widely observed in chronically stressed dementia caregivers (Lutgendorf et al., 1999; 2008; von Kanel, Dimsdale, & Ancoli-Israel et al., 2006). Taken together, these findings indicate that caring for a partner with dementia might prematurely age the immune system, and in so doing, accelerate the risk for a host of age related diseases. HPA axis hyperactivity (i.e., sustained hypersecretion of cortisol) and concomitant reduction in immune system GC sensitivity has been implicated as one physiological
pathway that translates chronic caregiver stress into persistent low grade inflammation (Kiecolt-Glaser et al., 2009).

Physiological findings in older dementia caregivers might be interpreted in the context of allostatic load. That is, over activation of the HPA axis might, by suppressing cell mediated immunity or reducing the sensitivity of immune cells to the immunosuppressive effects of cortisol, confer increased vulnerability for infectious, or inflammatory related diseases. Indeed, the stress of caring or a partner with dementing illness has been linked with accelerated rates of morbidity and mortality (Schulz & Beach, 1999; Vitaliano et al., 2003).

However, older caregivers are a unique population, typically contending with pre-existing endocrine and immune impairments associated with their advancing age (i.e., senescence). That is, cumulative exposure to cortisol with age causes hippocampal neuron loss and concomitant reduction in negative feedback sensitivity to GCs, culminating in unrestrained HPA activity forming a feed forward cascade (Sapolsky, Krey, & McEwen, 1986). The neuroendocrine hypothesis of immune senescence states that age related changes in basal HPA activity (i.e., cortisol cascade) are a major determinant for age related immune abnormalities such as poorer Th1 directed immunity and increased levels of inflammatory biomarkers (Bauer, 2005). Stress (i.e., caregiver) induced endocrine and immune impairments might compound pre-existing endocrine and immune senescence to produce physiological alterations more profound than those typically observed in the context of chronic caregiver stress. As such, researchers have started to examine whether the endocrine and immune impairments apparent in older dementia caregivers might extend to relatively young (i.e., parental) caregivers (of children with additional
complex needs), who are not contending with age related alterations in these physiological systems.

Indeed, caregivers of children with additional complex needs face demands that far exceed those of typically developing children such as increased physical pressures, social isolation, concern about the child’s future, financial hardship and having to negotiate a fragmented service system (Baker, Blacher, Crnic, & Edelbrock, 2002; Bank et al., 2006; Fletcher et al., 2012; Seltzer et al., 2009). Statistics have suggested that compared with normative controls (i.e., parents of typically developing children), caregivers of children with additional complex needs such as autism are more likely to experience serious financial problems, as well as difficulties accessing family support services (Parish & Cloud, 2006). In addition, more than 50% of caregivers are forced to reduce or stop working altogether to provide full time care for the impaired child (Kogan et al., 2008). Statistics have also indicated that caregivers of children with autism dedicate more than 43 hours per week to the caregiver role (Jarbrink, Fombonne, & Knapp, 2003), over 30% more than parents of typically developing children (Crowe & Florez, 2006). In the same way older dementia caregivers can expect the stress of the caregiver experience to last between 3-15 years (Vitaliano et al., 2003), relatively young caregivers of children with additional complex needs might anticipate the physical, emotional and financial burden associated with the caregiver role to extend well into the late adulthood of the child (Kuster & Merkle, 2004). Unsurprisingly, caregivers of children with additional complex needs have reported increased psychological morbidity relative to normative controls (Baker et al., 2002; Bella, Garcia, & Spadari-Bratfisch, 2011; Dabrowska & Pisula, 2010; Gallagher & Whiteley, 2012b; Ha, Greenberg, & Seltzer, 2011; Hedov, Annren, & Wikblad, 2000; Ingersoll &
However, fewer studies have assessed the physiological consequences of caregiver stress in younger populations. Research by Vedhara et al (2002) revealed that relatively young caregivers of patients with multiple sclerosis (MS), a severe physical impairment, and age matched non caregiving controls could not be differentiated on components of cell mediated and humoral immunity. From these findings, it might be concluded that the effect of chronic caregiver stress on immune dysfunction is exclusive to older caregivers, and as such, requires an already impaired (i.e., aged) immune system to manifest. Interestingly, however, post hoc analyses revealed the MS caregivers, while significantly more stressed than controls, were less distressed by the caregiver experience when compared against a group of older dementia caregivers. These findings raise an important question, whether preservation of immune efficacy in relatively young MS caregivers might be attributed to the absence of immune senescence, or relative lack of psychological distress associated with the caregiver experience. One model that has shed light on these competing explanations concerns the study of relatively young (i.e., parental) caregivers of children with additional complex needs such as autism, a condition characterised by the same kinds of distressing cognitive and behavioural impairments that affect dementia patients (Floyd & Gallagher, 1997; Hastings, Daley, Burns, & Beck, 2006). Indeed, relative to caregivers of patients with physical impairments such as MS, psychological distress is considerably greater and the caregiver experience much more intense in caregivers of patients with distressing cognitive and behavioural impairments such as dementia (Pinquart & Sorensen, 2003) and autism (Bouma & Schweitzer, 1990; Ergh, Rapport, Coleman, & Hanks, 2002). Researchers...
recently demonstrated that, unlike caregivers of patients with physical impairments such as MS (Vedhara et al., 2002), antibody response to influenza vaccination was diminished in caregivers of children experiencing distressing cognitive and behavioural impairments such as autism (Gallagher et al., 2009). Dysregulated immunity, manifested by increased markers of inflammation such as CRP, has also been observed in caregivers of children experiencing the severe cognitive and behavioural sequelae associated with glioblastoma multiforme, the most aggressive form of primary brain cancer (Miller et al., 2008; Rohleider et al., 2009). Taken together, these findings indicate that it is not the age of the caregiver that matters (i.e., senescence), rather that specific characteristics of the care recipient (i.e., nature of the impairment) and associated variations in the intensity of the caregiver experience dictate whether chronic caregiver stress becomes an issue for disease relevant physiological processes such as the immune response.

To extend work in this area, study two compared relatively young caregivers of children experiencing severe cognitive (i.e., autism) and behavioural (i.e., ADHD) impairments with a normative control group on a range of psychophysiological outcomes. Research has suggested the negative impact of caring for child with additional complex needs on immune function might be mediated by problem behaviours of the care recipient (Gallagher et al., 2009). Few studies, however, have actually assessed the psychosocial antecedents that might account for immune dysfunction in younger care providers (Vitaliano et al., 2003). To add to the relatively small research base in this area, study two also considered whether social support, typically diminished in relatively young caregivers (Edworthy, 2005; Fletcher, et al., 2012; Gallagher et al., 2010; Lach et al., 2009) and often associated with immune alterations (Kiecolt-Glaser et al., 2009; Lutgendorf et al., 2000;
Moynihan et al., 2004), might underlie the effect of chronic caregiver stress on dysregulated immunity. Indeed, reduced social support has been implicated as one psychosocial determinant for perturbations in other disease relevant physiological processes such as systolic blood pressure (Gallagher & Whiteley, 2012a) and basal HPA activity (Kim & Knight, 2008) in relatively young caregivers.
1.5 Social Support and Written Emotional Disclosure

The stress buffering hypothesis states that socially supported individuals (i.e., those with family and friends that provide emotional and material resources) appraise stressful situations to be less threatening, and by consequence, are protected against maladaptive changes in disease relevant physiological processes (Cohen & Ashby Wills, 1985). For example, in the context of a chronically stressful life event such as breast cancer, greater social support can mitigate psychological distress and buffer against endocrine (Turner-Cobb, Sephton, Koopman, Blake Mortimer, & Spiegel, 2000) and immune alterations (Costanzo et al., 2005). Indeed, the stress ameliorating effect of social support has been demonstrated in a large scale meta analysis (Sörensen et al., 2002), and more recent review paper (Kiecolt-Glaser et al., 2009). Converging with these findings, research has also revealed how dyadic support, i.e., the presence of a partner, loved one, or even a stranger, can attenuate cortisol (Ditzen et al., 2007; Kirschbaum et al., 1995) and cardiovascular reactivity (Lepore, Allen, & Evan, 1993; Phillips, Carroll, Hunt, & Der, 2006; Thorsteinsson, & James, 1999) to acutely stressful paradigms in laboratory settings.

Informal caregivers, however, face an extremely intense experience that seriously limits time available for social connectivity. For example, caregivers have reported fewer available social resources (Boyd, 2002; Esterling et al., 1996; Gallagher et al., 2010; Glaser et al., 2000; Kiecolt-Glaser et al., 1991), loss of friends (Fletcher, et al., 2012; Lach et al., 2009) and painful social isolation (Edworthy, 2005; Marsh, Kersel, Havill, & Sleigh, 1998; Wiles, 2003; Yantzi et al., 2006), relative to non caregiving controls. Caregivers of children with additional complex needs also find it difficult to regularly attend community support groups, despite good provision (Bank et al., 2006; Chien et al., 2011; Yantzi et al., 2006). Despite
these difficulties, socially supported caregivers appear to be protected against the harmful effects associated with the caregiver experience. For example, lower levels of psychological distress (Kiecolt-Glaser et al., 1987) and more adaptive immune function, as indexed by greater proliferative response to mitogenic stimulation (Baron, Cutrona, Hicklin, Russell, & Lubaroff, 1990; Kiecolt-Glaser et al., 1991) and Th1 induced secretion of NK cells in vitro (Esterling et al., 1994, 1996), has been observed in older dementia caregivers who reported greater social support. Adaptational outcomes such as reduced psychological morbidity and fewer incidences of negative health have also been reported in spousal caregivers who regularly attended caregiver support groups (Chien et al., 2011). The stress ameliorating effect of social support also seems to extend to relatively young caregivers. Indeed, socially supported caregivers of children with additional complex needs have reported lower levels of depression (Bozo et al., 2010; Ekas, Lickenbrock, & Whitman, 2010; Ergh, et al., 2002; Weiss, 2002; White & Hastings, 2004), loneliness (Kayfitz et al., 2010) and negative affect (Bromley, Hare, Davison, & Emerson, 2004; Ha et al., 2011; Ludlow, Skelly, & Rohleder, 2012; Pottie, Cohen, & Ingram, 2009; Pozo et al., in press), as well as fewer incidences of adverse physical health (Lin et al., 2009; Raina et al., 2005; Sawyer et al., 2010). Diminished psychological distress has also been reported in relatively young caregivers who received greater support from partners (Herman & Thompson, 1995), extended family members (Hastings, Thomas, & Delwiche, 2002) and other informal sources such as friends (Hastings & Johnson 2001; Smith, Oliver, & Innocenti, 2001). Recently, larger social networks predicted fewer depressive symptoms and more positive emotions across an 18 month period in relatively young caregivers of children with autism (Smith et al., 2011). However, fewer studies have assessed
whether social support can play a protective role with respect to perturbations in disease relevant physiological processes in relatively young caregivers. In a recent study, Gallagher and Whiteley (2012a) observed how socially supported caregivers of children with additional complex needs such as autism were protected against higher daytime systolic blood pressure, one well known physiological signature for cardiovascular disease risk.

To extend this relatively small research base, study three examined whether greater social support might ameliorate psychological distress, and in so doing, protect relatively young caregivers of children with autism/ADHD against alterations in basal HPA activity and the immune response.

It has been widely evidenced that quality of life for the care recipient is contingent on the health and happiness of the care provider (Addington, Coldham, Jones, Ko, & Addington, 2003; Burgess & Gutstein, 2007; Schor, 2003). Indeed, the harmful effects of caregiver stress can often impede caregivers’ ability to provide the consistency and quality of care needed by a child with additional complex needs (Beach, Schulz, Williamson, Miller, Weiner, & Lance, 2005; Groeneveld, Vermeer, van Ijzendoorn, & Linting, 2012; Hart & Kelley, 2006; Mockus-Parks & Novielli, 2000; Reinhard et al., 2008). As such, it becomes important to develop interventions that ameliorate the harmful effects of caregiver stress, with a view to improving care recipients’ quality of life. Lower levels of psychological distress (Gallagher-Thompson et al., 2000; Losada-Baltar et al., 2004; Ostwald, Hepburn, Caron, Burns, & Mantell, 1999; Wysocki et al., 2000) and enhanced immune efficacy, as indexed by greater antibody response to influenza vaccination (Vedhara et al., 2003), have been observed in chronically stressed caregivers who participated in cognitive behavioural therapy and psychoeducational skills training interventions. Indeed, meta
analysis has revealed that, in the context of chronic caregiver stress, psychophysiological outcomes are amenable to improvements through participation in psychotherapeutic (e.g., cognitive behavioural therapy) and education based interventions (Garcés et al., 2010; Sörensen et al., 2002). Other studies have demonstrated that formal respite care (i.e., periodically relieving caregivers of their responsibilities) might also be effective for alleviating caregiver induced stress. For example, lower anxiety and depression scores, as well as fewer complaints of negative health have been reported in chronically stressed caregivers receiving formal aid from respite services (Zarit et al., 1999). Indeed, a recent meta analysis concluded that formal respite care is one of the most effective interventions for coping with chronic caregiver stress (Garcés et al., 2010). However, these interventions are often expensive, time consuming and usually based outside the home, and as such, might not be well suited for informal caregivers who experience severe financial hardship (Kogan et al., 2008; Parish & Cloud, 2006), as well as logistical/practical challenges (e.g., difficulties arranging alternate and reliable supervision) that make it almost impossible to get out of the house (Bank et al., 2006; Edworthy, 2005; Yantzi et al., 2006).

Written emotional disclosure (WED) on the other hand is a simple, time and cost effective intervention that requires participants to write expressively about any stressful/traumatic experience for 15-20 minutes on three-four consecutive days (Pennebaker et al., 1988). Despite its apparent simplicity, disclosing stressful/traumatic events in writing has been linked with positive changes in a broad catalogue of health outcomes. For example, reduced levels of psychological distress (Hemenover, 2003; Lutgendorf et al., 1999) and positive physiological adjustments such as reduced cortisol reactivity to acutely stressful lab tasks (Sloan,
Marx, & Epstein, 2005; Smyth, Hockemeyer, & Tulloch, 2008) have been observed in participants who transformed their thoughts and feelings about stressful/traumatic life events into language. WED has also been linked with positive changes in immune function such as greater antibody response to vaccination (Petrie, Booth, Pennebaker, Davison, & Thomas, 1995; Stetler, Chen, & Miller, 2006) and higher T cell percentages (Booth, Petrie, & Pennebaker, 1997; Pennebaker et al., 1988; Petrie, Fontanilla, Thomas, Booth, & Pennebaker, 2004), as well as improvements in self reported somatic health status (Hemenover, 2003; Henry et al., 2010; Park & Blumberg, 2002). Indeed, the beneficial effects of WED on health related outcomes have been demonstrated in healthy (Fratarolli, 2006; Smyth, 1998) and medical populations (Averill, Kasarskis, & Segerstrom, 2013; Baikie & Wilhelm, 2005; Frisina, Borod, & Lepore, 2004; Wetherell et al., 2005), and in the context of chronically stressful life events such as PTSD (Koopman et al., 2006; Sloan, Marx, & Epstein, 2005; Smyth, 1998), bereavement (Kovac & Range, 2000) and breast cancer (Rosenberg et al., 2002).

The WED paradigm, usually applied in a controlled lab setting, has been modified to run in participants’ homes, and as such, might be especially well suited for informal caregivers who, despite their willingness, find it extremely difficult to participate in lab based studies (Wetherell et al., 2005). Previous research has indicated that implementing an at-home WED intervention in the context of chronically stressful life events is indeed, feasible. For example, in a recent study that assessed the feasibility and efficacy of expressive writing in chronically stressed cancer patients, adherence with the writing protocol was high. That is, 94% of participants reported no difficulties finding a quiet place in the home to complete the writing task, and 91% reported completion of the task with little or no
interruptions (Henry et al., 2010). Equally high levels of adherence with the writing protocol were recently demonstrated in the context of chronic caregiver stress (Ashley et al., 2011).

In its traditional format (i.e., where participants are free to write about any stressful/traumatic event), WED has yielded little in the way of health benefits for informal caregivers (Schwartz & Drotar, 2004). These null effects have also been replicated in caregivers directed to focus their writing exclusively on the stress associated with the caregiver experience (Barton & Jackson, 2008; Mackenzie et al., 2007). However, in a modification to the traditional WED paradigm, Ashley et al (2011) recently reported on psychological improvements such as reduced anxiety and depression scores in caregivers lower in alexithymia (and therefore, better able to express their feelings linguistically), who wrote for 20 minutes on three consecutive days about positive life experiences. Indeed, the beneficial effects of positive written disclosure on psychophysiological parameters have been widely evidenced (Burton & King, 2004, 2008; King & Miner, 2000; Wing, Schutte, & Byrne, 2006). Neatly dovetailing with these findings, diminished levels of psychological distress (Cheshire, Barlow, & Powell, 2012; Kayfitz et al., 2010; Kim, Schulz, & Carver, 2007; McCausland & Pakenham, 2003) and steeper diurnal cortisol slopes, which is indicative of more adaptive HPA function (Moskowitz & Epel, 2006), have also been observed in caregivers who reported finding more positive consequences (i.e., benefits) amidst the stress of the caregiver experience. Indeed, strengthened intimate relationships, changes in priorities of life goals, greater sensitivity to family issues and overall appreciation for life/loved ones are only a few examples of the kinds of benefits reported by informal caregivers in the midst of chronic caregiver stress (Markoulakis, Fletcher, & Bryden, 2012; Samios
et al., 2009). Interestingly, writing expressively about the benefits associated with chronically stressful life events has been found to positively influence health related outcomes. For example, lower depression scores and fewer incidences of negative health were reported in chronically stressed breast cancer patients directed to write for 20 minutes on three consecutive days about the benefits associated with their disease experience (Henry et al., 2010; Stanton et al., 2002). In view of these findings, it is perhaps surprising that only one study to date has applied a positive written disclosure intervention in chronically stressed caregivers (Ashley et al., 2011).

To address the paucity of research in this area, study four assessed the feasibility and efficacy of an at-home written benefit finding intervention, including the assessment of psychophysiological indices, in caregivers of children with autism/ADHD.
1.6 Summary and Conclusions

Several issues have been addressed in this literature review; firstly, few studies have examined the physiological processes that might underlie greater incidences of common health problems in young otherwise healthy individuals experiencing higher perceived levels of stress. Consequently, study one assessed whether perceived stress related disparities in self reported incidences of common health complaints might be mediated by differential patterns of cortisol secretion.

With regard the caregiver component of the programme, alterations in endocrine and immune parameters, indicative of greater vulnerability for ill health, have been widely evidenced in older dementia caregivers (Vedhara et al., 1999; Bauer et al., 2000). However, fewer studies have examined whether the negative impact of chronic caregiver stress on endocrine and immune function is exclusive to older dementia caregivers who, because of senescence, are already contending with pre-existing, age associated alterations in these physiological systems. Immune dysfunction, manifested by poorer antibody response to vaccination (Gallagher et al., 2009) and elevated markers of inflammation (Miller et al., 2008; Rohleder et al., 2009), has been observed in younger caregivers of children with additional complex needs such as autism, a condition characterised by the same kinds of cognitive and behavioural impairments that affect dementia patients, but not in younger caregivers of patients with physical impairments such as MS (Vedhara et al., 2002). Taken together, these findings suggest the immune dysfunction apparent in the context of chronic caregiver stress is not exclusive to older dementia caregivers (i.e., does not require an already aged immune response to manifest), but might extend to younger caregivers of children with additional complex needs such as autism, a condition that shares many clinical manifestations of dementia, and is therefore comparable in
terms of caregiver burden. To extend research in this area, study two examined the psychophysiological consequences of caring for a child with autism/ADHD, conditions that are characterised by the same kinds of distressing cognitive and behavioural impairments experienced by older dementia patients (Baron-Cohen, 2004; Johnson & Reader, 2002). Social support, typically lower in relatively young caregivers (Fletcher, et al., 2012; Gallagher & Whiteley, 2012b) and often linked with dysregulated immunity (Kiecolt-Glaser et al., 2009; Lutgendorf et al., 2000), was also examined as one psychosocial pathway that might link chronic caregiver stress with immune dysfunction. Indeed, few studies have examined the psychosocial antecedents that might underlie the negative impact of chronic caregiver stress (in younger populations) on disease relevant physiological processes such as the immune response (Gallagher et al., 2009; Vitaliano et al., 2003).

