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Mortensen, Jesper

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Informal caregiving and diurnal patterns of salivary cortisol: the Whitehall II cohort study

Jesper Mortensen, Nadya Dich, Alice Jessie Clark, Cecilia Høst Ramlau-Hansen, Jenny Head, Mika Kivimäki, Meena Kumari, Naja Hulvej Rod

Jesper Mortensen, PhD fellow, Department of Public Health, University of Copenhagen, 1123 Copenhagen, Denmark

Nadya Dich, Assistant Professor, Department of Public Health, University of Copenhagen, 1123 Copenhagen, Denmark

Alice Jessie Clark, Assistant Professor, Department of Public Health, University of Copenhagen, 1123 Copenhagen, Denmark

Cecilia Høst Ramlau-Hansen, Professor, Department of Public Health, Aarhus University, 8000 Aarhus, Denmark

Jenny Head, Professor, Department of Epidemiology and Public Health, University College London, WC1E 7HB London, United Kingdom

Mika Kivimäki, Professor, Department of Epidemiology and Public Health, University College London, WC1E 6BT London, United Kingdom and Clinicum, Faculty of Medicine, FI-00014 University of Helsinki, Helsinki, Finland

Meena Kumari, Professor, Institute for Social and Economic Research, University of Essex, CO4 3SQ, Colchester, United Kingdom

Naja Hulvej Rod, Professor, Department of Public Health, University of Copenhagen, 1123 Copenhagen, Denmark

Correspondence to: Jesper Mortensen, Jemo@sund.ku.dk, +45 353-37918, Section of Social Medicine, Department of Public Health, University of Copenhagen, 1123 Copenhagen, Denmark

Abstract

The objective was to investigate the association between aspects of informal caregiving and diurnal patterns of salivary cortisol, with special attention to the potential moderating effect of gender and work status. The study population was composed of 3,727 men and women from the British Whitehall II study. Aspects of caregiving included the relationship of caregiver to recipient, weekly hours of caregiving, and length of caregiving. Salivary cortisol was measured six times during a weekday. Diurnal cortisol profiles were assessed using the cortisol awakening response (CAR) and diurnal cortisol slopes. Results showed that men, but not women, providing informal care had a blunted CAR compared with non-caregivers ($P_{Interaction}$=0.03). Furthermore, there was a dose-response relationship such that more weekly hours of informal care was associated with a more blunted CAR for men (P_{trend}=0.03). The blunted CAR for men was especially pronounced in short-term caregivers those in paid work. In women, the steepest cortisol slope was seen among those in paid work who provided informal care ($P_{Interaction}$=0.01). In conclusion, cortisol profiles differ between male and female informal caregivers. Compared to non-caregivers, male caregivers had a blunted CAR which has previously been associated with chronic stress and burnout, while female caregivers did not. The CAR association among men was especially pronounced for those providing many weekly hours of care, short-term caregivers, and caregivers in paid work.

Keywords: Informal caregiving; Cortisol; work life; cohort study

Introduction

Caregiving for disabled and elderly people entails a large burden for the formal health care services. Informal caregiving, which is defined as unpaid care for a sick, disabled, or elderly family member or other closely related person (USLegal, 2017) saves the health care services great expenses annually (AARP - Public policy Institute, 2015). It is estimated that 34% of the population in 20 European countries provide any type of informal care for a family member, and 8% provide intensive care categorized as ≥11 hours of care a week (Verbakel et al., 2017). A growing population of elderly (WHO, 2012) will most likely also result in a growing population of people living with disabilities, and it is expected that the formal health care services will be under increasing pressure in the coming years (Beesley, 2006). Consequently, informal care is expected to play an important role in relieving the formal health care system of the caregiving burden (Bremer et al., 2017).

Informal caregiving may be associated with positive feelings, such as sense of personal accomplishment and gratification, an increase of family cohesion, and a sense of personal growth (Yu et al., 2017). However, informal caregiving may also be perceived as stressful and burdensome for the caregiver (Alfaro et al., 2013; Hunt GG, Reinhard S, Greene R, Whiting GC, Feinberg LF, 2015; Wolff, Spillman, Freedman, Kasper, 2016). In a large study among informal caregivers, it was found that 46% experienced emotional stress, 32% experienced physical strain, 18% experienced economical strain, and 22% felt that their health had gotten worse as a result of caregiving (Hunt GG, Reinhard S, Greene R, Whiting GC, Feinberg LF, 2015).

Informal caregiving is more common among women compared to men (Beesley, 2006; Hunt GG, Reinhard S, Greene R, Whiting GC, Feinberg LF, 2015; Verbakel et al., 2017), although gender differences in informal caregiving have become less evident in recent years (Beesley, 2006). Some studies have shown that women who provide informal care in general take part in heavier caregiving tasks than men, and more often take the role as primary caregiver (Beesley, 2006; Navaie-Waliser et al., 2002; Neal et

al., 1997; Verbakel et al., 2017). Furthermore, it has been suggested that sex affects HPA responsiveness to psychosocial stress (Kajantie and Phillips, 2006). Based on this, women providing informal care may be more prone to chronic stress compared with their male counterparts, although little research is available to confirm or refute this hypothesis.

A potential health consequence of chronic stress is dysregulation of the cardiovascular system caused by a cascade of neural and physiological responses including hyper- or hypo-activation of the hypothalamic-pituitary-adrenal (HPA) axis (Dragoş and Tănăsescu, 2010). HPA activity stimulates secretion of the stress hormone cortisol by the adrenal glands (Adam, Hawkley, Kudielka, Cacioppo, 2006). Cortisol levels have a natural circadian rhythm, with a high level upon awakening and a rapid increase during the first 30-45 minutes from awakening, called the cortisol awakening response (CAR) (Adam and Kumari, 2009; Stalder, Kirschbaum, Kudielka, Adam, Pruessner, Wüst, Dockray, Smyth, Evans, Hellhammer, Miller, Wetherell, Lupien, Clow, 2015). Following the awakening response, cortisol levels decline during the day with a nadir around midnight (Adam and Kumari, 2009). The CAR and cortisol slope are components of the diurnal cortisol profile, which have been suggested to characterize response to acute and chronic stressors (Adam and Kumari, 2009). An increased CAR is suspected to be a marker of acute psychological stress, while a blunted CAR may be a marker of chronic stress, depression, and burnout (Leggett, Zarit, Kim, Almeida, Klein, 2015; Miller, Chen, Zhou, 2007). Accordingly, steep cortisol slopes suggest a more rapid decline in cortisol levels throughout the day, whereas cortisol slopes with values closer to zero reflect flatter diurnal rhythms, indicating that the bedtime cortisol level is heightened or that the person may be in poor health status with difficulties unwinding before nighttime (Adam and Kumari, 2009).

According to a recent systematic review, providing informal caregiving for a family member with dementia is associated with higher cortisol levels throughout the day, although the findings are not completely consistent (Allen, Curran, Duggan, Cryan, Chorcoráin, Dinan, Molloy, Kearney, Clarke, 2017). Some of the studies found a higher cortisol level at awakening for caregivers and blunted CAR (de Vugt,

Jolles, van Osch, Stevens, Aalten, Lousberg, Verhey, 2006; Fonareva, Amen, Zajdel, Ellingson, Oken, 2011) while other studies reported flattened diurnal cortisol slopes (Allen, Curran, Duggan, Cryan, Chorcoráin, Dinan, Molloy, Kearney, Clarke, 2017). A limitation in those studies is that they did not investigate specific aspects of informal caregiving, such as the relationship with the care recipient, weekly hours of caregiving as a measure of intensity, and duration of caregiving, which may all affect the physiological stress response imposed by informal caregiving (Allen, Curran, Duggan, Cryan, Chorcoráin, Dinan, Molloy, Kearney, Clarke, 2017). For example, it has been suggested that caregiving for a spouse may be more detrimental for health compared to caregiving for a parent (Lee, Colditz, Berkman, Kawachi, 2003) due to more weekly hours of caregiving (AARP - Public policy Institute, 2015). In line with this, many weekly hours of caregiving may also be associated with providing care for a family member in the household, which is a considerable emotional burden (AARP - Public policy Institute, 2015). In addition, providing long-term caregiving may be more detrimental for the caregiver's health because of the ongoing burden as opposed to short-term caregiving, (Capistrant, Moon, Glymour, 2012; Capistrant, Moon, Glymour, 2013). This raises the hypothesis that caregiving for a spouse, providing caregiving for many weekly hours, and long-term caregiving may be associated with particularly blunted CAR and flattened cortisol slopes, as markers of chronic stress (Adam and Kumari, 2009; Leggett, Zarit, Kim, Almeida, Klein, 2015; Miller, Chen, Zhou, 2007).

Simultaneously providing informal care and being active in the labor market may constitute a double burden (Goode, 1960; Longacre, Valdmanis, Handorf, Fang, 2016). Previous analyses of data from the British Whitehall II study have shown that the CAR was greater on workdays compared to weekends, suggesting that work-life is associated with acute stressors (Kunz-Ebrecht, Kirschbaum, Marmot, Steptoe, 2004). However, no previous studies with the statistical power to address effect modification, have investigated whether having paid work as an additional source of stress would increase the adverse effect of informal caregiving on diurnal cortisol levels. Thus for informal caregivers, we hypothesize that the long-term consequence of combining caregiving responsibilities with paid work may make some individuals more prone to be chronically stressed (Goode, 1960; Longacre, Valdmanis, Handorf, Fang, 2016), resulting

in blunted CAR and flattened cortisol slopes (Adam and Kumari, 2009; Leggett, Zarit, Kim, Almeida, Klein, 2015; Miller, Chen, Zhou, 2007).