The stress buffering hypothesis states that socially supported individuals appraise stressful events to be less threatening, and as such, are protected against perturbations in physiological processes relevant for health and well being (Cohen & Ashby-Wills, 1985). Indeed, reduced psychological morbidity has been observed in relatively young caregivers of children with autism (Bromley et al., 2004; Pottie et al., 2009; Sawyer et al., 2010; Smith et al., 2011; Weiss, 2002) and other additional complex needs (Bozo et al., 2010; Ha et al., 2010; Lin et al., 2009; White & Hastings, 2004) who reported greater social support. However, fewer studies have assessed whether social support can protect relatively young caregivers against maladaptive changes in disease relevant physiological mechanisms such as the HPA axis and immune response. In a recent study, Gallagher and Whiteley (2012a) demonstrated an inverse relationship between social support and daytime systolic
blood pressure in relatively young caregivers of children with autism and other additional complex needs.

**Study three** aimed to add to this relatively small research base by assessing whether greater social support might ameliorate psychological distress, and in so doing, protect relatively young caregivers of children with autism/ADHD against alterations in basal HPA activity and the immune response.

Quality of life for the care recipient is inextricably linked with the psychophysiological well being of the care provider (Addington et al., 2003; Burgess et al., 2007). Indeed, decrements in caregivers’ emotional and physical health can seriously undermine the quality of care they provide (Beach et al., 2005). As such, it becomes important to develop interventions that alleviate caregiver related stress, with a view to improving care recipients’ quality of life. Positive psychophysiological adjustments such as lower depression scores and enhanced immune response to influenza vaccination, have been observed in caregivers who participated in psychotherapeutic (i.e., cognitive behavioural therapy) and psychoeducational skills training interventions (Gallagher-Thompson et al., 2000, 2001; Losada-Baltar et al., 2004; Sörensen et al., 2002; Vedhara et al., 2003). Adaptational outcomes such as lower levels of depression and anxiety have also been reported in caregivers receiving formal aid from respite services (Zarit et al., 1999). However, financial (Donelan et al., 2002; Kogan et al., 2008) and logistical/practical challenges such as difficulties arranging alternate and reliable supervision (Bank et al., 2006; Edworthy, 2005) often prevent caregivers’ access to these varied and potentially effective interventions.

**WED** on the other hand is a simple, cheap and time effective intervention that has been adapted to run in participants’ homes, and at any time of day (Wetherell et
al., 2006), and as such, might be especially well suited for informal caregivers. In terms of its efficacy, WED has been linked with adaptive changes in a broad catalogue of health outcomes in healthy (Booth et al., 1997; Petrie et al., 1995; Sloan et al., 2005) and medical populations (Smyth et al., 1999; Wetherell et al., 2005), and in the context of chronically stressful life events such as bereavement (Kovac & Range, 2000) and breast cancer (Low et al., 2006; Rosenberg et al., 2002), but not informal caregiving (Barton & Jackson, 2008; Mackenzie et al., 2007; Schwartz & Drotar, 2004). However, in a modification to the traditional WED paradigm, reduced anxiety and depression scores were recently reported in caregivers lower in alexithymia, who focussed their writing on positive life experiences (Ashley et al., 2011). Converging with these findings, diminished levels of psychological distress and steeper diurnal cortisol slopes (Moskowitz & Epel, 2006), which is indicative of more adaptive HPA activity, has been observed in caregivers who reported finding more positive consequences (i.e., benefits) amidst the stress of the caregiver experience. Interestingly, writing about the benefits associated with stressful life events has been linked with adaptational outcomes such as reduced anxiety and depression scores, as well as fewer reported incidences of ill health (Henry et al., 2010; Stanton et al., 2002).

To date, only one study has assessed the efficacy of positive written disclosure in the context of chronic caregiver stress (Ashley et al., 2011). To address the paucity of research in this area, study four used a randomised control trial to assess the feasibility and efficacy of an at-home written benefit finding intervention for coping with the stress of caring for a child with autism/ADHD.
2. Programme Methods

2.0 Chapter Overview

General information pertaining to measures, procedures and protocols applied throughout the programme is discussed here. However, information specific to individual studies will be detailed within subsequent chapters.

2.1 Psychosocial Assessment

Psychological distress was quantified using the 10 item Perceived Stress Scale (PSS) (Cohen, Kamarck, & Mermelstein, 1983). The PSS, though not a diagnostic instrument, measures to what extent participants perceived their lives to be overwhelming, uncontrollable and unpredictable over the previous month. Scale responses range from 0 (never) to 4 (very often). A total PSS score is calculated, first by reversing scores on the four positive items (4, 5, 7, & 8), and then by summing across all items. A total PSS score can range from 0-40 with higher scores indicating greater perceived levels of stress.

Anxiety and depression were assessed using the Hospital Anxiety and Depression Scale (HADS) (Zigmond & Snaith, 1983). The HADS is composed of 14 items, seven reflecting anxiety and seven reflecting depression, and is scored along a four point scale ranging from 0 (never) to 3 (considerable). Items for the two subscales are summed to yield a total score, with higher scores (range 0-21) indicating higher levels of anxiety and depression. Clinical guidelines suggest that anxiety and depression scores (a) between 0-7 are normal, (b) 8-10 indicate borderline mood disorder and (c) > 11 are indicative of probable mood disorder (Snaith, 2003).
Incidences of 54 common health problems were quantified using the Pennebaker Inventory of Limbic Languidness (PILL) (Pennebaker, 1982). Scale responses range from 1 (never/almost never) to 5 (more than once per week). Items experienced by the respondent more than once per month are summed to formulate an index of total problem frequency.

Social support was assessed using the Interpersonal Support Evaluation Checklist (ISEL) (Cohen & Hoberman, 1983). The scale comprises 40 statements that measure participants’ availability of social support across four subscales: appraisal support (availability of confidants to discuss one’s problems), belonging support (availability to interact with others), tangible support (availability of material aid), and self esteem based support (availability of a favourable comparison when comparing oneself with others). Scale responses range from 0 (definitely false) to 3 (definitely true). Items are counterbalanced, i.e., half are positive statements regarding social support and half are negative, therefore reverse scoring is applied as necessary. Total scores for each subscale are calculated by summing across items, with higher scores indicating greater social support.

2.2 Salivary Cortisol

2.2.1 Basic Issues

Released from the adrenal cortex, approximately 90% of cortisol is bound to blood borne carriers such as corticosteroid-binding-globulin (CBG) and erythrocytes, while only a small percentage (5-10%) circulates unbound, or free (Kirschbaum & Hellhammer, 1989). Only free cortisol, which is measurable in saliva, acts on target tissues and receptors to elicit physiological changes (Ekins, 1990; Kirschbaum & Hellhammer, 2000). Therefore, salivary cortisol provides one index of the
biologically active fraction of the hormone. Saliva, unlike blood, can be collected noninvasively and without the supervision of trained medical personnel, and as such, has become the preferred method of physiological assessment in psychophysiological research (Kudielka, Gierens, Hellhammer, Wust, & Schlotz, 2012). For all studies reported here, cortisol was sampled using the Salivette (Sarstedt, Ltd); that is, participants were asked to chew sterile cotton swabs for one-two minutes and deposit saturated swabs into plastic collection tubes.

2.2.2 State Influences

On any single day, diurnal cortisol measurement can be influenced by state variables. Indeed, findings have revealed a steeper CAR in early risers (Edwards, Evans, Hucklebridge, & Clow, 2001; Federenko et al., 2004; Kudielka & Kirschbaum, 2003; Stalder, Hucklebridge, Evans & Clow, 2009), and in participants exposed to 250 lux (Thorn, Hucklebridge, Esgate, Evans, & Clow, 2004) and 800 lux light in the morning (Scheer & Buijs, 1999). Findings have also revealed that the CAR, while apparent in conditions of complete darkness after waking, is enhanced if exposed to simulated dawn over the same time period (Thorn et al., 2004). Both an inverse (Williams, Magid, & Steptoe, 2005) and positive relationship (Griefahn & Robens, 2008) between CAR magnitude and sleep duration have been observed, as has diminished waking levels of cortisol in participants who reported poorer sleep quality (Lasicewitz, Hendrickx, Talbot, & Dye, 2008). More recently, a steeper CAR was observed in participants who reported greater levels of arousal (Thorn, Hucklebridge, Evans, & Clow, 2009).

To increase the reliability of diurnal cortisol measurement, researchers have recommended that participants collect between four-six saliva samples (Saxbe, 2008).
on two (or more) consecutive weekdays (Hellhammer et al., 2007; Stalder, Evans, Hucklebridge, & Clow, 2010). To capture important measures that summarise aspects of the diurnal cortisol pattern, all participants in the current programme of work were instructed to collect salivary cortisol at waking, 30 minutes post waking, 1200h and 2200h on two (studies two, three and four), or three (study one) consecutive weekdays.

2.2.3 Protocol Adherence

Non adherence with the saliva collection protocol can invalidate the reliability of resultant cortisol data. Indeed, the increase in cortisol between waking and 30 minutes post waking can be as great as 100%, and therefore, even small timing errors might have ramifications for the values obtained (Clow, Thorn, Evans, & Hucklebridge, 2004). The negative impact of inaccurately timed morning samples in relation to waking, or to each other, on estimates of CAR magnitude has been widely evidenced. Indeed, delays of more than 10 minutes between waking and collection of the waking cortisol sample (i.e., where cortisol has already started to rise), or where collection of the waking and 30 minute post waking samples deviated by more than 10 minutes from the requested 30 minute time interval (i.e., that fails to capture the cortisol apex) have been linked with reduced CAR magnitude (Broderick, Arnold, Kudielka, & Kirschbaum, 2004; Dockray, Bhattacharyya, Molloy, & Steptoe, 2008; Griefahn & Robens, 2011; Kupper et al., 2005; Okun et al., 2010; Wright & Steptoe, 2005). Where research has linked disruptions in the CAR with pathophysiological conditions such as subclinical atherosclerosis (Eller et al., 2005) and clinically verified coronary artery disease (Bhattacharyya et al., 2008), non
adherence with the saliva collection protocol might have implications for interpretation of the CAR, and its clinical relevance.

Some researchers have turned to electronic monitoring devices such as MEMS track caps (that record the date and time of each sample) and wrist actigraphy (that monitor periods of movement/non movement) as a more objective check on timing compliance (Kudielka, Hawkley, Adam, & Cacioppo, 2007; Smyth, Clow, Thorn, Hucklebridge, & Evans, in press). Researchers have recommended that objective assessments of sampling accuracy are used only when the scientific gains outweigh the substantial financial costs (Adam & Kumari, 2009). However, data from recent studies indicated that subjective, self report measures (i.e., paper diaries) of timing compliance are not only preferred by participants (Kraemer et al., 2006), but perform equally well when compared to more objective, electronic devices for evaluating adherence with the saliva collection protocol (Seltzer et al., 2010; Stalder et al., 2009; Okun et al., 2010; Wolf et al., 2008). In addition, it was recently demonstrated that electronic monitoring devices (i.e., MEMS track caps) often produce missing data points, and as a result, might lead to erroneous interpretations of diurnal cortisol measurement (Ailinger, Black, & Lima-Garcia, 2008; Hall et al., 2011).

For the current programme of work, all participants were provided paper adherence diaries and asked to report waking and collection times as accurately as possible on all sampling days. In accordance with previous work, a compliance window of more than 10 minutes between waking and collection of the waking cortisol sample was applied as criteria for the exclusion of erroneous cortisol data (Wright & Steptoe, 2005). Cortisol data were also excluded if self reported collection times for the waking and 30 minute post waking samples deviated by more than 10
minutes from the requested 30 minute time interval (Kudielka et al., 2007). It should be acknowledged that other procedures controlling for suspected protocol non-adherence might have been applied such as excluding data for participants who displayed no post waking cortisol rise (O’Connor et al., 2009; Thorn, Hucklebridge, Evans, & Clow, 2006).

Adam and Kumari (2009) indicated that adherence with the saliva collection protocol might be optimised if participants are provided: (a) clear sampling instructions, (b) a precise definition of waking, (c) training to accurately collect saliva, and (d) a simple way to return saliva samples. For the current programme of work, all participants were provided detailed written instructions that emphasised the time sensitive nature of the hormone, and were trained to accurately collect saliva using the Salivette (Sarstedt, Ltd). In addition, for 45 minutes prior to the collection of any sample, participants were instructed to abstain from behaviours known to affect the measurement of cortisol in saliva. These included: (a) consumption of food, caffeinated and/or alcoholic beverages (b) use of nicotine, (c) brushing teeth, including use of mouthwash and/or antacids, and (d) exercise (Kirschbaum & Hellhammer, 1992; Kudielka, Hellhammer, & Wust, 2009). Waking was defined as: ‘when your eyes open and you are ready to get up’ (Adam & Kumari, 2009; Cohen et al., 2006). In all cases, participants were asked to remain supine for collection of the waking cortisol sample, but they were free to collect the 30 minute post waking sample either supine or standing, as research has revealed no posture related disparities (i.e., supine vs. standing) in CAR magnitude (Hucklebridge, Mellins, Evans, & Clow, 2002). Participants were provided prepaid addressed envelopes for returning samples and adherence diaries to the research team by post. Salivary cortisol remains stable at room temperature for five days (Clements & Parker, 1998),
and even up to several weeks later (Kirschbaum & Hellhammer, 1989; Kudielka et al., 2012), and as such, degradation during collection and postage periods was not anticipated.

2.2.4 Biochemical Assays

In accordance with recent guidelines, all returned samples were immediately stored frozen at -20°C until biochemical assay (Kudielka et al., 2012). For study one, after defrosting, salivettes were centrifuged for 10 minutes, 400 x g at 20°C and tested in-house using a time resolved competitive luminescence immunoassay, IBL Hamburg, Germany. For studies two-four, saliva samples were tested in-house using an enzyme-linked immunosorbant assay (ELISA), Salimetrics Ltd, Suffolk, England. No differences were observed between techniques in terms of accuracy such that inter and intra assay coefficients of variation ranged between 3.9%-6.0% and 7.1%-4.6% respectively. To minimise inter-assay variation, samples from each participant were tested on the same immunoassay plate.

2.2.5 Treatment of Cortisol Data

To normalise distributions, raw cortisol values were log_{10} transformed and data for each sampling day used to derive three markers of basal HPA activity. First, the CAR was calculated. Area under the curve (AUC) captures total cortisol volume across the post waking period and has been widely applied as a measure of the cortisol response to waking (Fries et al., 2009). However, calling for a more consistent measure of the CAR, Clow Hucklebridge, Stalder, Evans, & Thorn (2010) recently recommended that measures of overall cortisol volume across the post waking period (i.e., AUC) be substituted with a single measure of reactivity from
waking. In addition, findings from a recent meta analysis suggested the change in cortisol from waking values is, compared with measures of overall cortisol volume (i.e., AUC), a more appropriate measure for assessing HPA activity post waking (Chida & Steptoe, 2009). For all studies of the thesis, the CAR was calculated as the difference between cortisol values at waking and 30 minutes post waking (Kudielka et al., 2012; Kunz-Ebrecht et al., 2004; Steptoe, Kunz-Ebrecht, Brydon, & Wardle, 2004). Second, all four cortisol values were summed to calculate mean cortisol output across the day (Brant, Wetherell, Lightman, Crown, & Vedhara, 2009; Grossi, Perski, Lundberg, & Soares, 2001; O’Connor et al., 2009). Finally, the slope of diurnal change was calculated to assess how well participants fitted the typical descending cortisol pattern. This was achieved by estimating a linear regression line for each participant that predicted cortisol from time since waking (Smyth et al., 1997). Steeper cortisol slopes indicate a greater rate of diurnal decline and are represented by smaller $\beta$ values (larger negative values). Higher $\beta$ values (as they approach, or cross zero) indicate flatter diurnal slopes and provide one marker of dysregulated HPA activity. The CAR represents phasic psychophysiological processes specific to the transition from sleep to wakefulness, and therefore, is independent from diurnal variations in HPA activity (Edwards, Clow, Evans & Hucklebridge, 2001; Fries et al., 2009; Wilhelm, Born, Kudielka, Schlotz & Wust, 2007). To avoid any effect of the CAR on estimates of the diurnal cortisol slope, cortisol values at 30 minutes post waking were excluded from slope calculations (Brant et al., 2009; Cohen et al., 2006; Matthews et al., 2006).
3. Study One

3.0 Chapter Overview

Research involving young otherwise healthy individuals has linked higher perceived levels of stress with increased susceptibility and severity for common health problems such as upper respiratory illness (Cohen et al., 1991, 1993, 1999). However, fewer studies have examined the physiological antecedents that might underlie these effects. Consequently, study one assessed whether perceived stress related disparities in subjective reports of common health problems might be mediated by differential patterns of cortisol secretion.

Participants who reported higher perceived levels of stress displayed flatter diurnal cortisol slopes, largely accounted for by hypersecretion of cortisol in the evening. Mean levels of cortisol across the day were also elevated in the higher stress group, as were reported incidences of common health problems. Findings reported here converge with other studies that have observed atypical patterns of cortisol secretion (Abercrombie et al., 2004; O’Connor et al., 2009) and greater perceptual representations of common health problems (Cohen et al., 1999; Hellhammer et al., 2004; Sjögren et al., 2006) in young otherwise healthy individuals experiencing higher perceived levels of stress. Perceived stress related disparities in common health problems were appreciably reduced when mean diurnal cortisol output was statistically controlled. These findings indicate that dysregulated HPA activity, widely implicated in the aetiologies of severe pathologic conditions such as subclinical atherosclerosis (Hamer et al., 2010; Matthews et al., 2006; Seldenrijk et al., 2012) and coronary artery disease (Bhattacharyya et al., 2008), partially mediated the effect of higher perceived levels of stress on greater incidences of the kinds of common health complaints that typically affect young otherwise healthy individuals.
3.1 Methods

3.1.1 Participants

A sample of 47 female undergraduate students responded to an e-mail inviting participation in a study investigating: ‘perceived levels of stress, stress hormones and common health problems’. Participants were recruited according to strict exclusion criteria which included: (a) past or present disorders of the endocrine system, (b) irregular medical history, (c) taking prescription medication excluding oral contraceptives, (d) excessive alcohol intake, defined as more than two alcoholic beverages per day, (e) excessive caffeine intake, defined as more than 10 cups of tea and/or coffee per day, (f) smoker and (g) working night shift. Participants who satisfied these criteria were invited to provide informed consent following an ethically approved protocol by the Department of Psychology Ethics Committee. All participants were recompensed £10.00. Data for one participant was excluded based on cortisol values greater than 75 nmol/L (Kunz Ebrecht et al., 2004). Four participants who failed to return any saliva samples and seven participants who failed to complete the primary health measure were also excluded. Analyses were conducted on a final sample of 35 participants.

3.1.2 Potential Confounds

To determine whether demographic, behavioural or biomedical factors might be acting as confounds, data were collected on participants’ age, weight, body mass index (BMI), time of waking, use of oral contraceptives and phase of the menstrual cycle.
3.1.3 Psychological Outcomes

Perceived levels of stress were quantified using the 10 item PSS (see Chapter Two for full details of the PSS). In accordance with previous research, participants were dichotomised into higher (range: 16-26) and lower (range: 3-14) stress groups based on responses to the PSS (Burns, Drayson, Ring, & Carroll, 2002; Epel et al., 2004; O’Connor et al., 2009). To create distinctly higher and lower groups, three participants who scored at the median were excluded. Therefore, statistical analyses were conducted on a final sample of 32 participants. Self reported incidences of common health problems were assessed using the PILL (see Chapter Two for full details of the PILL). All measures achieved excellent psychometrics in the sample (all alpha’s > 0.87).

3.1.4 Physiological Outcomes

To capture important parameters of the diurnal cortisol pattern, participants collected saliva samples at waking, 30 minutes post waking, 1200h and 2200h on three consecutive weekdays (see Chapter Two for full details of the saliva collection protocol and checks on adherence). Repeated measures analysis of variance (ANOVA) was used to explore possible within person differences in cortisol data per individual sample point. Findings revealed a significant main effect of time ($F(2.35, 70.36) = 137.02, p < 0.01, \eta_p^2 = 0.82$), reflecting normal diurnal variation of cortisol secretion. However, no between day differences ($F(2, 60) = 0.45, p = 0.64, \eta_p^2 = 0.15$) or day x time interaction effect ($F(4.82, 144.52) = 1.27, p = 0.28, \eta_p^2 = 0.04$) was observed. In addition, no between day differences emerged with respect to derived HPA indices, CAR magnitude ($F(2, 60) = 0.58, p = 0.57, \eta_p^2 = 0.02$) or diurnal cortisol slope ($F(2, 60) = 0.27, p = 0.70, \eta_p^2 = 0.01$). Having failed to detect
within person differences, cortisol values for each sampling point were averaged across sampling days to obtain more reliable markers of basal HPA activity (Abercrombie et al., 2004; Miller et al., 2008; O’Connor et al., 2009).