The objective of the present study was to investigate the association between informal caregiving and diurnal patterns of salivary cortisol, with special focus on the nature of the relationship with the care recipient, weekly hours of providing caregiving, and duration of caregiving, for men and women separately. We examined whether this association was more pronounced for caregivers in paid work compared to those who have retired or otherwise not in the labor market. ~~Investigating this, we used data from the Whitehall II cohort study, which provides detailed data on salivary cortisol in a large population of adults with a working history in the civil service. The use of the large Whitehall II data allows for the examination of intersections of carer versus non-carer, taking the importance of gender and employment into consideration. This has only been feasible to a lesser extent in previous smaller studies (Allen, Curran, Duggan, Cryan, Chorcoráin, Dinan, Molloy, Kearney, Clarke, 2017).~~

Methods

Study participants came from the Whitehall II study, which was established in 1985 and included 10,308 British civil servants from 20 London-based departments (Marmot and Brunner, 2005). We applied data from phase 9 (2007-2009) due to the availability of detailed data on caregiving and cortisol. A total of 6,761 people (response rate 84.5%) participated in this wave. From the 5,963 people, who agreed to participate in the cortisol collection, 4,128 participants had valid cortisol measures (Appendix A: Flowchart). Excluding those with missing data on caregiving information (n=263) and participants with missing data information on covariates (n=138), the study population encompassed 3,727 participants.

Informal caregiving

People were asked if they provided regular care for any of the following in a self-administered questionnaire: a) Children, b) Grandchildren, c) Disabled or ill partner/spouse, d) Disabled or ill parent, e) Other disabled or ill relative, f) Disabled or ill friend. Participants with affirmative response to question c, d, e or f constituted the group of informal caregivers, since caregiving for children and grandchildren is not included in the definition of informal caregiving, unless they are chronically sick or disabled (USLegal, 2017).

In addition, participants were asked how many hours per week they provided care for each of these persons. In line with previous research, we classified participants into three groups depending on the hours of weekly caregiving: 1-8 hours, 9-20 hours, 20-100 hours, and >100 hours weekly (Lee et al., 2003). Investigating duration of caregiving, long-term caregivers were categorized as those, who answered that they provided care for an aged or disabled relative in the preceding phase (2-3 years earlier) and with affirmative response to questions c, d or e, at phase 9. Short-term caregivers were categorized as those, who provided informal caregiving at phase 9, but not in the preceding phase. Category f (care for a disabled or ill friend) was not included in the analyses of duration of caregiving, since the question in the previous phase concerns caregiving for relatives only.

Cortisol

Participants were requested to provide six saliva samples in salivettes over the course of a normal weekday at waking, 30 minutes from awakening, 2.5 hours from awakening, 8 hours from awakening, 12 hours from awakening, and at bedtime. Participants were instructed not to brush teeth or eat or drink anything for 15 min before sample collection. An instruction booklet was used to record information on the day of sampling including date of collection, wake time, and time each sample was taken. Participants were requested to send samples back by post. On receipt of samples, they were stored at -80° Celsius until they were sent to laboratory in dry ice conditions for subsequent analysis. Salivettes were centrifuged at 3,000 revolutions per minute for 5 minutes, resulting in a clear supernatant of low viscosity. Salivary cortisol

levels were measured using a commercial immunoassay with chemiluminescence detection (CLIA; IBLHamburg, Hamburg, Germany). The lower concentration limit of this assay is 0.44 nmol/L; intra- and inter assay coefficients of variance were below 8%. Any sample over 50 nmol/L was reanalyzed (Badrick, Kirschbaum, Kumari, 2007).

Covariates

Confounders were selected based on current knowledge of factors which may influence caregiving and diurnal cortisol levels (Adam and Kumari, 2009), using Directed Acyclic Graphs (Appendix B)(Greenland, Pearl, Robins, 1999). Analyses were adjusted for age, ethnicity, married/cohabiting, annual household income (<20,000£, 20,000-50,000£, >=50,000£) (Goren, Gilloteau, Lees, DiBonaventura, 2014), employment status (in main effect analyses on caregiving), and wakeup time (Adam and Kumari, 2009). Based on results from a study (Burton, Chen, Conti, Pransky, Edington, 2004) and in line with our previous studies on informal caregiving (Mortensen, Clark, Lange, Andersen, Goldberg, Ramlau-Hansen, Head, Kivimäki, Madsen, Leineweber, Lund, Rugulies, Zins, Westerlund, Rod 2016; Mortensen, Dich, Lange, Alexanderson, Goldberg, Head, Kivimäki, Madsen, Rugulies, Vahtera, Zins, Rod, 2016), we argue that lifestyle factors such as current smoking and high alcohol consumption, leisure time physical inactivity are likely to be a consequence of caregiver stress. Therefore, we have not included these factors as confounders in our analyses, as this would attenuate the effect of caregiving stress on cortisol levels mediated through these pathways. This is also the case with psychological disease such as depression, which is a likely consequence of caregiver stress (Capistrant, Berkman, Glymour, 2014; Pinquart and Sörensen, 2003).

Statistical analyses

The CAR was calculated by subtracting the cortisol measure at awakening from the cortisol measure at approximately 30 minutes from awakening. Conventionally, analyses were restricted to wakeup samples that were collected within 10 minutes of waking because of a reduced CAR in those with longer delays (Kudielka, Broderick, Kirschbaum, 2003). Furthermore, the second measure was restricted to samples collected no later than 45 min from awakening (Kudielka, Broderick, Kirschbaum, 2003). The CAR was further analyzed in a standard regression model:

$$Y_i = \alpha' X_i + \varepsilon_i$$

where Yi denotes the CAR, α is a vector of regression coefficients, X_i is a vector constructed from the above mentioned covariates, and ε_i is independent (between subjects) error terms assumed to be mean zero and with same variance. Cortisol slopes were analyzed in a multilevel linear regression model with random slopes and intercept, and hours as the time unit. In the multilevel model, measurement time was used as a level one identifier and person as a level two identifier. Interaction with time was included in the model for the caregiving exposure and included confounders:

$$\log \tilde{Y}_i = \widetilde{\alpha'} \widetilde{X}_\iota + \widetilde{\gamma'} \widetilde{X}_\iota t_{i,j}) + \left(\beta_{0,i} + \beta_{1,i} t_i\right) + \widetilde{\varepsilon_{\iota,J}}$$

where $\widetilde{X}_\iota$ is the relevant set of covariates (see above), $t_{i,j}$ is the jth measurement time of subject i, β's are person specific random effects assumed to be mean zero, and $\widetilde{\varepsilon_{\iota,J}}$ is residual variation. The five time points from the 30 min peak until nadir was used for calculating cortisol slopes (Adam and Kumari, 2009). Because of right-skewed distributions of diurnal cortisol levels, data on cortisol slopes were log transformed. For all analyses, participants with cortisol values outside ±3 SD of the mean were considered outliers and removed from analyses (Adam and Kumari, 2009; Stalder, Kirschbaum, Kudielka, Adam, Pruessner, Wüst, Dockray, Smyth, Evans, Hellhammer, Miller, Wetherell, Lupien, Clow, 2015). Also, participants, who woke up before 4am or after 11am, were removed from analyses as diurnal cortisol patterns may not be accurately determined with such measurements (Karlamangla, Friedman, Seeman, Stawski, Almeida, 2013). Both CAR (nmol/L) and cortisol slope (log nmol/L/hour) were analyzed as the outcome in the following models, separately for men and women: We tested the difference in β-coefficients (95% CI) in the following

analyses, with non-caregivers as the reference: informal caregiving, caregiving according to care recipients (either spouse or parent, relative, and friend), weekly caregiving hours categorized as 1-8 hours, 9-20 hours, 20-100 hours, and >100 hours, and finally short- and long-term caregiving. Test for trend on weekly hours of caregiving was analyzed using the categorical variable generated. We also created four mutual exclusive categories to investigate the double burden of caregiving and work: 1. Non-caregivers, non-workers, 2. Non-caregivers, workers, 3. Caregivers, non-workers, 4. Caregivers, workers. We assessed additive interaction by including a product term in the model (von Elm, Altman, Egger, Pocock, Gøtzsche, Vandenbroucke, 2007). For difference in the CAR, a negative β represents a blunted response, while for the difference in cortisol slope, a negative β represents a steeper slope. For the CAR, Cohen's d were calculated, with effect sizes of 0-0.20 standard deviations representing small effects, 0.20-0.50 standard deviations representing medium effects, and >0.50 representing large effects (Cohen J, 1992).

Different approaches have been suggested in analyzing the CAR (Stalder, Kirschbaum, Kudielka, Adam, Pruessner, Wüst, Dockray, Smyth, Evans, Hellhammer, Miller, Wetherell, Lupien, Clow, 2015). Based on this, we made sensitivity analyses for CAR adjusted for the cortisol level at wakeup, and secondly we analyzed the ratio between the CAR and time difference between the wake-up measure and peak response (Adam and Kumari, 2009; Stalder, Kirschbaum, Kudielka, Adam, Pruessner, Wüst, Dockray, Smyth, Evans, Hellhammer, Miller, Wetherell, Lupien, Clow, 2015). These other approaches to analyze the association between informal caregiving and the CAR yielded similar results as the main analyses (Appendix C). According to recommendations by International Society of Psychoneuroendocrinology, we present cortisol means and standard deviations for the cortisol response at wakeup (Stalder, Kirschbaum, Kudielka, Adam, Pruessner, Wüst, Dockray, Smyth, Evans, Hellhammer, Miller, Wetherell, Lupien, Clow, 2015), showing that there was no difference in wakeup response for caregivers and non-caregivers (Appendix C).