3.1.5 Additional Measures of Protocol Adherence

One participant reported collection of the waking and 30 minute post waking samples more than 10 minutes outside the requested 30 minute time interval on the second sampling day. In accordance with previous work, protocol adherent data from the remaining sampling days was averaged and taken forward for statistical analyses (Mommersteeg, Heijnen, Kavelaars, Lorenz, & Doornen, 2006; Sjögren et al., 2006).

3.1.6 Treatment of Results

A series of chi square ($\chi^2$) and one way ANOVAs were used to assess demographic, behavioural and biomedical disparities between the groups. One way ANOVAs were also used to assess group differences in derived HPA indices and reported incidences of common health problems, with partial eta squared as the effect size ($\eta_p^2$). Differences at individual time points and mean diurnal cortisol output were assessed by way of a two (group: higher vs. lower stress) x four (time: waking, 30 minutes post waking, 1200h and 2200h) mixed ANOVA. Huynh-Feldt correction was applied whenever sphericity was violated. Following the guidelines of Baron and Kenny (1986), variation in basal stress hormone activity was assessed as one potential mediator for perceived stress related disparities in common health problems. That is, one way analysis of covariance (ANCOVA) was applied to remove any variance in reported incidences of common health problems that could be attributed to group differences in cortisol output. These mediation techniques have
been applied to test similar physiological parameters, and in similar populations (Cohen et al., 1999; Miller et al., 2002).

3.2 Results

3.2.1 Potential Confounds

Table 3.1 presents demographic, behavioural and biomedical data by stress group. Chi square revealed no group differences with respect to use of oral contraceptives ($\chi^2 = 2.03$, df = 1, $p = 0.15$) or phase of the menstrual cycle ($\chi^2 = 0.89$, df = 1, $p = 0.35$). One way ANOVA also yielded no group disparities in age, weight, BMI or time of waking (all $p$s > 0.07, all $\eta^2_p < 0.11$). Therefore, none of these variables were included in further statistical analyses.

Table 3.1

Means and Standard Deviations for Demographic, Behavioural and Biomedical Characteristics by Stress Group

<table>
<thead>
<tr>
<th></th>
<th>Higher stress (n = 16)</th>
<th>Lower stress (n = 16)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age</strong></td>
<td>19.3 (0.9)</td>
<td>19.1 (1.2)</td>
</tr>
<tr>
<td><strong>Weight</strong></td>
<td>129.5 (14.8)</td>
<td>138.4 (9.4)</td>
</tr>
<tr>
<td><strong>BMI</strong></td>
<td>21.4 (2.1)</td>
<td>22.4 (1.2)</td>
</tr>
<tr>
<td><strong>Time of waking</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Day one</td>
<td>09.05</td>
<td>08.34</td>
</tr>
<tr>
<td>Day two</td>
<td>08.38</td>
<td>08.26</td>
</tr>
<tr>
<td>Day three</td>
<td>09.13</td>
<td>08.22</td>
</tr>
<tr>
<td><strong>Oral contraceptives</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>7</td>
<td>11</td>
</tr>
<tr>
<td>No</td>
<td>9</td>
<td>5</td>
</tr>
<tr>
<td><strong>Menstrual cycle</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phase one</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>Phase two</td>
<td>10</td>
<td>8</td>
</tr>
</tbody>
</table>
3.2.2 Physiological Outcomes

Table 3.2 presents means and standard deviations for perceived levels of stress, HPA indices and common health problems by stress group. Mixed ANOVA yielded a significant main effect of time ($F(2.3, 68.7) = 120.18$, $p < 0.01$, $\eta_p^2 = 0.80$), reflecting the typical descending pattern of cortisol secretion across the day. Data also revealed a significant main effect of group ($F(1, 30) = 4.99$, $p = 0.03$, $\eta_p^2 = 0.14$), reflecting higher mean diurnal output of cortisol in the higher stress group. A significant group x time interaction effect was also observed ($F(2.3, 68.7) = 3.59$, $p = 0.03$, $\eta_p^2 = 0.11$). Follow up post hoc comparisons revealed significantly elevated cortisol in the higher stress group at 2200h only ($F(1, 30) = 7.65$, $p < 0.01$, $\eta_p^2 = 0.20$). Therefore, flatter diurnal cortisol slopes in the higher stress group, as evidenced by larger beta values ($F(1, 30) = 5.69$, $p = 0.02$, $\eta_p^2 = 0.16$), were largely accounted for by HPA hyperactivity in the evening.

Table 3.2
Means and Standard Deviations for Perceived Levels of Stress, HPA Indices and Incidences of Common Health Problems by Stress Group

<table>
<thead>
<tr>
<th></th>
<th>Higher stress (n = 16)</th>
<th>Lower stress (n = 16)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Perceived stress</strong></td>
<td>21.1 (3.1)</td>
<td>11.4 (3.0)</td>
</tr>
<tr>
<td><strong>HPA indices</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Waking (nmol/L)</td>
<td>12.3 (3.4)</td>
<td>10.6 (3.0)</td>
</tr>
<tr>
<td>30 minutes post waking (nmol/L)</td>
<td>15.2 (5.6)</td>
<td>15.5 (5.0)</td>
</tr>
<tr>
<td>1200h (nmol/L)</td>
<td>7.9 (3.4)</td>
<td>6.2 (2.8)</td>
</tr>
<tr>
<td>2200h (nmol/L)</td>
<td>4.4 (3.3)</td>
<td>2.3 (1.9)</td>
</tr>
<tr>
<td>Diurnal cortisol slope ($\beta$)</td>
<td>-.64 (0.3)</td>
<td>-.84 (0.2)</td>
</tr>
<tr>
<td>CAR (nmol/L)</td>
<td>1.9 (0.2)</td>
<td>4.9 (0.1)</td>
</tr>
<tr>
<td>Mean diurnal cortisol output (nmol/L)</td>
<td>10.1 (0.1)</td>
<td>8.7 (0.1)</td>
</tr>
<tr>
<td><strong>Incidences of common health problems</strong></td>
<td>20.1 (9.4)</td>
<td>13.4 (7.5)</td>
</tr>
</tbody>
</table>

*BMI, body mass index; CAR, cortisol awakening response; nmol/L, nanomoles per litre*
Diurnal patterns of cortisol secretion for participants in the higher and lower stress groups are presented in Figure 3.1. Though blunted for participants experiencing higher perceived levels of stress, group disparities in CAR magnitude approached, but failed to reach statistical significance ($F(1, 30) = 3.23, p = 0.08, \eta_p^2 = 0.10$).

Figure 3.1 Diurnal patterns of cortisol secretion in the higher (n = 16) and lower (n = 16) stress groups. Error bars represent standard error of the mean ($\log_{10}$ data presented for illustrative purposes).
3.2.3 Incidences of Common Health Problems

Data revealed that subjective reports of ill health were significantly greater in the higher stress group \((F (1, 30) = 4.83, p = 0.04, \eta_p^2 = 0.14)\).

3.2.4 Mediation Analysis

According to Baron and Kenny (1986), mediation requires that: (a) the effect of the independent variable (perceived levels of stress) on the outcome variable (common health problems) is significant, (b) the path from the independent variable (perceived levels of stress) to the mediator (cortisol output) is significant, (c) the path from the mediator (cortisol output) to the outcome variable (common health problems) is significant, and for complete mediation, (d) the association between the independent variable (perceived levels of stress) and the outcome variable (common health problems) is appreciably reduced after controlling for the effect of the hypothesized mediator (cortisol output). Data reported here satisfy criteria (a) and (b), such that participants with higher perceived levels of stress reported greater incidences of common health problems \((p = 0.04, \eta_p^2 = 0.14)\) and displayed elevated mean diurnal cortisol output \((p = 0.03, \eta_p^2 = 0.14)\), and flatter diurnal cortisol slopes \((p = 0.02, \eta_p^2 = 0.14)\). To satisfy criteria (c), bivariate correlation revealed a significant positive relationship between mean diurnal cortisol output and common health problems \((r = 0.33, n = 32, p = 0.03)\). Presented in Figure 3.2 is a path model displaying the relationships between perceived levels of stress, mean diurnal cortisol output and incidences of common health problems.
Statistically controlling for mean diurnal cortisol output caused a substantial reduction in the percent variance of self reported incidences of common health problems explained by perceived levels of stress from $\eta^2_p = 0.14$ (in the original analysis) to $\eta^2_p = 0.08$. That is, after partialing out the effect of mean levels of cortisol across the day, the effect size for perceived levels of stress on common health problems was reduced by 43.9%. Perceived stress related disparities in common health problems also became non-significant under these conditions ($F (1, 29), 2.55, p = 0.12, \eta^2_p = 0.08$). These findings suggest the negative impact of higher perceived levels of stress on subjective reports of ill health was partially mediated by HPA hyperactivity.
3.3 Discussion

Study one investigated the effect of perceived stress on basal organisation of the HPA axis and subjective reports of common health problems in a sample of young otherwise healthy individuals. Dysregulated HPA activity, manifested by flatter cortisol slopes (largely accounted for by higher evening values) and elevated mean diurnal cortisol output was apparent in the higher stress group. Self reported incidences of common health problems were also greater in participants experiencing higher perceived levels of stress. Findings reported here converge with other studies involving young otherwise healthy individuals that have individually linked higher perceived levels of stress with atypical patterns of cortisol secretion (Abercrombie et al., 2004; O’Connor et al., 2009) and greater subjective reports of common health complaints (Hellhammer et al., 2004; Sjögren et al., 2006). These data are also commensurate with studies that have observed disruptions in other disease relevant physiological processes such as antibody response to vaccination in participants experiencing higher perceived levels of stress (Burns et al., 2002).

The immunomodulatory properties of cortisol have been well documented in the stress literature (Elenkov, 2004; McEwen et al., 1997; Sapolsky et al., 2000), and therefore, it was predicted that differential patterns of cortisol secretion might account for the group effect on ill health. Indeed, perceived stress related disparities in reported incidences of ill health were appreciably reduced when mean diurnal cortisol output was statistically controlled. These data implicate cortisol hypersecretion as one physiological determinant for greater incidences of common health problems in young otherwise healthy individuals experiencing higher perceived levels of stress. Findings reported here also converge with other studies that have implicated dysregulated HPA activity and concomitant alterations in
proinflammatory cytokine secretion, as one physiological pathway that links higher perceived levels of stress with more severe episodes of common health problems such as upper respiratory illness (Cohen et al., 1999).

The present study has a number of shortcomings. The cross sectional nature of the study precludes drawing causal inferences. That is, proinflammatory cytokines, which are released in response to common health problems such as infectious pathologies (Dantzer et al., 2008; Elenkov, 2004), might have been responsible for increased HPA activity in the higher stress group. The sample size, while modest, is comparable in magnitude (O’Connor et al., 2009) or exceeds (Cohen et al., 1999) other studies that have assessed similar research questions, and in similar populations. Study one does have other shortcomings, most of which are shared by subsequent studies in the thesis. Therefore, shared programme limitations will be considered in the general discussion chapter at the end of the thesis.

In conclusion, participants experiencing higher perceived levels of stress displayed atypical cortisol secretion patterns characterised by flatter diurnal cortisol slopes and elevated mean levels of cortisol across the day. Self reported incidences of common health problems were also greater in the higher stress group. Data indicated that HPA hyperactivity partially mediated the effect of higher perceived levels of stress on greater incidences of the kinds of common health problems that typically affect young otherwise healthy individuals.

3.4 Concluding Remarks

As a programme of work, the thesis set out to investigate the psychophysiological pathways that link chronic stress with increased vulnerability for ill health. Findings from study one implicate atypical patterns of cortisol secretion
as one physiological determinant for greater incidences of ill health in young otherwise healthy individuals experiencing higher perceived levels of stress. As the first of the programme, study one provided a valuable learning experience with respect to the accurate collection of salivary biomarkers and helped better inform important protocol decisions in future studies. As a logical next step in the programme, study two used the caregiver control model to look more closely at the psychophysiological consequences of chronic stress.
4. Study Two

4.0 Chapter Overview

The study of informal caregivers has become one well established model for assessing the psychophysiological consequences of chronic stress. Increased psychological morbidity and perturbations in endocrine and immune parameters, indicative of increased vulnerability for ill health, have been widely evidenced in older caregivers of partners with dementing illness (Bauer et al., 2000; Dabrowska & Pisula, 2010; Da Roza Davis & Cowen, 2001; Gallagher-Thompson et al., 2006; Kiecolt-Glaser et al., 2003; Vedhara et al., 1999; von Kanel, Dimsdale, & Mills et al., 2006). However, older caregivers are a unique population, who are typically contending with pre-existing endocrine and immune impairments associated with their advancing age (Bauer, 2005; Sapolsky, 1987), thus making it difficult to disentangle stress induced physiological alterations from the natural effects of senescence.

Therefore, researchers have started to consider whether chronic caregiver stress results in endocrine and immune dysfunction in relatively young caregivers, who are not contending with age related impairments in these physiological systems. To date, findings have been mixed. That is, Vedhara et al (2002) demonstrated that younger caregivers of patients with multiple sclerosis (MS), a severe physical impairment, and age matched non caregiving controls could not be differentiated on humoral response to influenza vaccination. These findings suggest the adverse effect of chronic caregiver stress on immune function might be exclusive to older dementia caregivers, and as such, requires an already impaired immune system (i.e., senescence) to manifest. However, diminished antibody response to pneumococcal vaccination was recently observed in relatively young caregivers of children with
additional complex needs such as autism (Gallagher et al., 2009), a condition characterised by the same kinds of distressing cognitive and behavioural impairments that affect dementia patients. Elevated markers of inflammation such as IL-6 and CRP have also been observed in younger caregivers of children experiencing the severe cognitive and behavioural sequelae associated with aggressive brain cancer (Miller et al., 2008; Rohleder et al., 2009). Indeed, relative to caregivers of patients with severe physical impairments such as MS, psychological distress is considerably greater and the caregiver experience much more intense in caregivers of patients with distressing cognitive and behavioural impairments such as dementia (Pinquart & Sorensen, 2003) and autism (Bouma & Schweitzer, 1990). Taken together, these findings indicate it is not caregivers’ age (i.e., senescence) that is important, rather specific characteristics of the care recipient (i.e., the nature of the impairment) and concomitant variations in the intensity of the caregiver experience that dictate whether caregiver stress becomes an issue for disease relevant physiological processes such as the immune response.

Consequently, study two compared relatively young caregivers of children experiencing distressing cognitive (i.e., autism) and behavioural (i.e., ADHD) impairments with a normative control group on a range of psychophysiological outcomes. Study two also examined whether social support, which is typically diminished in relatively young caregivers (Edworthy, 2005; Fletcher, et al., 2012; Gallagher & Whiteley, 2012b; Lach et al., 2009) and often linked with immune alterations (Kiecolt-Glaser et al., 2009; Lutgendorf et al., 2000; Moynihan et al., 2004), might underlie the effect of chronic caregiver stress on immune dysfunction. To assess the additive effects of caring for a typically developing child, a group of non parents, analogous in makeup to those used in study one was also recruited.
In accordance with previous research, preliminary analyses revealed that caregivers of children with autism and ADHD were statistically indistinguishable with respect to all psychophysiological outcomes (Donnenberg & Baker, 1993; Lach et al., 2009). Data also revealed that parents of typically developing children and the group of non parents could not be differentiated on all measures of interest. These findings suggest it is not parenting per se, rather that caring for a child with additional complex needs dictates whether caregiver stress becomes an issue for health related outcomes. On the basis of these findings, autism and ADHD caregivers were treated as one uniform group, and were compared in all subsequent analyses with a single control group comprising parents of typically developing children, and non parents.

Data revealed that psychological morbidity was significantly greater in the caregivers. Indeed, caregivers’ mean anxiety and depression scores satisfied current criterion for borderline clinical mood disorder compared with normative scores in the control group (Snaith, 2003). Caregivers also reported significantly diminished social support and greater incidences of common health problems. However, findings revealed no caregiver related disparities with respect to basal stress hormone activity; indeed, both caregivers and controls displayed the typical descending pattern of cortisol across the day and were comparable on HPA markers, CAR magnitude, diurnal cortisol slope and mean diurnal cortisol output. Data further revealed that concentrations of the proinflammatory biomarker, CRP were significantly elevated in the caregiver group. Taken together, these findings converge with other recent studies that have demonstrated increased psychological morbidity (Bella et al., 2011; Gallagher, Phillips, Oliver, & Carroll, 2008; Seltzer et al., 2009; Smith et al., 2012), increased reports of ill health (Ha et al., 2011; Ingersoll & Hambrick, 2011; Lach et
al., 2009; Smith et al., 2012), lower social support (Gallagher & Whiteley, 2012b; Ludlow et al., 2012) and dysregulated immunity, manifested by increased markers of inflammation (Miller et al., 2008; Rohleder et al., 2009) in relatively young caregivers of children with additional complex needs. Failure to observe caregiver related disparities in basal cortisol secretion patterns will be discussed in the context of the HPA rebound effect (Miller et al., 2007).

4.1 Methods

4.1.1 Participants

A sample of 56 caregivers of children with autism/ADHD was recruited from local and regional support groups, special schools and charities across North East England. A group of 22 parents of typically developing children and 22 non parents were sought from an under/postgraduate and staff population. All caregivers were recruited according to strict criteria which included: (a) aged between 18-60 years, (b) providing care for at least one child (aged between 3-19 years) with a clinical diagnosis of autism or ADHD, who was (c) living at home on a full time basis, (d) not providing care for another individual with chronic illness, (e) not pregnant, breast feeding, taking any steroidal and/or hormone replacement medication, (f) not experiencing any chronic illness and (g) not working night shift. Research has indicated that parent carers experience increased emotional distress throughout the diagnostic process (Graungaard & Skov, 2007). Therefore, to focus on stressful experiences exclusive to the caregiver role, and in line with relevant previous research, only parents of children aged 3-19 years were eligible to take part (Gallagher et al., 2009; Hastings et al., 2006). Parents of typically developing children and non parents were recruited according to the same criteria, though to be
eligible, must not have been: (a) experiencing any chronically stressful life events (e.g., divorce, bereavement), or (b) providing care for an individual with chronic illness. Participants who satisfied these criteria were invited to provide informed consent following an ethically approved protocol endorsed by the Department of Psychology Ethics Committee.

Of the 100 participants recruited in total, three withdrew citing time constraints and 16 failed to provide any/sufficient saliva, and/or blood for biochemical assay. Adherence checks identified four participants with delays of more than 10 minutes between waking and collection of the waking cortisol sample on both sampling days. In addition, two participants reported collection of the waking and 30 minute post waking samples more than 10 minutes outside of the requested 30 minute time interval on both sampling days. These six participants were also excluded. As a likely indicator of infection at the time of assessment, three participants were excluded based on CRP concentrations greater than 10mg/l (Fuligni et al., 2009; Taylor, Lehman, Kiefe, & Seeman, 2006), as was one participant who disclosed a clinically verified psychological disorder. Statistical analysis was conducted on a final sample of 71 participants comprising 39 autism/ADHD caregivers, 17 parents of typically developing children and 15 non parents. All participants were recompensed £10.00 for their time.

4.1.2 Potential Confounds

Demographic, behavioural and biomedical data were collected to assess whether caregiver related disparities in psychophysiological outcomes might reflect the contribution of other variables.
4.1.3 Psychosocial Outcomes

Perceived stress was quantified using the 10 item PSS. The HADS was used to measure anxiety and depression, and the ISEL used to assess social support. The PILL was used to quantify incidences of 54 common health problems (see Chapter Two for full details of all psychosocial measures). All instruments achieved excellent psychometrics in the sample (all Cronbach’s alphas > 0.86).

4.1.4 Endocrine Outcomes

To capture important measures that summarise aspects of the diurnal cortisol pattern, participants collected saliva samples at waking, 30 minutes post waking, 1200h and 2200h on two consecutive weekdays (see Chapter Two for full details of the saliva collection protocol and checks on adherence). Repeated measures ANOVA was used to explore possible within person differences in cortisol data per individual sample point. Violated assumptions were corrected with Huynh Feldt. Findings revealed a significant main effect of time ($F(2.6, 150.5) = 447.9, p < 0.01, \eta^2_p = 0.89$), reflecting the typical descending pattern of cortisol secretion. However, no between day differences ($F(1, 57) = 0.09, p = 0.77, \eta^2_p < 0.01$) or day x time interaction effect ($F(2.6, 147.5) = 2.02, p = 0.19, \eta^2_p = 0.03$) was observed. Data also revealed no between day differences with respect to CAR magnitude ($F(1, 57) = 1.13, p = 0.29, \eta^2_p = 0.02$) or diurnal cortisol slope ($F(1, 57) = 0.15, p = 0.70, \eta^2_p < 0.01$). Having failed to detect within person differences, cortisol values for each sampling point were averaged across sampling days to obtain more reliable indices of basal HPA activity (O’Connor et al., 2009; Turner-Cobb et al., 2010).
4.1.5 Additional Measures of Protocol Adherence

One participant reported a delay of more than 10 minutes between waking and collection of the waking cortisol sample, and five participants reported collection of their waking and 30 minute post waking samples more than 10 minutes outside of the requested 30 minute time interval on one sampling day. An additional four participants failed to complete any adherence checks, and three participants failed to collect sufficient quantities of saliva for biochemical assay on one sampling day. In accordance with previous research, protocol adherent data from the remaining sampling day was taken forward for statistical analyses (Mommersteeg et al., 2006; Sjögren et al., 2006).

4.1.6 Immune Outcomes

Venous blood was collected and the obtained plasma assessed for concentrations of the inflammatory biomarkers, IL-6 and CRP. Fasting blood was taken from all participants between 10am-12pm to control for diurnal variations. Plasma samples frozen at -80°C were thawed and IL-6 quantified in duplicate using a commercial high-sensitivity enzyme-linked immunosorbant assay (Quantikine human IL-6, R&D Systems, Minneapolis, MO). This assay has a minimum detection threshold of 0.70 pg/ml. Mean intra and inter-assay coefficients were 6.9% and 9.6% respectively. CRP was assessed in duplicate by high sensitivity ELISA (CRP, Kalon Biological Ltd, Guildford, UK). This assay has a minimum detection threshold of 0.2 mg/l. Mean intra and inter-assay coefficients were 5.2% and 12.0% respectively. IL-6 ($D (71) = 0.21, p < 0.01$) and CRP ($D (71) = 0.27, p < 0.01$) data were log$_{10}$ transformed to correct for skewed distribution.
4.1.7 Treatment of Results

Preliminary analyses revealed that caregivers of children with autism and ADHD were statistically indistinguishable on all psychophysiological outcomes which included: psychological distress (all $ps > 0.45$, all $\eta_p^2 < 0.02$), social support (all $ps > 0.14$, all $\eta_p^2 < 0.06$), common health problems ($F(1, 37) = 0.79$, $p = 0.78$, $\eta_p^2 < 0.01$), HPA indices (all $ps > 0.07$, all $\eta_p^2 < 0.08$) and markers of inflammation (all $ps > 0.08$, all $\eta_p^2 < 0.08$). Data also revealed that parents of typically developing children and the group of non parents could not be differentiated on all measures of interest (all $ps > 0.08$, all $\eta_p^2 < 0.09$). In view of these findings, autism and ADHD caregivers were treated as one uniform group, an approach widely applied in previous research involving relatively young caregivers of children with additional complex needs (Gallagher & Whiteley, 2012a; Seltzer et al., 2009, 2010). Caregivers were compared in all subsequent analyses with a single control group composed of parents of typically developing children and non parents. Table 4.1 presents means and standard deviations for psychophysiological outcomes by group.

A series of chi square ($\chi^2$) and one way ANOVAs were used to examine group differences in demographic, behavioural and biomedical factors. Caregiver related disparities with respect to psychological distress (i.e., perceived stress, anxiety and depression), social support, ill health, HPA indices and inflammatory markers were explored using a series of one way ANCOVAs, with partial eta squared as the effect size ($\eta_p^2$). All between group comparisons were adjusted for multiple comparisons using Bonferroni. Two (group: caregivers and controls) x four (time: waking, 30 minutes post waking, 1200h and 2200h) mixed ANCOVA was used to explore group differences in cortisol data per individual sample point and mean output across the day. Huynh Feldt correction was applied to correct violations
of sphericity as necessary. Following the guidelines of Baron and Kenny (1986), ANCOVA was used to assess whether variations in social support might account for immune disparities between the groups. Indeed, these mediation techniques have been widely applied in previous research involving chronically stressed caregivers (Bozo et al., 2010; Miller et al., 2002; Vitaliano et al., 2005).

4.2 Results

4.2.1 Potential Confounds

Summary characteristics for the two groups are presented in Table 4.1. Caregivers and controls were statistically indistinguishable on gender, weight, BMI, marital status, number of children, annual household income, use of nicotine, alcohol consumption, oral contraceptives, phase of the menstrual cycle, frequency of exercise and time of waking (all \( ps > 0.11 \), all \( \eta^2_p < 0.05 \)). However, caregivers were significantly older than controls (\( F(1, 69) = 21.7, p < 0.01, \eta^2_p = 0.24 \)) and more likely to be using antidepressants (\( \chi^2 = 6.81, df = 1, p < 0.01 \)). Therefore, age and use of antidepressants were statistically controlled in all subsequent analyses.

Table 4.1

Means and Standard Deviations for Demographic, Behavioural and Biomedical Characteristics by Group

<table>
<thead>
<tr>
<th></th>
<th>Caregivers (n = 39)</th>
<th>Controls (n = 32)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Gender</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>5</td>
<td>8</td>
</tr>
<tr>
<td>Female</td>
<td>34</td>
<td>24</td>
</tr>
<tr>
<td><strong>Age</strong></td>
<td>44.6 (7.1)</td>
<td>36.0 (8.4)</td>
</tr>
<tr>
<td><strong>Weight</strong></td>
<td>159.9 (41.9)</td>
<td>151.1 (26.6)</td>
</tr>
<tr>
<td><strong>BMI</strong></td>
<td>26.3 (6.1)</td>
<td>24.6 (3.5)</td>
</tr>
<tr>
<td><strong>Nicotine (cigarettes per week)</strong></td>
<td>1.9 (5.7)</td>
<td>1.1 (4.0)</td>
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</table>
Table 4.1 (continued)

<table>
<thead>
<tr>
<th></th>
<th>Caregivers (n = 39)</th>
<th>Controls (n = 32)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alcohol (units per week)</td>
<td>5.8 (7.5)</td>
<td>7.3 (7.9)</td>
</tr>
<tr>
<td>Exercise (occasions per week)</td>
<td>2.9 (2.4)</td>
<td>2.4 (1.8)</td>
</tr>
<tr>
<td>Annual income (£)</td>
<td>37142 (29115)</td>
<td>34026 (19236)</td>
</tr>
<tr>
<td>Number of children</td>
<td>2.2 (1.1)</td>
<td>1.7 (0.6)</td>
</tr>
<tr>
<td>Time of waking (day one)</td>
<td>7.20 (1.1)</td>
<td>7.20 (0.1)</td>
</tr>
<tr>
<td>Time of waking (day two)</td>
<td>7.08 (1.0)</td>
<td>7.30 (1.0)</td>
</tr>
<tr>
<td>Marital status</td>
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<tr>
<td>Partnered</td>
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<td>22</td>
</tr>
<tr>
<td>Not partnered</td>
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</tr>
<tr>
<td>Menstrual cycle</td>
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</tr>
<tr>
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</tr>
<tr>
<td>Phase two</td>
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<tr>
<td>Oral contraceptives</td>
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<td>Yes</td>
<td>4</td>
<td>6</td>
</tr>
<tr>
<td>No</td>
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</tr>
<tr>
<td>Antidepressant medications</td>
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</tr>
<tr>
<td>No</td>
<td>29</td>
<td>31</td>
</tr>
</tbody>
</table>

4.2.2 Psychosocial Outcomes

Table 4.2 presents means and standard deviations for psychophysiological outcomes by group. Perceived stress ($F(1, 67) = 22.1, p < 0.01, \eta_p^2 = 0.25$), anxiety ($F(1, 67) = 20.6, p < 0.01, \eta_p^2 = 0.24$) and depression scores ($F(1, 67) = 13.5, p < 0.01, \eta_p^2 = 0.17$) were significantly higher in the caregivers, as were subjective reports of ill health ($F(1, 67) = 13.6, p < 0.01, \eta_p^2 = 0.17$). Caregivers also scored significantly lower on the appraisal ($F(1, 67) = 6.1, p = 0.02, \eta_p^2 = 0.08$), self esteem ($F(1, 67) = 9.7, p < 0.01, \eta_p^2 = 0.13$) and belonging ($F(1, 67) = 5.0, p = 0.03, \eta_p^2 = 0.07$) subscales of the ISEL relative to the control group.
Table 4.2

**Means and Standard Deviations for Psychophysiological Outcomes by Group**

<table>
<thead>
<tr>
<th></th>
<th>Caregivers (n = 39)</th>
<th>Controls (n = 32)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Perceived stress</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HADS</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anxiety</td>
<td>22.7 (6.3)</td>
<td>16.1 (4.8)</td>
</tr>
<tr>
<td>Depression</td>
<td>9.9 (4.5)</td>
<td>5.4 (3.2)</td>
</tr>
<tr>
<td><strong>ISEL subscales</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Appraisal</td>
<td>21.7 (6.8)</td>
<td>25.1 (5.4)</td>
</tr>
<tr>
<td>Tangible</td>
<td>21.6 (6.6)</td>
<td>23.6 (5.1)</td>
</tr>
<tr>
<td>Self esteem</td>
<td>17.4 (4.9)</td>
<td>20.6 (3.2)</td>
</tr>
<tr>
<td>Belonging</td>
<td>20.1 (6.6)</td>
<td>23.3 (5.1)</td>
</tr>
<tr>
<td><strong>Incidences of common health problems</strong></td>
<td>17.5 (11.1)</td>
<td>9.8 (6.4)</td>
</tr>
<tr>
<td><strong>HPA indices</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Waking (nmol/L)</td>
<td>14.9 (6.0)</td>
<td>13.4 (4.4)</td>
</tr>
<tr>
<td>30 minutes post waking (nmol/L)</td>
<td>20.4 (8.4)</td>
<td>19.7 (6.5)</td>
</tr>
<tr>
<td>1200h (nmol/L)</td>
<td>6.9 (5.8)</td>
<td>5.9 (2.4)</td>
</tr>
<tr>
<td>2200h (nmol/L)</td>
<td>2.9 (5.8)</td>
<td>2.7 (3.3)</td>
</tr>
<tr>
<td>Diurnal cortisol slope ($\beta$)</td>
<td>-0.84 (0.3)</td>
<td>-0.83 (0.4)</td>
</tr>
<tr>
<td>CAR (nmol/L)</td>
<td>5.5 (8.6)</td>
<td>6.3 (6.1)</td>
</tr>
<tr>
<td>Mean diurnal cortisol output (nmol/L)</td>
<td>11.3 (4.7)</td>
<td>10.4 (2.7)</td>
</tr>
<tr>
<td><strong>Proinflammatory biomarkers</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IL-6 (pg/mL)</td>
<td>1.6 (1.5)</td>
<td>1.5 (2.0)</td>
</tr>
<tr>
<td>CRP (mg/L)</td>
<td>1.3 (1.8)</td>
<td>0.6 (1.1)</td>
</tr>
</tbody>
</table>

**CAR**, cortisol awakening response; **CRP**, C-reactive protein; **HADS**, Hospital Anxiety and Depression Scale; **IL**, interleukin; **ISEL**, Interpersonal Support Evaluation Checklist; **mg/L**, milligrams per litre; **nmol/L**, nanomoles per litre; **pg/mL**, pictograms per millilitre

4.2.3 Endocrine Outcomes

A two (group: caregivers vs. controls) x four (time: waking, 30 minutes post waking, 1200h, and 2200h) mixed ANCOVA yielded a significant main effect of time ($F (2.5, 166.9) = 3.84, p = 0.02, \eta_p^2 = 0.05$) reflecting the typical decline in cortisol secretion across the day. No group disparities emerged with respect to mean diurnal cortisol output ($F (1, 67) = 0.42, p = 0.52, \eta_p^2 < 0.01$) or interaction effect ($F (2.5, 166.9) = 0.34, p = 0.72, \eta_p^2 < 0.01$). In addition, caregivers could not be differentiated from controls on CAR magnitude ($F (1, 67) = 0.03, p = 0.85, \eta_p^2 < 0.01$).
0.01) or diurnal cortisol slope \((F(1, 67), = 0.65, p = 0.42, \eta^2 = 0.01)\). Figure 4.1 displays diurnal patterns of cortisol secretion for caregivers and controls.

4.2.4 Immune Outcomes

Concentrations of the inflammatory marker, CRP were significantly higher in the caregivers \((F(1, 67) = 5.03, p = 0.03, \eta^2 = 0.07)\). However, groups could not be differentiated on concentrations of IL-6 \((F(1, 67) = 1.80, p = 0.19, \eta^2 = 0.03)\).

Figure 4.1 Diurnal patterns of cortisol secretion in the caregivers \((n = 39)\) and controls \((n = 32)\). Error bars represent standard error of mean (\(\log_{10}\) data presented for illustrative purposes).
4.2.5 Mediation Analysis

Lower social support was examined as one psychosocial determinant for higher CRP concentrations in the caregivers. Data yielded no significant relationships between scores on individual ISEL subscales and CRP concentrations (all $p$s > 0.36). Moreover, group differences in CRP were not appreciably reduced when scores on individual ISEL subscales were statistically controlled (all $p$s < 0.04, all $\eta_p^2$ < 0.07). Thus, the effect of chronic caregiver stress on higher proinflammatory activity was not mediated by diminished social support. Associations between caregiver stress, social support (ISEL) and CRP are presented in Figure 4.2.

![Figure 4.2](image.png)

**Figure 4.2** Path model displaying the relationships between caregiver stress, social support (ISEL) and CRP concentrations. CRP, C-reactive protein; ISEL, Interpersonal Support Evaluation Checklist

4.3 Discussion

Data reported here suggest the psychophysiological effects of stress apparent in older dementia caregivers might extend to relatively young caregivers of children.
with autism/ADHD, conditions that share many clinical manifestations of dementia. Indeed, relative to a normative comparison group, caregivers reported increased psychological morbidity, as indexed by higher perceived stress, anxiety and depression scores. In fact, caregivers’ mean anxiety and depression scores satisfied current criterion for borderline clinical mood disorder, compared with normative scores in the controls (Snaith, 2003). However, basal stress hormone activity did not differentiate the groups. Indeed, both caregivers and controls displayed the typical descending pattern of cortisol secretion across the day and were comparable on CAR magnitude, diurnal cortisol slope and mean diurnal cortisol output. These findings converge with recent studies that also demonstrated a disassociation between psychological and physiological markers in the context of caring for a child with additional complex needs (Miller et al., 2008; Rohleder et al., 2009). As one possible explanation, sustained elevated levels of cortisol (as a result of repeated stress) rebound to normal (and below normal) levels over time as a function of increased feedback sensitivity (Gunnar & Vasquez, 2001), or down regulation of pituitary CRH receptors (Raison & Miller, 2003). Indeed, meta analysis has revealed an inverse relationship between time since stressor onset and HPA output (Miller et al., 2007).

Data further revealed that concentrations of the inflammatory marker, CRP were significantly elevated in the caregivers. According to current clinical guidelines, caregivers’ mean concentrations of CRP (1.3 mg/L) indicated moderate risk for incident cardiovascular pathologies, compared with low risk in the control group (Pearson et al., 2003; Ridker et al., 2003). Proinflammatory agents also signal the brain to induce the sickness syndrome, a range of non specific sickness symptoms including malaise, nausea and headaches (Dantzer, 2001), and have been implicated in the aetiologies of other common health problems such as upper
respiratory illness (Cohen et al., 1999). In view of these findings, increased markers of inflammation might account for the group effect on common health problems reported here. Indeed, findings reported here converge with other studies that have observed greater frequencies of ill health in caregivers of children with additional complex needs relative to normative controls (Byrne et al., 2010; Gallagher & Whiteley, 2012b; Ha et al., 2011; Hedov, Annren, & Wikblad, 2000; Ingersoll & Hambrick, 2011; Lach et al., 2009; Mugno, Ruta, D'Arrigo, & Mazzone, 2007; Ones, Yilmaz, Cetinkaya, & Caglar, 2005; Seltzer et al., 2001; Smith et al., 2011).

The GC resistance model states that immune cells compensate for sustained elevated cortisol secretion (as a result of repeated stress) by down regulating GC receptor activity. Therefore, in cells such as macrophages, cortisol becomes less able to inhibit proinflammatory transcriptional control pathways (Miller et al., 2002, 2008; Rohleder, 2011), thus rendering the host more susceptible to diseases fostered by persistent low grade inflammation (Hamer et al., 2010; Hu et al., 2004; Ridker et al., 2003; Spence et al., 2008). Reduced immune system GC sensitivity and concomitant elevation of inflammatory biomarkers such as CRP has been observed in caregivers of children with additional complex needs. However, as was the case here, variations in basal stress hormone activity did not account for the group effect on proinflammatory activity (Miller et al., 2008; Rohleder et al., 2009). It might be contended that sustained elevated cortisol secretion and associated reduction in immune system GC sensitivity was responsible for increased markers of inflammation in the early stages of the caregiver experience. However, over time, cortisol levels might have rebounded to normal (Miller et al., 2007) while GC receptors on immune cells remained down regulated. Future studies might use more
prospective designs to track changes in basal stress hormone activity across the
caregiver experience as evidence for HPA rebound.

Research has demonstrated that, relative to non caregiving controls,
caregivers of children with additional complex needs experience painful social
isolation (Edworthy, 2005; Garcés et al., 2010) and have fewer opportunities for
social connectivity (Gallagher & Whiteley, 2012b; Lach et al., 2009; Weiss, 2002).
Moreover, lower social support has been implicated as one psychosocial determinant
for perturbations in basal HPA activity (Seltzer et al., 2009) and cardiovascular
functioning (Gallagher & Whiteley, 2012a) in the context of chronic caregiver stress.
In the present study, caregivers scored lower on the appraisal, belonging and self
esteem subscales of the ISEL; however, caregiver related disparities in CRP
concentrations were not appreciably reduced when social support scores were
statistically controlled. Thus, the negative impact of caring for a child with
autism/ADHD on immune function was not mediated by diminished social support.

The current study should be evaluated in the context of its limitations. The
cross sectional nature of the study precludes drawing causal inferences about the
direction of relationships between study variables. For example, proinflammatory
chemicals signal the brain to induce non specific symptoms of sickness including
low mood and feelings of depression (Dantzer, 2001; Elenkov, 2004), and as such,
might have been responsible for higher levels of psychological distress in the
caregiver group. More prospective designs will help clarify the direction and
temporal sequence of these relationships.

Outputs of the SAM axis (i.e., adrenaline, noradrenaline) modulate the
secretion of proinflammatory cytokines (Evans et al., 2000), as well as the expression
of GC receptors on immune cells (DeRijk et al., 1996). To this end, future studies
might examine outputs of the SAM axis as additional physiological intermediaries that translate chronic caregiver stress into increased proinflammatory activity.

In a recent study, problem behaviours of the care recipient were implicated as one psychosocial determinant for immune dysfunction in relatively young caregivers of children with additional complex needs such as autism (Gallagher et al., 2009). Indeed, it has been widely evidenced that problem behaviours of the care recipient exacerbate the harmful effects associated with the caregiver experience (Baker et al., 2002; Gallagher, Phillips, Oliver, & Carroll, 2008; Hastings, 2003; Lecavalier, Leone, & Wiltz, 2006; White & Hastings, 2004). Failure to assess whether care recipients’ problem behaviours mediated the effect of chronic caregiver stress on elevated markers of inflammation represents a significant and major limitation of the present study.

It is unclear why, as the driver for CRP synthesis from liver hepatocytes (Dantzer et al., 2008), elevated levels of IL-6 were not apparent in the caregiver group. As one possible explanation, regulation of CRP synthesis from the liver is contingent on a complex interplay between IL-6 and other proinflammatory cytokines such as IL-1 and TNF-α (Heinrich, Castell, & Andus, 1990). Therefore, future studies might assess additional markers of inflammation to gain a clearer understanding of proinflammatory dynamics in the context of chronic caregiver stress. Finally, research has demonstrated the potential immunomodulatory influence of statins (Bu, Griffin, & Lichtman, 2011; Iwata et al., 2012); however, use of statins was not assessed in the current study, and therefore, its effects are unknown.