Results

In the study population, 286 (10%) men and 128 (13%) women provided informal caregiving. More men than women were married or cohabiting, and men had higher average annual household income. Among women, caregivers were more likely to be married than non-caregivers, whereas male caregivers were not more likely to be married than non-caregivers (Table 1).

Informal caregiving and cortisol

We observed a sex difference in diurnal patterns of cortisol, with male caregivers having a more blunted CAR than female caregivers compared with non-caregivers ($P_{Interaction}$=0.03). As seen in figure 1, we found that providing care was associated with a blunted CAR (β: -1.39 % CI: -2.74;-0.04 (nmol/L)) (Cohen's d=0.14) in men. For women, we found that caregivers had a steeper CAR than non-caregivers (β: 1.33 % CI: -0.86;3.51 (nmol/L))(Cohen's d=0.10), but the results were not statistically significant. For cortisol slopes, we found no difference between caregivers and non-caregivers, neither for men (β: 0.001; 95% CI: -0.006;0.008 (log nmol/L/hour)), nor women (β: 0.001; 95% CI: -0.011;0.012 (log nmol/L/hour)).

In table 2, mean cortisol levels at wake-up, 30 minutes response and bedtime are displayed along with the CAR, for each exposure group. As seen among men, a mean CAR of ≤4 nmol/L was observed among those providing ≥20 hours of weekly caregiving, short-term caregivers, and caregivers in paid work. Whereas, for women the lowest CAR was observed in non-caregivers and short-term caregivers with mean CAR of <8 nmol/L.

Various aspects of informal caregiving and cortisol

As seen in table 3, for men, we found a dose-response relationship between weekly hours of caregiving and the CAR, showing that more hours of care was associated with a more blunted CAR (P_{trend}=0.03). Furthermore, among men, we found that short-term caregiving was associated with a markedly blunted CAR (β: -3.27; 95% CI: -5.35;-1.19 (nmol/L))(Cohen's d=0.30) and a non-significant tendency for a flattened slope (β: 0.009; 95% CI: -0.002;0.020 (log nmol/L/hour)) compared with non-caregiving. The same was not

found for long-term caregiving. For women, providing care for a spouse was associated with a markedly elevated CAR compared with non-caregivers (β: 4.53; 95% CI: 0.54;8.51 (nmol/L))(Cohen's d=0.33).

Effect modification from work life

As seen in table 4, when stratifying by work status, providing care was associated with a blunted response among men in paid work ($P_{Interaction}$=0.07); male caregivers in paid work had a blunted CAR (β: -3.23; 95% CI: -5.73;-0.72 (nmol/L)) (Cohen's d=0.26), while caregivers without paid work did not have a blunted CAR compared with men who did not work and did not provide care (β: -0.71; 95% CI: -2.33; 0.91 (nmol/L)) (Cohen's d=0.06). For women, there was an interaction between caregiving and paid work, with caregivers in paid work having a steeper cortisol slope than would be expected from the effects of caregiving and work alone ($P_{Interaction}$=0.01). Among non-caregiving women, we found that those in paid work had a flattened slope relative to those, who were not in paid work (β: 0.014; 95% CI: 0.004;0.025 (log nmol/L/hour)). Unadjusted and multiple adjusted analyses yielded similar results (Appendix C).

Discussion

We found that male caregivers had a blunted CAR compared to non-caregivers. This association was especially pronounced among those providing many weekly hours of care, and caregivers in paid work. For women, spousal caregivers had a greater CAR than non-caregivers did.

In a recent systematic review, being caregiver for a family member with dementia was associated with higher cortisol levels throughout the day (Allen, Curran, Duggan, Cryan, Chorcoráin, Dinan, Molloy, Kearney, Clarke, 2017), and studies included in the review, found a blunted CAR in caregivers compared with non-caregivers (de Vugt, Jolles, van Osch, Stevens, Aalten, Lousberg, Verhey, 2006; Fonareva, Amen, Zajdel, Ellingson, Oken, 2011). This association was driven by higher cortisol levels at wakeup for caregivers compared with non-caregivers. In line with this, individuals with low wakeup levels

of cortisol tend to have a steep CAR, whereas individuals with high wakeup levels have a blunted CAR (Stalder, Kirschbaum, Kudielka, Adam, Pruessner, Wüst, Dockray, Smyth, Evans, Hellhammer, Miller, Wetherell, Lupien, Clow, 2015). We found that male caregivers had a blunted CAR compared with non-caregivers in a dose-response relation, showing that more weekly hours of caregiving were associated with a more blunted CAR. In contrast to studies included in the review (de Vugt, Jolles, van Osch, Stevens, Aalten, Lousberg, Verhey, 2006; Fonareva, Amen, Zajdel, Ellingson, Oken, 2011), we found no difference in cortisol levels at wakeup between caregivers and non-caregivers. For women, we found that caregivers tended to have a steeper CAR compared with non-caregivers. Although this finding was not statistically significant, it is consistent with the hypothesis that caregiving responsibilities may influence the physiological stress response differently in men and women (Stafford, Gardner, Kumari, Kuh, Ben-Shlomo, 2013).