In conclusion, caring for a child with autism/ADHD exacts a considerable psychophysiological toll on the carer. Indeed, even in the absence of dysregulated HPA activity, caregivers displayed elevated markers of inflammation, and as such,
might be at greater risk for adverse cardiovascular events (Ridker, 2003), and other
diseases fostered by persistent low grade inflammation (Hamer et al., 2010; Hu et al.,
2004; Spence et al., 2008).

4.4 Concluding Remarks

Findings from study two indicated the harmful effects apparent in older dementia
caregivers might extend to relatively young caregivers of children with
autism/ADHD, conditions characterised by the same kinds of distressing cognitive
and behavioural impairments that affect dementia patients. Indeed, relative to a
normative comparison group, caregivers reported increased psychological morbidity
and greater incidences of common health problems. Concentrations of the
proinflammatory biomarker, CRP were also elevated in the caregiver group. These
findings converge with other studies that have observed immune dysfunction,
manifested by increased markers of inflammation, in caregivers of children
experiencing the cognitive and behavioural sequelae associated with aggressive brain
cancer (Miller et al., 2008; Rohleder et al., 2009). However, data revealed that
variations in social support did not account for the group effect on proinflammatory
activity.

Research has indicated that greater social support might protect against the
psychophysiological consequences associated with chronic caregiver stress. Indeed,
socially supported caregivers of children with additional complex needs have
reported diminished levels of psychological distress (Ekas et al., 2010; Smith et al.,
2011; Weiss, 2002; White & Hastings, 2004) and fewer negative health complaints
(Lin et al., 2009; Sawyer et al., 2010). However, fewer studies have assessed whether
greater social support might protect relatively young caregivers against disturbances
in disease relevant physiological processes. In a recent study, Gallagher and Whiteley (2012a) reported that socially supported caregivers of children with additional complex needs such as autism were relatively protected against higher daytime systolic blood pressure, one well established risk factor for cardiovascular pathologies. To extend the relatively small research base in this area, study three examined whether greater social support might ameliorate psychological distress, and in so doing, protect autism/ADHD caregivers against perturbations in the HPA axis and immune response.
5. Study Three

5.0 Chapter Overview

Findings from study two indicated that caring for a child with autism/ADHD exacts a significant psychophysiological toll on the carer, manifested by increased psychological morbidity, reduced social support, greater reports of ill health and dysregulated immunity. These findings converge with other studies that have observed similar harmful effects in the context of caring for a child with additional complex needs (Byrne, Hurley, Daly, & Cunningham, 2010; Gallagher et al., 2009; Seltzer et al., 2009, 2010; Smith et al., 2012). However, greater social support might protect against the harmful effects associated with the caregiver experience. Indeed, reduced psychological morbidity (Chien et al., 2011) and higher percentages of NK cells (Esterling et al., 1996; Kiecolt-Glaser et al., 1987), which is indicative of more adaptive immune function, has been observed in older dementia caregivers who reported greater social support. The stress ameliorating effect of social support would also seem to extend to relatively young caregivers. For example, socially supported caregivers of children with additional complex needs have reported reduced levels of psychological distress (Bromley et al., 2004; Ekas et al., 2010; Ergh et al., 2002; Ha et al., 2011; Hastings & Johnson, 2001; Herman & Thompson, 1995; Kayfitz et al., 2010; Pottie et al., 2009; Pozo et al., in press; Smith et al., 2011; Weiss, 2002; White & Hastings, 2004), as well as better physical health status (Lin et al., 2009; Raina et al., 2005; Sawyer et al., 2010). However, fewer studies have assessed whether greater social support might play a protective role with respect to disruptions of disease relevant physiological processes in relatively young caregivers. In a recent study, greater social support buffered caregivers of children with additional complex needs against
stress induced alterations in cardiovascular functioning (Gallagher & Whiteley, 2012a). Study three aimed to extend this relatively small research base by examining whether greater social support might mitigate psychological distress, and in so doing, protect autism/ADHD caregivers against perturbations in the HPA axis and immune response.

Data revealed a significant inverse relationship between perceived levels of stress and caregivers’ scores on the appraisal, belonging and self esteem subscales of the ISEL. Anxiety and depression scores, as well as subjective reports of ill health were also diminished in caregivers who scored higher on all individual ISEL subscales. Physiological support for the buffering effect of social support also emerged, such that caregivers who scored higher on the belonging subscale of the ISEL displayed more adaptive HPA functioning, as indexed by a steeper CAR. However, no statistically meaningful relationships emerged between social support scores and concentrations of the inflammatory marker, CRP.

As a buffer between caring for a child with autism/ADHD and increased reports of ill health, findings reported here indicate that greater social support might mitigate psychological distress, and in so doing, play a protective role with respect to maladaptive changes in basal stress hormone activity. In view of these findings, interventions that enhance social connectivity might be advantageous for improving the psychophysiological well being of relatively young caregivers of children with additional complex needs such as autism/ADHD.
5.1 Methods

5.1.1 Participants

Participants were recruited according to strict criteria which included: (a) aged between 18-60 years, (b) providing care for at least one child (aged between 3-19 years) with clinically verified autism or ADHD, who was (c) living at home on a full time basis, (d) not providing care for another individual with any form of chronic illness, (e) not pregnant, breast feeding, taking any steroidal and/or hormone replacement medication, (f) not experiencing any form of chronic illness, and (g) not working night shift. Participants who satisfied these criteria were invited to provide informed consent following an ethically approved protocol endorsed by the Department of Psychology Ethics Committee. Participants were recruited from regional support groups, special schools and charities across the North East of England, and were recompensed £10.00.

Of the 56 participants recruited in total, two withdrew citing time constraints and 12 failed to return any saliva samples, or collect sufficient volume of saliva and/or blood for biochemical assay. Adherence checks identified one participant with a delay of more than 10 minutes between waking and collection of the waking cortisol sample on both sampling days. This participant was also excluded. Data were also excluded for two participants who collected their waking and 30 minute post waking samples more than 10 minutes outside of the requested 30 minute time interval on both sampling days. Therefore, statistical analysis was conducted on a final sample of 39 participants. Means and standard deviations for demographic, behavioural and biomedical characteristics of the sample are presented in Table 5.1.
Table 5.1

**Demographic, Behavioural and Biomedical Characteristics of the Sample**

<table>
<thead>
<tr>
<th><strong>Gender (%)</strong></th>
</tr>
</thead>
</table>
| Mothers        | 90  
| Fathers        | 10  
| **Age, years (mean ± SD)** | 44.6 (7.1)  
| **Weight, lbs (mean ± SD)** | 159.6 (41.6)  
| **BMI (mean ± SD)** | 26.3 (6.1)  
| **Number of children (%)** |  
| 1 child        | 23  
| 2-3 children   | 64  
| > 3 children   | 13  
| **Marital status** |  
| Partnered      | 77  
| Not partnered  | 23  
| **Annual income (%)** |  
| < £20,000      | 26  
| £20,000-29,999 | 20  
| £30,000-49,999 | 34  
| > £50,000      | 10  
| **Nicotine (%)** |  
| Smoker         | 13  
| Non smoker     | 87  
| **Alcohol consumption, units per week (%)** |  
| < 5 units      | 62  
| 5-10 units     | 26  
| > 10 units     | 12  
| **Exercise, per week (%)** |  
| < 3 occasions  | 49  
| 3 or more occasions | 51  
| **Oral contraceptives (%)** |  
| Yes            | 12  
| No             | 88  
| **Antidepressant medications (%)** |  
| Yes            | 26  
| No             | 64

5.1.2 Potential Confounds

Variables that might provide alternative explanations for relationships between social support and psychophysiological outcomes were assessed. These included: gender, age, weight, BMI, alcohol consumption, use of nicotine, frequency
of exercise, oral contraceptives and antidepressant medications, number of children and annual household income.

5.1.3 Psychological Outcomes

Psychological stress was quantified using the 10 item PSS. The HADS was used to measure anxiety and depression, and social support was quantified using the ISEL. Incidences of 54 common health problems were measured using the PILL (see Chapter Two for full details of all psychosocial measures). Internal consistency in the sample was high (all Cronbach’s alphas > 0.86).

5.1.4 Physiological Outcomes

Basal organisation of the HPA axis was assessed by measuring salivary cortisol at waking, 30 minutes post waking, 1200h and 2200h on two consecutive weekdays. Indices of the CAR, diurnal cortisol slope and mean cortisol output were calculated (see Chapter Two for full details of the saliva collection protocol and checks on adherence). Venous blood was collected and the obtained plasma assessed for concentrations of the proinflammatory biomarker, CRP. Fasting blood was taken from all participants between 10am-12pm to control for diurnal variations. Plasma samples frozen at -80° were thawed and CRP quantified in duplicate using high sensitivity ELISA (CRP, Kalon Biological Ltd, Guildford, UK). This assay has a minimum detection threshold of 0.2 mg/l. Mean intra and inter-assay coefficients were 5.2% and 12.0% respectively. CRP data was positively skewed (D (39) = 0.27, p = < 0.01), and as such, was log_{10} transformed to normalise distributions.
5.1.5 Additional Measures of Protocol Adherence

Two participants reported delays of more than 10 minutes between waking and collection of the waking cortisol sample on one sampling day. In addition, three participants failed to complete adherence checks, and a further three participants failed to provide sufficient volume of saliva for biochemical assay on one sampling day. In all cases, protocol adherent data from the remaining sampling day was taken forward for statistical analysis (Mommersteeg et al., 2006; Sjögren et al., 2006).

5.2 Results

5.2.1 Potential Confounds

Bivariate correlation yielded a significant positive relationship between the diurnal cortisol slope ($\beta$) and both use of nicotine ($r = 0.58, n = 39, p < 0.01$), and number of children ($r = 0.45, n = 39, p < 0.01$). CAR magnitude was also positively related with use of nicotine ($r = 0.42, n = 39, p < 0.01$). A significant positive correlation between number of children and mean diurnal cortisol output was also observed ($r = 0.37, n = 39, p = 0.02$). To avoid spurious relationships between social support scores and psychophysiological outcomes of interest, subsequent analysis adjusted for these variables. Table 5.2 presents means and standard deviations for psychophysiological outcomes in the sample.
Table 5.2

Means and Standard Deviations for Psychophysiological Outcomes and Incidences of Common Health Problems in the Sample

<table>
<thead>
<tr>
<th>ISEL subscales (mean, SD)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Appraisal</td>
<td>21.7 (6.8)</td>
</tr>
<tr>
<td>Belonging</td>
<td>20.1 (6.6)</td>
</tr>
<tr>
<td>Tangible</td>
<td>21.6 (6.6)</td>
</tr>
<tr>
<td>Self esteem</td>
<td>17.4 (4.9)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Psychological outcomes (mean, SD)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Perceived stress</td>
<td>22.7 (6.3)</td>
</tr>
<tr>
<td>Anxiety</td>
<td>9.9 (4.5)</td>
</tr>
<tr>
<td>Depression</td>
<td>9.0 (4.3)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Incidences of common health problems (mean, SD)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>17.5 (11.1)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Cortisol indices (mean, SD)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Waking (nmol/L)</td>
<td>14.9 (6.0)</td>
</tr>
<tr>
<td>30 minutes post waking (nmol/L)</td>
<td>20.4 (8.4)</td>
</tr>
<tr>
<td>1200h (nmol/L)</td>
<td>6.9 (5.8)</td>
</tr>
<tr>
<td>2200h (nmol/L)</td>
<td>2.9 (5.8)</td>
</tr>
<tr>
<td>CAR (nmol/L)</td>
<td>5.5 (8.6)</td>
</tr>
<tr>
<td>Diurnal cortisol slope (β)</td>
<td>-0.85 (0.3)</td>
</tr>
<tr>
<td>Mean diurnal cortisol output (nmol/L)</td>
<td>11.3 (4.7)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Proinflammatory biomarkers</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>CRP (mg/L)</td>
<td>1.3 (1.8)</td>
</tr>
</tbody>
</table>

CAR, cortisol awakening response; CRP, C reactive protein; ISEL, Interpersonal Support Evaluation Checklist; mg/L, microgram per litre; nmol/L, nanomoles per litre

5.2.2 Psychological Outcomes and Social Support

Depression scores were significantly diminished in caregivers who scored higher on all individual ISEL subscales (all *p* < 0.03). Data further revealed a significant inverse relationship between perceived levels of stress and scores on the appraisal (*r* = -0.39, *n* = 39, *p* = 0.02) and self esteem subscales of the ISEL (*r* = -0.50, *n* = 39, *p* < 0.01). In addition, caregivers’ levels of anxiety were negatively correlated with scores on all individual ISEL subscales (all *p* < 0.04). Caregivers with higher scores on the appraisal (*r* = -0.53, *n* = 44, *p* < 0.01), belonging (*r* = -0.46, *n* = 44, *p* < 0.01) self esteem (*r* = -0.36, *n* = 44, *p* = 0.02) and tangible (*r* = -0.50, *n* =
44, $p < 0.01$) subscales of the ISEL also reported significantly fewer incidences of common health problems.

Table 5.3

*Correlation Coefficients for Scores on ISEL Subscales and Psychophysiological Outcomes*

<table>
<thead>
<tr>
<th>Psychological distress</th>
<th>Appraisal</th>
<th>Belonging</th>
<th>Tangible</th>
<th>Self esteem</th>
</tr>
</thead>
<tbody>
<tr>
<td>Perceived stress</td>
<td>-0.39*</td>
<td>-0.25</td>
<td>-0.22</td>
<td>-0.50**</td>
</tr>
<tr>
<td>Anxiety</td>
<td>-0.46**</td>
<td>-0.36*</td>
<td>-0.33*</td>
<td>-0.58**</td>
</tr>
<tr>
<td>Depression</td>
<td>-0.54**</td>
<td>-0.55**</td>
<td>-0.37*</td>
<td>-0.59**</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Incidences of common health problems</th>
<th>Appraisal</th>
<th>Belonging</th>
<th>Tangible</th>
<th>Self esteem</th>
</tr>
</thead>
<tbody>
<tr>
<td>CAR</td>
<td>0.25</td>
<td>0.32*</td>
<td>0.20</td>
<td>0.27</td>
</tr>
<tr>
<td>Diurnal cortisol slope</td>
<td>0.15</td>
<td>0.12</td>
<td>0.27</td>
<td>0.40</td>
</tr>
<tr>
<td>Mean diurnal cortisol output</td>
<td>-0.07</td>
<td>-0.07</td>
<td>0.03</td>
<td>-0.09</td>
</tr>
</tbody>
</table>

In proinflammatory biomarkers

| CRP                               | 0.31      | 0.16      | 0.04     | 0.16        |

$p = < 0.05$

$p = < 0.01$

5.2.3 *Physiological Outcomes and Social Support*

Partial correlation, adjusting for nicotine, yielded a significant positive association between CAR magnitude and scores on the belonging subscale of the ISEL ($r = 0.32$, $n = 39$, $p = 0.05$). However, the relationship between social support scores and other HPA indices, diurnal cortisol slope (all $ps > 0.10$) and mean diurnal cortisol output (all $ps > 0.49$) failed to reach statistical significance. Data also yielded no statistically meaningful relationships between scores on individual ISEL subscales and concentrations of the inflammatory marker, CRP (all $ps > 0.06$). Table 5.3
presents correlation coefficients between scores on individual ISEL subscales and psychophysiological outcomes.

5.3 Discussion

Caregivers’ mean scores for anxiety and depression satisfied clinical criterion for borderline mood disorder (Snaith, 2003), and as such, reinforces the importance for identifying buffers that alleviate caregiver related stress. Findings reported here converge with other studies that have demonstrated reduced psychological morbidity (Bozo et al., 2010; Ekas et al., 2010; Hastings et al., 2002; Kayfitz et al., 2010; Pottie et al., 2009) and fewer incidences of ill health (Lin et al., 2009; Sawyer et al., 2010) in socially supported caregivers of children with additional complex needs. Physiological support for the stress ameliorating effect of social support also emerged, such that greater CAR magnitude, which is indicative of more adaptive HPA function (Fries et al., 2009; Powell & Schlotz, 2013), was observed in caregivers who reported higher belonging based support. It was recently demonstrated that greater social support might protect caregivers of children with additional complex needs such as autism against higher daytime systolic blood pressure, one well known risk factor for increased disease risk, especially in relation to cardiovascular disorders (Gallagher & Whiteley, 2012a). Findings reported here indicate that greater social support is associated with reduced levels of psychological distress and might protect autism/ADHD caregivers against perturbations in other disease relevant physiological processes such as the HPA axis.

Indeed, cortisol is responsible for physiological processes such as cardiovascular arousal, energy mobilisation, and promotes a state of enhanced arousal (Sapolsky, 2000; Schulz, Kirschbaum, Pruessner, & Hellhammer, 1998). As
such, researchers have argued the CAR is adaptive, equipping the organism with the physiological resources needed to meet anticipated challenges of the day (Clow et al., 2004, 2012; Saxbe, 2008). For example, greater CAR magnitude has been observed in those experiencing increased work overload (Steptoe, 2000), challenging and novel situations (Brant et al., 2010), and chronic worry (Wust, Federenko, Hellhammer, & Kirschbaum, 2000). A steeper CAR has also been observed on the day of an anticipated social evaluative performance stressor (Rohleder et al., 2007) and on more demanding week (i.e., work) days, relative to more leisurely weekends (Kunz Ebrecht et al., 2004; Scholtz et al., 2004). In addition, chronically stressed caregivers whose dependents presented more problem behaviours also displayed greater CAR magnitude (De Vugt et al., 2005). Moreover, waking individuals in the middle of the night (Dettenborn et al., 2007), or after a short snooze in the early evening (Federenko et al., 2004) failed to elicit a cortisol rise. Therefore, anticipation of upcoming demands appears to be of major relevance for the CAR, a notion that has been posited in a recent review (Fries et al., 2009) and experimentally modelled paper (Powell & Schlotz, 2013).

However, attenuation of the CAR has been observed in the context of chronically stressful life events such as lower SES (Ranjit, Young, & Kaplan, 2005), higher general life stress (O’Connor et al., 2009), and in relatively young caregivers of children with additional complex needs such as autism (Seltzer et al., 2010). Moreover, reduced bioavailability of cortisol has been implicated in the aetiologies of stress related disorders such as burnout (Heim et al., 2000), chronic fatigue syndrome (van Houdenhove et al., 2009), as well as other functional somatic syndromes (Tak, Bakker, Slaets, & Rosmalen, 2010). These data highlight the
potential clinical relevance of a blunted CAR in autism/ADHD caregivers with diminished social support.

In conclusion, as a buffer between the stress of the caregiver experience and increased subjective reports of ill health, greater social support might reduce psychological distress, and in so doing, protect autism/ADHD caregivers against maladaptive changes in basal stress hormone activity. In view of these findings, interventions that enhance social connectivity might be effective for improving the psychophysiological well-being of relatively young caregivers of children with additional complex needs such as autism/ADHD.

5.4 Concluding Remarks

Findings from study two indicated the stress of caring for a child with autism/ADHD exacts a considerable psychophysiological toll on the carer. However, data from study three suggests that socially supported caregivers might be relatively protected against these harmful effects. Indeed, reduced psychological morbidity and a steeper CAR, which is indicative of more adaptive HPA activity, was observed in caregivers who reported greater social support. An inverse relationship between social support scores and caregivers’ reported incidences of ill health was also observed.

In view of these findings, the development and delivery of interventions that enhance social connectivity might be advantageous for improving the psychophysiological well-being of autism/ADHD caregivers. However, as an intervention strategy, increasing social connectivity in caregivers of children with additional complex needs is difficult to initiate. Indeed, despite good provision, support related interventions such as community support groups are often
inaccessible to caregivers due to logistical/practical challenges such as difficulties arranging alternative and reliable supervision, as well as anxieties over leaving the child in someone else’s care (Bank et al., 2006; Yantzi et al., 2006). Similarly, other potentially effective interventions such as cognitive behavioural therapy (Gallagher-Thomson & Steffen, 1994; Losada-Baltar et al., 2004; Ostwald et al., 1999; Wysocki et al., 2000; Vedhara et al., 2003), formal aid from respite services (Garcés et al., 2010; Zarit et al., 1999) and psychoeducational skills training (Gallagher-Thompson et al., 2000; Sörensen et al., 2002) are often expensive, time consuming and usually require participation outside the home. Informal caregivers, however, experience serious financial hardship (Donelan et al., 2002; Kogan et al., 2008), and owing to the sheer volume of work involved with the caregiver role, often have little available free time (Edworthy, 2005; Fletcher, et al., 2012). Therefore, there would appear to be a case for developing interventions that obviate some of the financial and logistical/practical challenges faced by informal caregivers.

Written emotional disclosure (WED) is a simple, time and cost effective intervention that can be run in participants’ homes at any time of day (Wetherell, et al., 2006), and as such, might be especially well suited for informal caregivers. Transforming thoughts and feelings about stressful life events into language has been linked with adaptive changes in a variety of health outcomes in healthy (Hemenover et al., 2003; Petrie et al., 1995) and medical populations (Baikie & Wilhelm, 2005; Smyth et al., 1999), and in the context of chronically stressful life events such as PTSD (Sloan et al., 2005) and breast cancer (Stanton et al., 2002). However, WED in its traditional format has yielded little in the way of health benefits for informal caregivers (Barton & Jackson, 2008; Mackenzie et al., 2007; Schwartz & Drotar, 2004). More recently, positive psychological adjustments such as lower anxiety and
depression scores were reported in caregivers lower in alexithymia (and therefore, better able to express their emotions in words), who focussed their writing on positive life experiences (Ashley et al., 2011). In addition, caregivers who reported finding more positive consequences (i.e., benefits) amidst the stress of the caregiver experience scored lower with respect to feelings of anxiety and depression (Kim et al., 2007; McCausland & Pakenham, 2003), and displayed steeper diurnal cortisol slopes, which is indicative of healthier HPA functioning (Moskowitz et al., 2006). Interestingly, adaptational outcomes such as lower levels of anxiety and fewer somatic health complaints have also been reported in chronically stressed breast cancer patients who wrote for 20 minutes on three consecutive days about the benefits associated with their disease experience (Henry et al., 2010).

To date, only one study has applied a positive writing intervention in the context of chronic caregiver stress (Ashley et al., 2011). Consequently, study four examined the feasibility and efficacy of an at-home written benefit finding intervention for coping with the stress of caring for a child with autism/ADHD.
6. Study Four

6.0 Chapter Overview

In study three, socially supported autism/ADHD caregivers reported diminished psychological morbidity, fewer incidences of ill health and displayed greater CAR magnitude, which is indicative of more adaptive HPA function. In view of these findings, it was concluded that increasing caregivers’ social connectivity might be advantageous for improving their psychophysiological well being.

However, financial (Kogan et al., 2008; Parish & Cloud, 2006) and logistical/practical challenges such as difficulties arranging alternate and reliable supervision (Bank et al., 2006; Yantzi et al., 2006), often prevent caregivers accessing community support groups, as well as other potentially effective interventions such as cognitive behavioural therapy (Gallagher-Thompson & Steffen, 1994; Losada-Baltar et al., 2004; Vedhara et al., 2003; Wysocki et al., 2000), psychoeducational skills training (Gallagher-Thompson et al., 2001; Sörensen et al., 2002) and formal aid from respite services (Garcés et al., 2010; Zarit et al., 1999).

Written emotional disclosure (WED) on the other hand is a cheap and time effective intervention that can run in participants’ homes, and at any time of day (Wetherell et al., 2005). Writing expressively about stressful/traumatic events for as little as 20 minutes on three-four consecutive days has been linked with beneficial effects on a broad catalogue of health outcomes in healthy (Smyth, 1998) and clinical populations (Averill et al., 2013; Baikie & Wilhelm, 2005; Frisina et al., 2004), and in the context of chronically stressful life events such as breast cancer (Henry et al., 2010; Rosenberg et al., 2002) and bereavement (Kovac & Range, 2000), but not informal caregiving (Barton & Jackson, 2008; Mackenzie et al., 2007; Schwartz & Drotar, 2004). More recently, reduced anxiety and depression
scores were observed in caregivers lower in alexithymia (and therefore, better able to express their emotions linguistically), who focussed their writing on positive life events (Ashley et al., 2011). Converging with these findings, caregivers often find positive consequences (i.e., benefits) amidst the stress of the caregiver experience such as greater appreciation for life/loved ones, and better priority of life goals (Kayfitz et al., 2010; Samios et al., 2009). Interestingly, writing expressively about the benefits associated with chronically stressful life events has been linked with adaptational outcomes such as reduced psychological morbidity and better physical health status (Henry et al., 2010; Stanton et al., 2002).

In view of these findings, it is perhaps surprising that only one study to date has applied a positive written disclosure intervention in the context of chronic caregiver stress (Ashley et al., 2011). To address the paucity of research in this area, study four investigated the feasibility and efficacy of an at-home written benefit finding intervention on psychophysiological outcomes in caregivers of children with autism/ADHD.

Data indicated that at-home written benefit finding and ambulatory assessments of psychophysiological functioning is feasible for autism/ADHD caregivers. Indeed, all caregivers completed the writing task on three consecutive days as instructed, and with little or no interruptions. Moreover, rate of attrition was comparable with other studies that have assessed the efficacy of expressive writing in chronically stressed caregivers (Barton & Jackson, 2008; Mackenzie et al., 2007; Schwartz & Drotar, 2004). However, several caregivers assigned to the benefit finding condition reported difficulties completing their essays, a finding that might partially explain the null effect of the writing task on psychophysiological outcomes.
6.1 Methods

6.1.1 Participants

A sample of 41 autism/ADHD caregivers was recruited for a study investigating: ‘the feasibility and efficacy of expressive writing for coping with the stress of caring for a child with autism/ADHD’. Participants were recruited from caregiver support groups, special schools and charities across the North East of England, and according to the following criteria: (a) aged 18-60 years, (b) providing care for at least one child (aged between 3-19 years) with a clinical diagnosis of autism or ADHD, who was (c) living at home on a full time basis, (d) not providing care for another individual with chronic illness, (e) not pregnant, breast feeding, taking any steroidal and/or hormone replacement medication, (f) not experiencing chronic illness, and (g) not working night shift. Participants who satisfied these criteria were invited to provide informed consent following an ethically approved protocol endorsed by the Department of Psychology Ethics Committee.

Of the 41 participants recruited in total, one withdrew citing time commitments. Four participants who failed to provide complete psychophysiological outcome data, and one participant who failed to return any writing booklets were also excluded. One participant was excluded based on cortisol concentrations greater than 75 nmol/L (Kunz Ebrecht et al., 2004), as were five participants with CRP concentrations greater than 10mg/L (Fuligni et al., 2009; Taylor et al., 2006). The intervention profile is displayed in Figure 6.1.
Figure 6.1 Intervention profile. CRP, C-reactive protein; mg/L, micrograms per litre; nmol/L, nanomoles per litre; WBNR, Writing booklets not returned; PNA, Protocol non adherent.

In addition, adherence checks identified one participant with a delay of more than 10 minutes between waking and collection of the waking cortisol sample on both sampling days at baseline, and at one month post writing. Data for this participant was also excluded. Therefore, statistical analysis was conducted on a final sample of 28 participants. Table 6.1 presents demographic, behavioural and biomedical data for the sample by writing condition.
Table 6.1

*Means and Standard Deviations for Demographic, Behavioural and Biomedical Characteristics by Writing Condition*

<table>
<thead>
<tr>
<th></th>
<th>Benefit Finding (n = 12)</th>
<th>Control condition (n = 16)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Gender</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>Female</td>
<td>12</td>
<td>13</td>
</tr>
<tr>
<td><strong>Age</strong></td>
<td>46.0 (4.4)</td>
<td>42.8 (4.2)</td>
</tr>
<tr>
<td><strong>Weight (lbs)</strong></td>
<td>145.0 (15.6)</td>
<td>159.6 (29.1)</td>
</tr>
<tr>
<td><strong>BMI</strong></td>
<td>25.3 (3.1)</td>
<td>25.8 (3.9)</td>
</tr>
<tr>
<td><strong>Education</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Secondary school</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>College</td>
<td>4</td>
<td>6</td>
</tr>
<tr>
<td>University undergraduate</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>University postgraduate</td>
<td>6</td>
<td>5</td>
</tr>
<tr>
<td><strong>Alcohol (units per week)</strong></td>
<td>3.6 (4.1)</td>
<td>6.0 (8.0)</td>
</tr>
<tr>
<td><strong>Exercise (occasions per week)</strong></td>
<td>4.3 (2.0)</td>
<td>2.4 (2.2)</td>
</tr>
<tr>
<td><strong>Number of children</strong></td>
<td>2.3 (1.0)</td>
<td>1.9 (0.6)</td>
</tr>
<tr>
<td><strong>Child problem behaviours</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Conduct</td>
<td>3.1 (1.6)</td>
<td>5.1 (2.8)</td>
</tr>
<tr>
<td>Hyperactivity</td>
<td>8.2 (2.3)</td>
<td>8.7 (1.4)</td>
</tr>
<tr>
<td>Emotional</td>
<td>5.2 (2.5)</td>
<td>5.4 (2.0)</td>
</tr>
<tr>
<td>Peer</td>
<td>5.7 (2.1)</td>
<td>5.6 (1.9)</td>
</tr>
<tr>
<td><strong>Years caregiving</strong></td>
<td>5.4 (4.1)</td>
<td>4.3 (3.3)</td>
</tr>
<tr>
<td><strong>Journal or diary</strong></td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>No</td>
<td>11</td>
<td>16</td>
</tr>
<tr>
<td><strong>Social media</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>10</td>
<td>11</td>
</tr>
<tr>
<td>No</td>
<td>2</td>
<td>5</td>
</tr>
<tr>
<td><strong>Support group member</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>6</td>
<td>7</td>
</tr>
<tr>
<td>No</td>
<td>6</td>
<td>9</td>
</tr>
<tr>
<td><strong>Marital status</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Partnered</td>
<td>9</td>
<td>13</td>
</tr>
<tr>
<td>Not partnered</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td><strong>Menstrual cycle</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phase one</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>Phase two</td>
<td>5</td>
<td>3</td>
</tr>
<tr>
<td><strong>Oral contraceptives</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>No</td>
<td>12</td>
<td>12</td>
</tr>
</tbody>
</table>
Table 6.1 (continued)

<table>
<thead>
<tr>
<th></th>
<th>Benefit Finding (n = 12)</th>
<th>Control Condition (n = 16)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Antidepressants</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>No</td>
<td>10</td>
<td>12</td>
</tr>
<tr>
<td><strong>Statins</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>No</td>
<td>11</td>
<td>14</td>
</tr>
</tbody>
</table>

6.1.2 Writing Interventions

Participants were assigned to the benefit finding or control condition using computer generated random numbers. In all cases, participants were unaware that they had been assigned to one of two writing conditions. For the traditional WED paradigm, participants are asked to write for 20 minutes on three-four consecutive weekdays about stressful/traumatic life events, or if randomised to the control condition, about emotionally neutral topics such as time management tasks (i.e., daily activities). In a modification to the traditional WED paradigm, participants here were directed to write for 20 minutes on three consecutive weekdays about the benefits of caring for a child with autism/ADHD, or if assigned to the control condition, to describe emotionally neutral landscape images in detail (Ashley et al., 2011). All participants wrote on their assigned topic at home. Indeed, completion of the disclosure task at home has been recommended in both clinical and chronically stressed populations, who might find it difficult to participate in lab based tasks (Vedhara et al., 2007). Participants were asked to write, without regard for spelling, sentence structure or grammar, at the same time on three consecutive weekdays, and to date all essays. It was made clear that writing should be completed in a quiet place (e.g., bedroom), thus minimising the possibility for interruptions (e.g., telephone,
doorbell, other people in the home). Participants were asked to return all essays to the research team for inspection and were assured of their confidentiality. It was made clear to all participants that essay feedback would not be provided.

6.1.3 Benefit Finding

Instructions for the task were informed by previous studies that also incorporated a benefit finding condition (Henry et al., 2010; Stanton et al., 2002). Participants were encouraged to really let go and to explore their innermost thoughts and feelings about the benefits of caring for a child with autism/ADHD. A definition and examples of benefit finding were included with written instructions to support participants’ completion of the task. That is, participants were told that people often find positive consequences (i.e., benefits) amidst stressful life events such as improved social relationships, greater sensitivity to family issues and overall appreciation for life/loved ones. It was suggested that participants write about positive consequences with respect to (a) their lives, (b) their goals and (c) their relationships. Participants were free to write about the same benefits each day or explore different benefits on different writing days. In accordance with previous studies, participants were instructed to repeat previous points, or try including more detail if they had run out of things to write (Ashley et al., 2011; Pennebaker et al., 1988).

6.1.4 Control Condition

For the traditional WED paradigm, participants assigned to the control condition are asked to write about emotionally neutral topics such as daily activities (Pennebaker et al., 1988; Vedhara et al., 2007; Wetherell et al., 2005). However,
caregivers’ daily activities are principally organised around the care recipient, and therefore, are not emotionally neutral. For the present study, participants randomised to the control condition were asked to describe landscape images in detail, a control task used in other disclosure studies involving informal caregivers (Ashley et al., 2011). Participants were asked to describe in detail one picture per day without including any personal information or opinions on the images. It was suggested that participants write about (a) items included in the image (e.g., trees, buildings, water etc), (b) item colours (e.g. very pale blue, lemon yellow, scarlet red etc), (c) number of items (e.g., three trees, nine flowers, one house etc) and (c) item positions (e.g. in the foreground, in front of something, to the left-hand edge of the picture, in-between other items etc). In accordance with previous studies, participants were instructed to repeat previous points, or try including more detail if they had run out of things to write (Ashley et al., 2011; Henry et al., 2010; Pennebaker & Chung, 2012).

6.1.5 Measures
6.1.5.1 Manipulation Checks

All essays were read by the principal investigator and dates inspected to verify adherence with the writing protocol (i.e., that writing had been completed on three consecutive weekdays). As an objective measure of essay content, participants’ essays were transcribed into a database and analysed using Linguistic Inquiry and Word Count (LIWC), a text analysis software that quantifies participants’ use of different categories of words (Pennebaker, Chung, Ireland, Gonzales, & Booth, 2007). To verify that participants had written about qualitatively different topics, percentages of positive (e.g., happy) and negative (e.g., upset) emotion, and cognitive insight words (e.g., think) were compared between writing conditions.
Consistent with task instructions, it was expected that essays of participants randomised to the benefit finding condition would contain a greater percentage of affective and cognitive insight words. Following each writing session, participants were also asked to rate the extent to which their disclosed material was (a) meaningful, (b) personal, (c) emotionally revealing, and (d) how much they had wanted to disclose the content of their essays to others. Responses were scored on a five point Likert scale (0 = not at all to 5 = a great deal). Congruent with task instructions, it was expected that participants in the benefit finding condition would rate their essays as being more meaningful, personal, and revealing of emotions. It was also expected the desire to share the disclosed material with others would be greater in the benefit finding condition. Participants’ physical engagement with the task was assessed using a short questionnaire. That is, immediately following each writing session, participants were asked to report (a) any interruptions to their writing (e.g., telephone, doorbell etc) and (b) how long the interruption lasted (1 = less than five minutes, 2 = more than five minutes). Researchers have recommended that data are excluded for participants who report interruptions to their writing that last more than five minutes (Zakowski, Ramati, Morton, Johnson, & Flanigan, 2004).

6.1.5.2 Potential Confounds

To determine whether demographic, behavioural or biomedical factors might be acting as confounds, all participants completed a short questionnaire at baseline that assessed: gender, age, weight, BMI, level of education, use of nicotine, alcohol consumption, frequency of exercise, use of oral contraceptives, antidepressant medications and statins, phase of the menstrual cycle, number of children, child problem behaviours, years spent caregiving and support group membership.
Participants also answered questions on normative opportunities for written disclosure such as keeping a journal/diary, and use of social media.

6.1.5.3 Psychosocial Outcomes

The PSS, HADS, PILL and ISEL were completed at baseline, one month and three months post writing (see Chapter Two for full details of all psychosocial measures). Psychosocial measures achieved excellent internal consistency at baseline, and at both follow up points (all Cronbach’s alphas > 0.85).

6.1.5.4 Endocrine Outcomes

Basal organisation of the HPA axis was assessed via salivary biomarkers at baseline, one month and three months post writing (see Chapter Two for full details of the saliva collection protocol and checks on adherence). Repeated measures ANOVAs were used to explore possible within person differences in cortisol data per individual sample point. Huynh Feldt correction was applied whenever sphericity was violated. Results revealed a significant main effect of time, reflecting normal diurnal variation of cortisol at baseline ($F (2.3, 51.3) = 96.6, p < 0.01, \eta_p^2 = 0.82$), one month ($F (1.8, 32.4) = 83.7, p < 0.01, \eta_p^2 = 0.82$), and at three months post writing ($F (3.0, 69.0) = 53.4, p < 0.01, \eta_p^2 = 0.70$). However, data yielded no between day differences (all $ps > 0.24$, all $\eta_p^2 < 0.08$) or day x time interaction effect at baseline, or either post writing follow up (all $ps > 0.42$, all $\eta_p^2 < 0.04$). In addition, no between day differences were observed with respect to CAR magnitude (all $ps > 0.81$, all $\eta_p^2 < 0.01$) or diurnal cortisol slope (all $ps > 0.51$, all $\eta_p^2 < 0.02$) at baseline, one month, or at three months post writing. In view of these findings, cortisol values...
for each sampling point were averaged across the two sampling days to obtain more reliable indices of basal HPA activity (O’Connor et al., 2009).

6.1.5.5 Immune Outcomes

Venous blood was collected and the obtained plasma assessed for concentrations of the proinflammatory biomarker, CRP at baseline, and at three months post writing. Fasting blood was taken from all participants between 10am-12pm to control for diurnal variations. CRP was assessed in duplicate by high sensitivity ELISA (CRP, Kalon Biological Ltd, Guildford, UK). This assay has a minimum detection threshold of 0.2 mg/l. CRP data was positively skewed at baseline ($D(28) = 0.22, p < 0.01$) and three months post writing ($D(28) = 0.23, p < 0.01$), and therefore, was log$_{10}$ transformed to normalise distributions.

6.1.6 Procedures

Consenting participants were provided a baseline pack to take home containing self report measures of psychological distress (PSS, HADS), social support (ISEL) and incidences of ill health (PILL). Baseline packs also contained materials for the ambulatory collection of salivary cortisol, as well as detailed written instructions that emphasised the importance of timely collection. Participants were asked to return baseline packs in prepaid addressed envelopes within two weeks. On receipt, materials to complete the writing intervention were sent to participants by post. Each writing pack contained: (a) three writing booklets, (b) an instruction sheet, (c) manipulation check questionnaires, and (d) a prepaid addressed envelope. All participants were contacted by telephone to confirm receipt of the writing materials and were given the opportunity to ask any questions regarding task completion. It
was at this point participants confirmed on which three days (and at what time) they would be available to write on their assigned topic. Research has indicated that regular contact with participants is critical for successful transfer of the WED paradigm to a field setting (Wetherell et al., 2005). For the present study, all participants were contacted by telephone 10 minutes before each writing session as a reminder to complete the writing task, and to reinforce the importance of strict protocol adherence. Calls were also made 10 minutes following each writing session to verify that participants had completed all manipulation check questionnaires and dated their essays. Manipulation checks and writing booklets were sealed in a prepaid addressed envelope and returned to the research team by post. Participants were followed up at one and three months post writing, at which point, assessments of psychological well being, social support, ill health and basal HPA activity were repeated. Venous blood was collected from participants at baseline and three months post writing to assess changes in CRP concentrations in response to the writing task. All participants were recompensed £40.00.

6.2 Results

6.2.1 Feasibility

Rate of attrition was comparable with other studies (with similar size samples) that have assessed the palliative effects of expressive writing in the context of chronic caregiver stress (Barton & Jackson, 2008; Mackenzie et al., 2007; Schwartz & Drotar, 2004). Researchers have recommended that, as an indicator of poor physical engagement with the task, data are excluded for participants who report interruptions to their writing that last more than five minutes (Zakowski et al., 2004). For the present study, however, all participants completed their essays with
minor (i.e., less than five minutes) or no interruptions. Manipulation checks further verified that all participants wrote on their assigned topic on three consecutive weekdays as instructed. These data support the feasibility of implementing an at-home written benefit finding intervention in chronically stressed caregivers of children with autism/ADHD.