We stratified analyses by work status and found that male caregivers in paid work had a blunted CAR compared to male non-caregivers who were not in paid work. For women, we found an interaction showing that caregivers in paid work had a steeper cortisol slope than would be expected from the effects of caregiving and work alone. A higher peak did not explain this result, as we did not find a higher CAR for working women also providing care. Steeper slopes have previously been associated with better health status, as it may indicate that these individuals find it easier to unwind before bedtime (Adam and Kumari, 2009). Thus, our result may point to a 'healthy caregiver effect', where more privileged women, who provide care, stay in the labor market due to good working conditions in contrast to less privileged women, who quit the labor market because of caregiving responsibilities. This explanation is partly supported by previous findings, which shows that less privileged individuals have flatter cortisol slopes (Karlamangla, Friedman, Seeman, Stawski, Almeida, 2013).

In contrast to our hypothesis, we found that short-term caregiving and not long-term caregiving, was associated with a blunted CAR in men. A possible explanation may be that long-term

caregivers have learned to cope with the role as caregiver, whereas short-term caregivers may struggle with adapting to the new role and the associated life changes. In line with this, a study among informal caregivers who were in paid work have shown that in severe care situations, caregivers adapt their work situation by taking time off work or having flexible work arrangements (Oldenkamp, Bültmann, Wittek, Stolk, Hagedoorn, Smidt, 2017); which may explain why the caregiving burden among those in paid work may be diminished in long-term caregivers.

Women providing spousal caregiving had a markedly elevated CAR compared with non-caregivers, whereas care for parents, friends, or other relatives was associated with a very small non-significant elevated CAR. We hypothesized a blunted CAR as a result of chronic stress or burnout, and we hypothesized that this would be more pronounced for spousal caregivers (Lee, Colditz, Berkman, Kawachi, 2003), because of high intensity caregiving from living in the same household (AARP - Public policy Institute, 2015). However, the elevated CAR observed in spousal caregivers may be a consequence of acute stress reactivity in the morning. This response may not necessarily be unhealthy, since results suggest that women providing spousal caregiving do not have flatter cortisol slopes than non-caregivers, indicating that they do not have more difficulties unwinding in the evening.

It has been proposed that cortisol concentrations measured in saliva in healthy adults increase by between 50% and 160% in the first 30 minutes post-awakening, corresponding to an average increase of 9 nmol/l (range 4–15 nmol/L) (Clow, Thorn, Evans, Hucklebridge, 2004). Based on this, a CAR of less than 4 nmol/L with medium effect sizes (Cohen J, 1992), which was observed among male caregivers providing ≥20 hours of weekly care, male short-term caregivers, and male caregivers in paid work, seems to be an unhealthy awakening response.

Methodological considerations

The study population encompassed British adults with a working history in the civil service of whom most had retired. This should be taken into consideration when generalizing the results. A limitation of this study is that cortisol was measured on a single weekday, and may therefore not necessarily be representative of their diurnal cortisol levels in general. Having cortisol measures for several weekdays in future studies would strengthened the evidence (Karlamangla, Friedman, Seeman, Stawski, Almeida, 2013). Another limitation is that we had no information on whether participants provided care for disabled or chronically ill children. Thus, there could potentially be informal caregivers in the non-caregiving group, which were not identified by the questionnaire. This misclassification would attenuate the true effect of informal caregiving on diurnal cortisol levels. Furthermore, we had no information on the type of caregiving provided or the severity of the disease of the care recipient, although there may be large differences in the caregiving burden, based on whether providing assistance with e.g. personal care or grocery shopping (Hunt GG, Reinhard S, Greene R, Whiting GC, Feinberg LF, 2015). In addition, caregiving for a spouse or parent with dementia may constitute a greater caregiving burden than care for a disabled relative with no cognitive deficits (Allen, Curran, Duggan, Cryan, Chorcoráin, Dinan, Molloy, Kearney, Clarke, 2017).

A high number of eligible participants did not return cortisol samples, probably due to the cumbersome task of collecting six saliva samples at particular time intervals. This non-compliance was prevalent in participants with older age and higher household income, but not with gender. It is possible that non-compliance is most common among those who are mostly stressed by the burden of informal caregiving. If this was the case, then our results would have presented a more healthy stress response in informal caregivers than would be seen in the total population of eligible participants. However, non-compliance was associated with higher household income, and thus spousal caregivers, which was hypothesized to be those mostly stressed by the burden of informal caregiving, are likely to be underrepresented among non-compliers.