However, four of 12 (33%) caregivers assigned to the benefit finding condition commented on difficulties writing about the benefits associated with the caregiver experience. Indeed, essays of these four caregivers focussed predominately on the negative/stressful aspects associated with caregiving. For example, one caregiver wrote: “I honestly don’t believe there are any positive aspects to having a child with autism”. Another wrote: “I find it very hard to think about the positive effects, I really think I would be lying if I did this”. Other comments included: “It is really difficult to say how I am affected positively by my son’s condition” and, “It is so hard to escape the negative thoughts”.

6.2.2 Baseline Comparisons and Equivalence of Conditions

A series of chi square ($\chi^2$) and one way ANOVAs revealed no group disparities with respect to gender, age, weight, BMI, level of education, use of nicotine, alcohol consumption, use of oral contraceptives and statins, phase of the menstrual cycle, number of children, years caregiving, support group membership and opportunities for normative written disclosure (all $ps > 0.06$, all $\eta_p^2 < 0.16$). However, participants assigned to the benefit finding condition exercised more frequently ($F (1, 26) = 5.80, p = 0.02 \eta_p^2 = 0.18$) and reported fewer problems with care recipients’ conduct behaviours ($F (1, 26) = 0.71, p = 0.04, \eta_p^2 = 0.17$). Therefore, these variables were statistically controlled in all subsequent analysis.
A series of one way ANOVAs revealed that writing conditions were statistically indistinguishable on psychological well being (all $ps > 0.19$, all $\eta^2_p < 0.07$), social support (all $ps > 0.53$, all $\eta^2_p < 0.02$) and subjective reports of ill health ($F(1, 26) = 0.33, p = 0.57, \eta^2_p = 0.01$) at baseline. Mixed ANOVA was applied to assess differences in cortisol data per individual sample point between writing conditions at baseline. Results were corrected with Huynh-Feldt whenever sphericity was violated. The within subject factor was time (waking, 30 minutes post waking, 1200h, and 2200h) and between subject factor was writing condition (benefit finding vs. control). Findings revealed a significant main effect of time ($F(2.59, 67.33) = 125.82, p < 0.01, \eta^2_p = 0.83$) reflecting normal variation in diurnal cortisol secretion. However, no group differences emerged with respect to mean diurnal cortisol output ($F(1, 26) = 0.10, p = 0.76, \eta^2_p < 0.01$) or pattern of change across the day ($F(2.59, 67.33) = 0.43, p = 0.70, \eta^2_p = 0.02$). One way ANOVA further verified that writing conditions were comparable with respect to CAR magnitude ($F(1, 26) = 0.36, p = 0.56, \eta^2_p < 0.01$) and diurnal cortisol slope ($F(1, 26) = 0.27, p = 0.61, \eta^2_p < 0.01$) at baseline. However, concentrations of the inflammatory marker, CRP were significantly elevated in the control condition at baseline ($F(1, 26) = 5.87, p = 0.02, \eta^2_p = 0.18$).

6.2.3 Manipulation Checks

Essays of participants assigned to the benefit finding condition were significantly more revealing of emotions ($F(1, 26) = 52.46, p < 0.01, \eta^2_p = 0.67$), more personal ($F(1, 26) = 71.80, p < 0.01, \eta^2_p = 0.73$) and meaningful ($F(1, 26) = 49.27, p < 0.01, \eta^2_p = 0.66$) compared with controls. The desire to share information disclosed in the essays was also significantly greater in the benefit finding condition.
Data from LIWC indicated that essays of participants assigned to the benefit finding condition included a higher percentage of positive \((F (1, 26) = 50.43, p < 0.01, \eta_p^2 = 0.66)\) and negative emotion \((F (1, 26) = 83.13, p < 0.01, \eta_p^2 = 0.76)\), and cognitive insight words \((F (1, 26) = 144.28, p < 0.01, \eta_p^2 = 0.85)\). Table 6.2 displays means and standard deviations for manipulation checks by writing condition. These results are in accordance with those generally found in written disclosure studies and indicate the writing intervention was effective (Ashley et al., 2011; Pennebaker et al., 2007; Vedhara et al., 2007).

Table 6.2

<table>
<thead>
<tr>
<th>LIWC</th>
<th>Benefit Finding (n = 12)</th>
<th>Control Condition (n = 16)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive emotion words (%)</td>
<td>4.2 (1.1)</td>
<td>1.3 (1.0)</td>
</tr>
<tr>
<td>Negative emotion words (%)</td>
<td>1.5 (0.5)</td>
<td>0.2 (0.2)</td>
</tr>
<tr>
<td>Cognitive insight words (%)</td>
<td>7.8 (1.7)</td>
<td>1.9 (0.8)</td>
</tr>
<tr>
<td>State VAS</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Emotionally revealing</td>
<td>3.2 (0.4)</td>
<td>1.6 (0.7)</td>
</tr>
<tr>
<td>Personal</td>
<td>3.7 (0.6)</td>
<td>1.6 (0.7)</td>
</tr>
<tr>
<td>Meaningful</td>
<td>3.5 (0.6)</td>
<td>1.7 (0.7)</td>
</tr>
<tr>
<td>Told others</td>
<td>2.4 (1.1)</td>
<td>1.3 (0.4)</td>
</tr>
</tbody>
</table>

LIWC, Linguistic Inquiry and Word Count; VAS, Visual Analogue Scales

6.2.4 Psychological Outcomes

Two way mixed ANCOVAs (controlling for exercise and child conduct behaviours) were used to assess differences in psychological well being, social support and common health problems over time between writing conditions (Ashley et al., 2011; O’Connor & Ashley, 2008). Violated assumptions were corrected using Huynh-Feldt. The within subject factor was time (baseline, one month and three
months post writing) and between group factor was writing condition (benefit finding vs. control). Means and standard deviations for psychophysiological outcomes by writing condition are displayed in Table 6.3.

Data yielded no main effect of time (all $p > 0.43$, all $\eta^2_p < 0.08$), condition (all $p > 0.51$, all $\eta^2_p < 0.02$), or time x condition interaction effect (all $p > 0.71$, all $\eta^2_p < 0.02$) with respect to perceived stress and anxiety scores. There was a trend toward a reduction in depression scores across the post writing period ($F(2, 42) = 2.93, p = 0.06, \eta^2_p = 0.12$); however, writing conditions were statistically indistinguishable as evidenced by no main effect of condition ($F(1, 21) = 11.13, p = 0.99, \eta^2_p < 0.01$) or time x condition interaction ($F(2, 42) = 1.39, p = 0.26, \eta^2_p = 0.06$). Thus, there was no effect of writing condition on psychological well being from baseline. Data also yielded no main effect of time (all $p > 0.07$, all $\eta^2_p < 0.13$), condition (all $p > 0.32$, all $\eta^2_p < 0.12$) or interaction effect (all $p > 0.10$, all $\eta^2_p < 0.11$) with respect to scores on all individual ISEL subscales, reflecting no effect of writing condition on social support across the follow up period. There was a trend toward a post writing reduction in reported incidences of ill health ($F(2, 42) = 3.03, p = 0.06, \eta^2_p = 0.13$); however, writing conditions could not be statistically differentiated, as evidenced by no main effect of condition ($F(1, 21) = 9.85, p = 0.25, \eta^2_p = 0.06$) or time x condition interaction effect ($F(2, 42) = 0.26, p = 0.77, \eta^2_p = 0.12$). Thus, there was no effect of writing condition on subjective reports of ill health across the follow up period.
Table 6.3

*Means and Standard Deviations for Psychophysiological Outcomes by Writing Condition*

<table>
<thead>
<tr>
<th></th>
<th>Benefit Finding (n = 12)</th>
<th>Control Condition (n = 16)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Perceived Stress</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>19.8 (6.7)</td>
<td>22.2 (6.3)</td>
</tr>
<tr>
<td>One month</td>
<td>17.8 (4.5)</td>
<td>21.6 (4.5)</td>
</tr>
<tr>
<td>Three months</td>
<td>18.3 (9.1)</td>
<td>22.8 (6.4)</td>
</tr>
<tr>
<td><strong>HADS (Anxiety)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>8.3 (3.7)</td>
<td>9.6 (3.7)</td>
</tr>
<tr>
<td>One month</td>
<td>8.3 (3.7)</td>
<td>8.9 (4.0)</td>
</tr>
<tr>
<td>Three months</td>
<td>7.2 (3.9)</td>
<td>9.6 (4.3)</td>
</tr>
<tr>
<td><strong>HADS (Depression)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>8.2 (4.4)</td>
<td>8.5 (3.2)</td>
</tr>
<tr>
<td>One month</td>
<td>8.0 (5.2)</td>
<td>9.1 (3.5)</td>
</tr>
<tr>
<td>Three months</td>
<td>6.4 (3.7)</td>
<td>8.9 (4.0)</td>
</tr>
<tr>
<td><strong>Common health problems</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>17.4 (12.3)</td>
<td>21.1 (7.7)</td>
</tr>
<tr>
<td>One month</td>
<td>15.3 (10.9)</td>
<td>17.2 (7.6)</td>
</tr>
<tr>
<td>Three months</td>
<td>14.5 (10.9)</td>
<td>17.4 (8.5)</td>
</tr>
<tr>
<td><strong>ISEL (appraisal)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>19.6 (7.0)</td>
<td>22.4 (10.2)</td>
</tr>
<tr>
<td>One month</td>
<td>21.5 (7.4)</td>
<td>21.4 (7.4)</td>
</tr>
<tr>
<td>Three months</td>
<td>20.3 (6.7)</td>
<td>21.8 (6.5)</td>
</tr>
<tr>
<td><strong>ISEL (tangible)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>18.9 (9.0)</td>
<td>21.6 (5.5)</td>
</tr>
<tr>
<td>One month</td>
<td>19.8 (8.8)</td>
<td>22.1 (6.5)</td>
</tr>
<tr>
<td>Three months</td>
<td>18.7 (7.9)</td>
<td>20.9 (5.7)</td>
</tr>
<tr>
<td><strong>ISEL (belonging)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>19.2 (8.0)</td>
<td>20.3 (4.7)</td>
</tr>
<tr>
<td>One month</td>
<td>20.6 (8.6)</td>
<td>20.2 (5.0)</td>
</tr>
<tr>
<td>Three months</td>
<td>20.3 (6.5)</td>
<td>20.4 (6.0)</td>
</tr>
<tr>
<td><strong>ISEL (self esteem)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>16.6 (6.0)</td>
<td>16.8 (4.4)</td>
</tr>
<tr>
<td>One month</td>
<td>17.9 (6.2)</td>
<td>16.5 (3.9)</td>
</tr>
<tr>
<td>Three months</td>
<td>17.2 (5.7)</td>
<td>16.8 (5.2)</td>
</tr>
<tr>
<td><strong>CAR (nmol/L)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>3.3 (4.0)</td>
<td>3.6 (6.1)</td>
</tr>
<tr>
<td>One month</td>
<td>2.5 (2.5)</td>
<td>2.2 (4.2)</td>
</tr>
<tr>
<td>Three months</td>
<td>3.7 (9.5)</td>
<td>5.9 (5.5)</td>
</tr>
</tbody>
</table>
6.2.5 Endocrine Outcomes

Two way mixed ANCOVAs were used to assess differences in CAR magnitude, diurnal cortisol slope and mean diurnal cortisol output over time between writing conditions. Results were corrected with Huynh-Feldt whenever sphericity was violated. Data revealed no main effect of time (all ps > 0.13, all $\eta_p^2 < 0.09$), condition (all ps > 0.55, all $\eta_p^2 < 0.02$) or time x condition interaction effect (all ps > 0.25, all $\eta_p^2 < 0.06$) with respect to all HPA indices. Thus, there was no effect of writing condition on basal stress hormone activity across the follow up period. Mixed ANCOVAs were also used to assess differences in cortisol data per individual sample point between writing conditions at the one month and three month post writing follow up. Data revealed a significant main effect of time (all ps < 0.01, all $\eta_p^2 > 0.27$), reflecting the typical descending pattern of cortisol secretion across the day; however, no main effect of condition (all ps > 0.36, all $\eta_p^2 < 0.04$) or condition x time interaction effect (all ps > 0.35, all $\eta_p^2 < 0.05$) was apparent. That is, writing

Table 6.3 (continued)

<table>
<thead>
<tr>
<th></th>
<th>Benefit Finding (n = 12)</th>
<th>Control Condition (n = 16)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Diurnal cortisol slope ($\beta$)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>-0.87 (0.2)</td>
<td>-0.85 (0.3)</td>
</tr>
<tr>
<td>One month</td>
<td>-0.96 (0.1)</td>
<td>-0.93 (0.1)</td>
</tr>
<tr>
<td>Three months</td>
<td>-0.60 (0.5)</td>
<td>-0.80 (0.2)</td>
</tr>
<tr>
<td><strong>Mean diurnal cortisol output (nmol/L)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>7.2 (1.6)</td>
<td>7.5 (2.9)</td>
</tr>
<tr>
<td>One month</td>
<td>6.7 (3.0)</td>
<td>6.6 (2.0)</td>
</tr>
<tr>
<td>Three months</td>
<td>7.5 (3.3)</td>
<td>8.0 (2.9)</td>
</tr>
<tr>
<td><strong>CRP concentrations (mg/L)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>1.1 (1.0)</td>
<td>1.7 (2.0)</td>
</tr>
<tr>
<td>Three months</td>
<td>0.9 (0.7)</td>
<td>2.1 (2.2)</td>
</tr>
</tbody>
</table>

CAR, cortisol awakening response; CRP, C-reactive protein; HADS, Hospital Anxiety and Depression Scale; ISEL, Interpersonal Support Evaluation Checklist; mg/L, milligrams per litre; nmol/L, nanomoles per litre
conditions could not be differentiated on diurnal cortisol secretion patterns at one month or three months post writing.

6. 2.6 Immune Outcomes

CRP concentrations were significantly elevated in the control condition at baseline \((F(1, 26) = 5.87, p = 0.02, \eta^2_p = 0.18)\). However, after statistically adjusting for baseline differences, writing conditions could not be differentiated on CRP concentrations at three months post writing \((F(1, 20) = 0.22, p = 0.64, \eta^2_p = 0.01)\).

6.3 Discussion

Study four assessed the feasibility and efficacy of an at-home written benefit finding intervention, including ambulatory assessments of psychophysiological functioning, in caregivers of children with autism/ADHD. Data indicated that all participants completed their essays on three consecutive weekdays as instructed, and experienced few, if any interruptions. Level of attrition was also comparable with other studies (with similar size samples) that have applied an expressive writing intervention in the context of chronic caregiver stress (Barton & Jackson, 2008; Mackenzie et al., 2007; Schwartz & Drotar, 2004). These data indicate that at-home written benefit finding is a feasible intervention for relatively young caregivers of children with additional complex needs such as autism/ADHD.

In terms of its efficacy, writing conditions could not be statistically differentiated on psychological well being, social support or incidences of ill health across the follow up period. However, it was noteworthy that caregivers assigned to the benefit finding condition, whose mean anxiety and depression scores placed them in the range of borderline clinical mood disorder at baseline, scored within the
normative range at three months post writing (Snaith, 2003). That is, while statistically indistinguishable from controls on psychological well being from baseline, caregivers who wrote about the benefits of caring for a child with autism/ADHD reported a clinically meaningful reduction in anxiety and depression scores post writing. These data indicate that written benefit finding, a time and cost effective intervention that can be carried out at home, might be effective for alleviating caregiver related stress. With regard basal stress hormone activity, writing conditions could not be differentiated on CAR magnitude, diurnal cortisol slope or mean diurnal cortisol output across the follow up period. In addition, no support emerged for the adaptive effects of written benefit finding on immune function, such that writing conditions were comparable on concentrations of the inflammatory marker, CRP at three months post writing.

WED has been linked with beneficial effects on a broad catalogue of health outcomes in chronically stressed individuals such as breast cancer patients (Henry et al., 2010; Rosenberg et al., 2002) and bereaved individuals (Low, Stanton, & Danoff-Burg, 2006), but not informal caregivers (Barton & Jackson, 2008; Mackenzie et al., 2007; Schwartz & Drotar, 2007). However, in a modification to the traditional WED paradigm, positive psychological adjustments such as lower anxiety and depression scores were recently observed in caregivers lower in alexithymia (and therefore, better able to express their feelings linguistically), who wrote expressively about positive life experiences (Ashley et al., 2011). Neatly dovetailing with these findings, caregivers often find positive consequences (i.e., benefits) amidst the stress of the caregiver experience such as a greater appreciation for life/loved ones and better priority of life goals (Kayfitz et al., 2010; Samios et al., 2009). Interestingly, writing about the benefits associated with chronically stressful life events has been linked
with adaptational outcomes such as improved psychological well being and better somatic health status (Henry, et al., 2010; Stanton et al., 2002). Thus, findings reported here are at odds with relevant previous studies that have demonstrated health benefits in caregivers (Ashley et al., 2011), and in other chronically stressed individuals (Henry et al., 2010; Stanton et al., 2002) who wrote expressively about positive life experiences, or the positive consequences associated with stressful life events.

However, research has indicated that variables related to individual differences might moderate disclosure efficacy (O’Connor & Ashley, 2008). For example, in a study of informal caregivers, Ashley et al (2011) found that alexithymia, a condition characterised by difficulties identifying and expressing emotions linguistically, moderated the effect of positive written disclosure on psychological outcomes. That is, an interaction effect was observed, such that reduced anxiety and depression scores were reported by caregivers lower in alexithymia who wrote expressively about positive life experiences. More recently, research indicated that individuals more ambivalent about emotional expression (i.e., those who found it difficult, or didn’t want to express their emotions) were more likely to benefit from expressive writing (Averill et al., 2013). Accordingly, failure to assess the moderating effect of alexithymia, or other individual difference variables related to emotional openness (e.g., ambivalence about emotional expression) on the efficacy of the written benefit finding task represents a significant limitation of the present study. Indeed, writing expressively about the benefits associated with the caregiver experience might have produced positive psychophysiological adjustments for caregivers who were more (Ashley et al., 2011) or less (Averill et al., 2013) able to identify and express their feelings linguistically.
Four of 12 of caregivers assigned to the benefit finding condition reported difficulties completing their essays, a finding which might partially explain the null effect of the writing task on psychophysiological outcomes. Indeed, finding difficulties writing about the benefits associated with stressful life events is not a new concept. For example, chronically stressed breast cancer patients reported similar problems when asked to write about the benefits associated with their disease experience (Henry et al., 2010). Inspection of these four caregivers’ essays revealed that much of the disclosed information focussed on the negative/stressful aspects of the caregiver experience. However, writing about stressful life events (Schwartz & Drotar, 2004), or exclusively about the stress of the caregiver experience (Barton & Jackson, 2008; Mackenzie et al., 2007) has yielded little in the way of health benefits for informal carers. In fact, researchers recently suggested that writing about stressful life events triggers perseverative cognition (i.e., ruminative stress related thinking), and as such, might seriously undermine the effectiveness of stress related disclosure (O’Connor, Walker, Hendrickx, Talbot, & Schaefer, 2013). Despite some difficulties completing the writing task, caregivers assigned to the benefit finding condition, most of who satisfied criterion for clinical anxiety and depression at baseline, scored within the normative range at three months post writing (Snaith, 2003). That several caregivers commented on difficulties completing their essays highlights the potential importance for identifying techniques that improve caregivers’ engagement with the benefit finding task.

As one possible technique, video interaction guidance (VIG) captures carer-recipient interactions on film and has been used to help caregivers of children with cerebral palsy identify communicative strengths within the carer-recipient dyad (Fukkink, 2008; Kennedy & Sked, 2008; Wadnerkar, Pirinen, Haines-Bazrafshan,
Rodgers, & James, 2010). Accordingly, video feedback techniques might also be useful for helping autism/ADHD caregivers identify and reflect on benefits associated with the caregiver experience. Future studies, therefore, might examine whether VIG, either as a standalone technique or primer to the written benefit finding task, is effective for improving the psychophysiological well being of autism/ADHD caregivers.