Almost three quarters of the study population were men, which could be considered as a strength, since many studies on informal caregiving are primarily based on women. However, there may have been insufficient power to observe an association in women due to smaller numbers. On the other hand, we found that gender modified the association statistically significantly, showing that male caregivers had a blunted CAR compared with female caregivers. Thus, although only a modest difference in the CAR was observed among men and women, these gender differences are interesting since most previous research on informal caregiving have highlighted that women may be more prone to stress from caregiving, due to a greater caregiving burden in women, compared with men (Navaie-Waliser et al., 2002; Verbakel et al., 2017).

Conclusion

To our knowledge, this is the first study investigating various aspects of informal caregiving such as the relation with the care recipient, weekly hours of proving caregiving, and caregiving duration in relation to diurnal cortisol levels. For male caregivers we found a blunted CAR, which in other studies has been associated with chronic stress and burnout. This observed association was especially pronounced for those those providing many weekly hours of care, short-term caregivers, and caregivers in paid work. For female caregivers we found the opposite tendency, showing an elevated CAR compared with non-caregiving women, which was especially pronounced among spousal caregivers. Our findings indicate that men and women providing informal caregiving may have different cortisol profiles. Further research is needed to examine whether our results are generalizable beyond UK citizens with a working history in the civil service.

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Table 1. Baseline characteristics of the study population

Study population	Men (n=2,772)		Women (n=955)	
	Caregivers (n=286)	Non-caregivers (n=2,486)	Caregivers (n=128)	Non-caregivers (n=827)
Age, mean (±SD)	66 (±6)	66 (±6)	65 (±6)	66 (±6)
Married/cohabitating	84%	84%	62%	52%
In paid work	30%	32%	23%	25%
Non-Caucasian	6%	5%	10%	9%
Low annual household income (<20,000£)	16%	13%	30%	39%