The findings of this study must be discussed in the context of its limitations. The sample size, while comparable in magnitude to other studies that have assessed the efficacy of expressive writing in caregivers (Barton & Jackson, 2008; Mackenzie et al., 2007; Schwartz & Drotar, 2004), might have precluded the observation of statistically meaningful effects. Indeed, data revealed a notable trend toward a significant main effect of time, reflecting reduced depression scores and fewer incidences of ill health post writing. These data hint at a non specific positive effect of writing per se, such that improvements in psychological well being and self reported health were apparent irrespective of writing condition. Indeed, by temporarily distracting caregivers from their responsibilities (Ashley et al., 2011), or by eliciting positive emotions (Wetherell et al., 2005), writing about landscape images might produce adaptive changes in health related outcomes. Given the observed feasibility of the protocol, the null effects reported here should be considered preliminary until substantiated with a larger sample.

In conclusion, data indicated that at-home written benefit finding is a feasible intervention for relatively young autism/ADHD caregivers. That is, all caregivers completed the writing task on three consecutive days as instructed and experienced only minimal interruptions. However, no support emerged for the adaptive effects of written benefit finding on psychophysiological outcomes which included:
psychological well being, social support, reported ill health, diurnal cortisol secretion patterns and proinflammatory activity. While statistically indistinguishable from controls on anxiety and depression scores post writing, caregivers assigned to the benefit finding condition, one third of who reported difficulties completing the writing task, no longer satisfied criterion for borderline clinical mood disorder at the three month follow up (Snaith, 2003). These data underscore the potential importance for identifying techniques that improve caregivers’ engagement with the benefit finding task. It was recommended that VIG might help autism/ADHD caregivers identify and reflect on benefits associated with the caregiver experience, and as such, might be an advantageous technique for improving their psychophysiological well being.

6.4 Concluding Remarks

Feasibility data indicated that all caregivers completed the writing task on three consecutive days and with only minimal interruptions. Moreover, rate of attrition was comparable with other studies (with similar size samples) that have applied an expressive writing intervention in the context of chronic caregiver stress (Barton & Jackson, 2008; Mackenzie et al., 2007). In terms of its efficacy, data revealed no effects of writing about the benefits of caring for a child with autism/ADHD on any psychophysiological outcomes of interest. Findings reported here are odds with relevant previous studies that have observed health benefits in caregivers, and in other chronically stressed individuals who wrote about positive life experiences (Ashley et al., 2011), or the positive consequences (i.e., benefits) associated with chronically stressful life experiences (Henry et al., 2010; Stanton et al., 2002).
However, research has indicated that individual difference variables related to emotional openness such as alexithymia (Ashley et al., 2011; O’Connor & Ashley, 2008) and ambivalence over emotional expression (Averill et al., 2013), might moderate disclosure efficacy. Indeed, writing expressively about the benefits associated with the caregiver experience, a task that requires caregivers to express their innermost feelings and emotions, is likely to be moderated by characteristics of the individual. Failure to assess the moderating effect of individual difference variables related to emotional openness represents a major limitation of the present study.

Data revealed that caregivers in the benefit finding condition, whose mean anxiety and depression scores satisfied criterion for borderline clinical mood disorder at baseline, scored within the normative range at three months post writing (Snaith, 2003). That is, despite several reports of difficulties completing the writing task, caregivers who wrote about the benefits of caring for a child with autism/ADHD reported a clinically meaningful reduction in psychological distress post writing. These data underscore the importance for identifying techniques that increase caregivers’ engagement with the written benefit finding task. It was recommended that VIG, which captures natural carer-recipient interactions on film, might help autism/ADHD caregivers identify and reflect on benefits associated with the caregiver experience, and as such, might be effective for improving psychophysiological outcomes.
7. General Discussion

As a programme of work, the thesis set out to investigate the psychophysiological pathways that link chronic stress with increased vulnerability for ill health. As the first in thesis, study one examined whether perceived stress related disparities in common health problems might be mediated by differential patterns of cortisol secretion. Findings indicated that dysregulated HPA activity, widely implicated in the aetiologies of severe pathologic conditions such as aspects of the metabolic syndrome (Rosmond, 2005) and adverse cardiovascular events (Matthews et al., 2006; Seldenrijk et al., 2012), partially mediated the effect of higher perceived levels of stress on greater incidences of the kinds of common health problems that typically affect young, otherwise healthy individuals. Study one also provided a valuable learning experience with respect to the accurate collection of salivary biomarkers, and helped inform important protocol decisions such as the continued use of paper adherence diaries in future experimental studies.

As a logical next step in the programme, study two used the caregiver control model to look more closely at the psychophysiological consequences of chronic stress. In particular, study two considered: (a) whether the physiological impairments apparent in older dementia caregivers might extend to relatively young caregivers of children with autism/ADHD, and (b) whether social support, typically diminished in relatively young caregivers (Fletcher et al., 2012; Gallagher et al., 2010, 2012a, 2012b; Lach et al., 2009; Seltzer et al., 2001) and associated with immune dysfunction (Kiecolt-Glaser et al., 2002, 2009; Lutgendorf et al., 2000), might underlie the negative impact of chronic caregiver stress on immune function. Data confirmed the a priori assumption that autism/ADHD caregivers are more stressed than controls. Indeed, caregivers’ mean anxiety and depression scores satisfied
current criterion for borderline clinical mood disorder, while scores for parents of typically developing children fell within the normative range (Snaith, 2003). Caregivers also reported greater incidences of common health problems and diminished social support. Immune dysfunction, manifested by higher concentrations of the proinflammatory biomarker, CRP was also apparent in the caregiver group. Indeed, caregivers’ mean concentrations of CRP (1.3 mg/L) satisfied clinical criteria for moderate cardiovascular disease risk, compared with low risk in the controls (Pearson et al., 2003; Ridker et al., 2003). However, data revealed that caregiver related disparities in proinflammatory activity were not mediated by variations in social support.

Research has demonstrated that, in the context of caring for a child with additional complex needs, greater social support can mitigate psychological distress (Bozo et al., 2010; Ekas et al., 2010; Gallagher & Whiteley, 2012a; Ha et al., 2011; Pozo et al., in press; Smith et al., 2011; Weiss, 2002; White & Hastings, 2004) and protect against increased reports of negative health (Lin et al., 2009; Raina et al., 2005; Sawyer et al., 2010). However, fewer studies have examined whether greater social support can protect relatively young caregivers against perturbations in disease relevant physiological processes such as the HPA axis and immune response. Data from study three revealed diminished psychological morbidity and fewer incidences of ill health in socially supported caregivers of children with autism/ADHD. To the extent that caregivers reported greater belonging based support, they also displayed greater CAR magnitude, which is indicative of more adaptive HPA function (Fries et al., 2009; Powell & Scholtz, 2013). These findings converge with other recent studies that have demonstrated how greater social support can protect caregivers of children with additional complex needs against perturbations in other disease
relevant physiological parameters such as systolic blood pressure (Gallagher & Whiteley, 2012a). Taken together, these findings indicate that increasing caregivers’ social connectivity might be advantageous for improving their psychophysiological well being. Indeed, quality of life for a child with additional complex needs is contingent on the health and happiness of the care provider (Addington et al., 2003; Burgess et al., 2007; Mockus-Parks & Novielli, 2000; Schor, 2003).

Socially supportive interventions, while potentially effective for coping with chronic caregiver stress (Chien et al., 2010), are often inaccessible to informal caregivers. Indeed, despite good provision, caregivers are often unable to attend community support groups owing to difficulties arranging alternative and reliable supervision, as well as anxieties about leaving the child in someone else’s care (Bank et al., 2006; Edworthy, 2005; Gallagher-Thompson et al., 2006). Similarly, psychotherapeutic interventions such as cognitive behavioural therapy (Gallagher-Thompson & Steffen, 1994; Losada-Baltar et al., 2004; Ostwald et al., 1999; Vedhara et al., 2003; Wysocki et al., 2000), psychoeducational skills training (Sörensen et al., 2002) and formal aid from respite services (Garcés et al., 2010; Zarit et al., 1999) are often expensive, time consuming and usually based outside of the home. However, informal caregivers often experience severe financial hardship (Donelan et al., 2002; Kogan et al., 2008; Parish & Cloud, 2006) and painful social isolation (Yantzi et al., 2006; Wiles, 2003), thus find it difficult to utilise these varied and potentially effective interventions.

Written emotional disclosure (WED) on the other hand is a simple, time and cost effective intervention that can be run in participants’ homes at any time of day (Wetherell et al., 2005). Beneficial effects of WED on psychophysiological outcomes have been widely observed in the context of chronically stressful life
events such as PTSD (Koopman et al., 2006; Sloan et al., 2005; Smyth et al., 1998), bereavement (Kovac & Range, 2000) and breast cancer (Rosenberg et al., 2002), but not informal caregiving (Barton & Jackson, 2008; Mackenzie et al., 2007; Schwartz & Drotar, 2004). However, reduced anxiety and depression scores were recently reported in caregivers lower in alexithymia, and who wrote for 20 minutes on three consecutive days about positive life experiences (Ashley et al., 2011). Converging with these findings, caregivers often report finding positive consequences (i.e., benefits) amidst the stress of the caregiver experience such as greater appreciation for life/loved ones and improved personal relationships (Kayfitz et al., 2010; Markoulakis et al., 2012). Interestingly, writing about the benefits associated with chronically stressful life events has been linked with adaptive changes in a variety of health related outcomes (Henry et al., 2010; Stanton et al., 2002). Data from study four indicated that at-home written benefit finding is a feasible intervention for caregivers of children with autism/ADHD. Indeed, all caregivers completed the writing task on three consecutive weekdays as instructed and experienced only minimal interruptions. In terms of its efficacy, data revealed no statistically meaningful differences between writing conditions on any psychophysiological outcomes across the follow up period. However, it was noteworthy that caregivers assigned to the benefit finding condition, most of who satisfied criterion for borderline clinical mood disorder at baseline, scored within the normative range at three months post writing (Snaith, 2003). These data suggest that written benefit finding might be effective for alleviating caregiver related stress. That one third of caregivers assigned to the benefit finding condition reported difficulties completing the writing task make this finding of even greater importance. It was recommended that video feedback techniques, which have been
previously used to improve communication within the carer-recipient dyad (Wadnerkar et al., 2010), might also be useful for helping caregivers better engage with the benefit finding task.

7.0 Programme Limitations

Studies presented here share several limitations. First, the cross sectional nature of the work precludes drawing inferences about the observed relationship between study variables. Future studies might better delineate causality by using more prospective designs with larger sample numbers.

Throughout the thesis, measurement of the CAR was based on saliva collection at waking and 30 minutes post waking only. Sampling across a more protracted period (e.g., at waking, 15, 30 and 45 minutes post waking) would have provided a more robust measure of the CAR (Pruessner et al., 1997). However, it was reported in a recent meta analysis that estimates of CAR magnitude are typically based on collection of saliva at two time points only, waking and 30 minutes post waking (Adam & Kumari, 2009), as was the case throughout the thesis.

Research has indicated that single day collection can bias aspects of the cortisol pattern such as the CAR to state, rather that trait characteristics (Hellhammer et al., 2007). To gain a more reliable estimate of basal HPA activity, researchers have recommended that saliva be collected at four-six time points (Stalder et al., 2009) on two (or more) consecutive weekdays (Hellhammer et al., 2007; Stalder et al., 2010). For the current programme of work, saliva was collected at four time points (waking, 30 minutes post waking, 1200, and 2200h) on two (studies two, three and four) or three (study one) consecutive weekdays. Sampling across a more protracted period (e.g., waking, 30 minutes post waking, +1 hour, +4 hours, + 9 hours and +14 hours),
as has been the case in previous studies (Miller et al., 2008), might have provided a more robust measure of basal stress hormone activity. However, informal caregivers are a notoriously hard to reach population (Gallagher & Whiteley, 2012a) with little available free time (Edworthy, 2005; Marsh et al., 1998), and as such, it was important for retention/completion rates to keep the saliva collection protocol manageable in size.

Adam & Kumari (2009) indicated that adherence with the saliva collection protocol might be optimised if participants are provided: (a) clear sampling instructions, (b) a precise definition of waking, (c) training to accurately collect saliva, and (d) a simple way to return saliva samples. In all studies of the thesis, steps were taken to encourage participants’ strict adherence with the saliva collection protocol. That is, all participants were provided detailed sampling instructions that emphasised the importance of exact time of sampling, and were trained to accurately collect saliva using the Salivette (Sarstedt, Ltd). Participants were also provided a precise definition of waking, defined as: ‘when your eyes open and you are ready to get up’ (Adam & Kumari, 2009; Cohen et al., 2006), as well as prepaid addressed envelopes to return saliva samples to the research team by post. In addition, all participants were instructed to record waking and collection times as accurately as possible on all sampling days using paper diaries. Electronic monitoring devices such as MEMS track caps (that record the date and time of each sample) or wrist actigraphy (that can differentiate wake and sleep periods through monitoring activity) could have provided a more objective check on timing compliance (Adam & Kumari, 2009; Smyth et al., in press), but can be prohibitively expensive (Adam & Kumari, 2009). Moreover, MEMS track caps often produce missing data, and as a result, might invalidate the reliability of diurnal cortisol measurement (Ailinger et al.,
Subjective measures (i.e., paper diaries) on the other hand are preferred by participants (Kraemer et al., 2006), and according to several recent studies, perform equally well in comparison with more objective, electronic measures for evaluating adherence with the saliva collection protocol (Okun et al., 2010; Seltzer et al., 2009, 2010; Stalder et al., 2009; Wolf et al., 2008). Therefore, self report measures of timing compliance (i.e., paper diaries) were used in all studies throughout the thesis. It should be acknowledged, however, that other procedures controlling for suspected protocol noncompliance might have been applied such as excluding data for participants who displayed no cortisol rise post waking (O’Connor et al., 2009; Thorn et al., 2006).

Findings reported here might be confounded as a consequence of unmeasured lifestyle variables such as quality/duration of sleep. Indeed, an inverse association between sleep quality and psychological distress has been demonstrated in older caregivers of partners with dementia (von Kanel, Dimsdale, & Ancoli-Israel et al., 2006), and in relatively young caregivers of children with additional complex needs such as autism (Gallagher et al., 2010). As such, findings reported here might not be independent of sleep related factors. Similarly, attributes of the care recipient such as child problem behaviours (Gallagher et al., 2008, 2009, 2012b) and intelligence (Baker et al., 2002) have been associated with the psychophysiological well being of the care provider. Accordingly, future studies involving chronically stressed caregivers could usefully employ more thorough assessments of sleep measurement parameters, as well as specific attributes of the care recipient. However, it should be noted that statistically meaningful findings were maintained following adjustment for the most likely demographic (e.g., gender, age, education etc), behavioural (e.g.,
nicotine, alcohol, exercise etc) and biomedical (e.g., BMI, medication etc) confounds.

Autism is a complex genetic condition which might imply that some caregivers share milder versions of the disorder (Le Couteur, Haden, Hammal, & McConachie, 2008; Piven et al., 1997). Deficits related to emotion recognition are a hallmark symptom of autism (Jones et al., 2011), and as such, might have affected caregivers’ ability to engage with the benefit finding intervention, a task that requires participants to interpret and express their innermost thoughts and emotions in words. Therefore, it will be important for future studies to incorporate standardised assessment tools for measuring caregivers’ autistic symptoms and to statistically control for their potential confounding effects.

7.1 Future Directions

In study two of the thesis, immune dysfunction, as evidenced by elevated concentrations of the proinflammatory biomarker, CRP was apparent in autism/ADHD caregivers. According to current clinical guidelines, caregivers mean concentrations of CRP (1.3 mg/L) indicate moderate risk for cardiovascular pathologies, compared with low risk in the control group (Ridker et al., 2003). These findings converge with other studies that have demonstrated elevated systolic blood pressure, one well known biomarker for cardiovascular disease risk, in relatively young caregivers of children with additional complex needs (Gallagher & Whiteley, 2012a; Yamaki, Hsieh, & Heller, 2009). Previous research observed a positive relationship between CRP concentrations and pulse wave velocity, one composite measure of elastic and muscular arterial stiffness (Spence et al., 2008), which has been linked with adverse cardiovascular events such as myocardial infarction and
coronary atherosclerosis (Weber et al., 2005). As such, future studies might assess inflammatory markers such as CRP alongside measures of arterial stiffness to better delineate cardiovascular vulnerability in the context of caring for a child with additional complex needs such as autism/ADHD. Similarly, other studies have used ultrasound techniques to examine the progression rate of intima media thickness, one non invasive marker of arterial wall alteration (in the carotid artery) and early asymptomatic indicator of subclinical atherosclerosis, in relation to cardiovascular risk factors such as stress hormones. Indeed, Eller et al (2005) revealed the four year progression rate of intima media thickness was significantly accelerated in participants with a steeper CAR. Future studies might use more prospective designs to assess the significance of inflammatory agents such as CRP for the progression rate of intima media thickness in chronically stressed caregivers of children with additional complex needs.

Findings from study two also confirmed the a priori assumption that caregivers of children with autism/ADHD were more stressed than controls, such that caregivers’ mean anxiety and depression scores satisfied criterion for borderline clinical mood disorder (Snaith, 2003). Caregivers also displayed a wide range of other negative effects such as diminished social support, greater incidences of common health problems and dysregulated immunity, as indexed by increased markers of inflammation. Perturbations in cognitive health indicators such as memory and attention have been widely evidenced in chronically stressed individuals (Evans & Shamberg, 2009; Lupien, Maheu, Tu, Fiocco, & Schramek, 2007; McEwen & Sapolsky, 1995; Peavy et al., 2009), but have been rarely assessed in the context of chronic caregiver stress. To date, research has indicated that, relative to normative controls, informal caregivers perform poorer on working memory and selective
attention tasks (Mackenzie, Smith, Hasher, Leach, & Behl, 2007). Impaired cognition might have serious practical and clinical implications for autism/ADHD caregivers, who are often responsible for coordinating complex medication schedules and arranging important medical appointments, as well as other tasks of daily functioning (Bank et al., 2006; Edworthy, 2005; Green et al., 2006). As such, future studies might examine the impact of caring for a child with additional complex needs on important cognitive domains such as prospective memory, which refers to the process of remembering to do things at some future point in time (Heffernan, Clark, Bartholomew, Ling, & Stephens, 2010).

Findings from study three indicated that socially supported autism/ADHD caregivers might be protected against the harmful effects associated with the caregiver experience. Indeed, diminished levels of psychological distress, fewer incidences of common health problems and more adaptive HPA activity, as evidenced by a steeper CAR, was observed in caregivers who reported greater social support. These findings might be useful for informing the decisions of health care professionals as they relate to helping relatively young caregivers of children with additional complex needs cope more effectively with caregiver related stress.

Data from study four indicated at-home written benefit finding was a feasible intervention for autism/ADHD caregivers. However, in terms of its efficacy, data yielded no statistically meaningful differences between writing conditions with respect to psychological well being, social support, reported ill health, HPA indices or inflammatory markers across the follow up period. Despite several reports of difficulties completing the writing task, caregivers assigned to the benefit finding condition, most of who met criterion for clinical mood disorder at baseline, scored within the normative range at three months post writing (Snaith, 2003). These data
highlight the importance for identifying techniques that increase caregivers’ engagement with the benefit finding task. Video feedback techniques (e.g., VIG) have been used by caregivers of children with cerebral palsy to identify, reflect and successfully build on communicative strengths that exist within the carer-recipient dyad (Fukkink et al., 2008; Wadnerkar et al., 2010). Accordingly, video feedback might also be useful for helping autism/ADHD caregivers identify and reflect on discrepancies between the way they perceive the caregiver experience (i.e., without benefits) and the positive moments captured on film. Therefore, future studies might assess whether video feedback, either as a standalone technique or primer to the written benefit finding task, might be effective for improving the psychophysiological well being of autism/ADHD caregivers.
8. References


Castle, S., Wilkins, S., Heck, E., Tanzy, K., & Fahey, J. (1995). Depression in caregivers of demented patients is associated with altered immunity: Impaired proliferative capacity, increased CD8⁺, and a decline in lymphocytes with surface signal transduction molecules (CD38⁺) and a cytotoxic marker (CD56⁺ CD8⁺). *Clinical and Experimental Immunology, 101*, 487-493.


support moderates caregiver distress. *Journal of Head Trauma Rehabilitation, 17*(2), 155-174.


von Kanel, R., Dimsdale, J. E., Ancoli-Israel, S., Mills, P. J., Patterson, T. L., McKibbin, C., et al. (2006). Poor sleep is associated with higher plasma proinflammatory cytokine interleukin-6 and procoagulant marker fibrin D-


adrenal responses following inhalation of 35% CO$^2$.

*Psychoneuroendocrinology, 31*, 736-747.