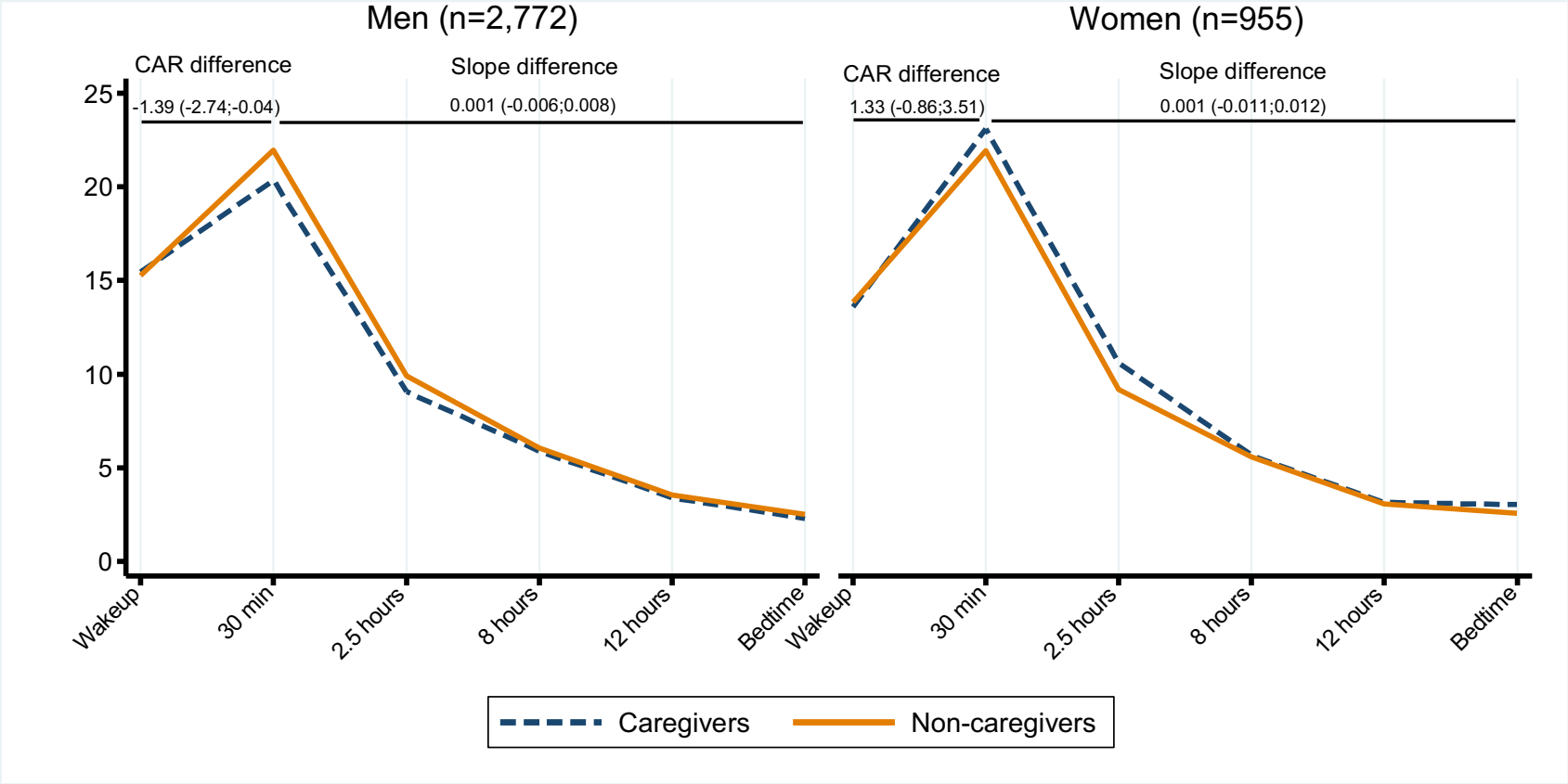


Figure 1. Difference in cortisol awakening response (CAR) and cortisol slope for informal caregivers vs non-caregivers

The CAR estimates is presented as difference in β (95%CI) (nmol/L) and slope estimates is presented as difference in β (95%CI) (log nmol/L/hour). Multiple adjusted for age, ethnicity, marital status, employment status, income category, wake-up time. Interaction between informal caregiving and gender for the CAR (p=0.03). For difference in the CAR, a negative β represents a blunted response, and for the difference in cortisol slope, a negative β represents a steeper slope.

Table 2. Various aspects of informal caregiving and cortisol levels

	Men (n=2,772)				Women (n=955)			
	Cortisol at wake-up	**Cortisol at 30 min**	**Cortisol at bedtime**	**CAR**	**Cortisol at wake-up**	**Cortisol at 30 min**	**Cortisol at bedtime**	**CAR**
Informal caregiving:								
Caregivers	15.0 (±7.3)	19.9 (±10.2)	2.3 (±2.2)	4.9 (±10.1)	13.6 (±6.6)	22.6 (±13.0)	2.9 (±3.0)	9.0 (±12.7)
Non-caregivers	14.9 (±7.3)	21.3 (±11.4)	2.5 (±2.4)	6.4 (±11.1)	13.9 (±7.8)	21.8 (±11.8)	2.5 (±2.1)	7.9 (±11.6)
Care recipient:								
Spouse	14.6 (±6.4)	19.1 (±8.8)	2.5 (±1.9)	4.5 (±9.1)	12.5 (±6.0)	24.1 (±14.7)	2.8 (±2.5)	11.7 (±12.7)
Parent, relative or friend	15.1 (±7.7)	20.3 (±10.8)	2.2 (±2.4)	5.1 (±10.5)	14.1 (±6.8)	22.0 (±12.3)	3.0 (±2.1)	8.0 (±12.6)
Weekly caregiving hours:								
>100 hours	16.0 (±8.8)	20.0 (±7.8)	1.9 (±1.2)	4.0 (±8.5)	12.5 (±6.3)	21.3 (±14.9)	3.2 (±3.2)	8.8 (±12.7)
20-100 hours	16.7 (±7.7)	20.0 (±9.3)	2.0 (±1.5)	3.3 (±10.0)	13.2 (±8.2)	22.7 (±12.7)	2.8 (±2.7)	9.5 (±14.8)
9-20 hours	14.4 (±7.6)	19.3 (±11.2)	2.4 (±3.1)	4.9 (±10.3)	12.7 (±6.9)	21.0 (±13.4)	3.6 (±4.5)	8.3 (±12.5)
1-8 hours	14.7 (±6.2)	20.7 (±10.3)	2.4 (±2.2)	6.0 (±10.3)	14.5 (±5.8)	24.5 (±12.8)	2.4 (±2.3)	10.0 (±12.2)
Duration of caregiving:								
Long-term caregiving	15.3 (±7.4)	20.7 (±10.2)	2.0 (±1.4)	5.5 (±9.8)	13.7 (±7.0)	23.5 (±14.0)	3.0 (±2.9)	9.8 (±13.0)
Short-term caregiving	14.8 (±7.6)	17.9 (±9.2)	2.5 (±3.0)	3.1 (±9.5)	14.8 (±6.7)	22.6 (±12.0)	3.2 (±3.5)	7.8 (±12.9)
Informal caregiving and paid work:								
Caregivers, in paid work	16.2 (±8.2)	19.6 (±9.9)	2.1 (±1.6)	3.3 (±11.3)	14.8 (±8.2)	23.5 (±8.7)	2.3 (±1.6)	8.7 (±9.4)
Caregivers, not in paid work	14.4 (±6.9)	20.0 (±10.3)	2.3 (±2.5)	5.6 (±9.4)	13.3 (±6.1)	22.4 (±14.1)	3.1 (±3.2)	9.1 (±13.6)
Non-caregivers, in paid work	14.7 (±7.0)	21.5 (±11.2)	2.5 (±2.6)	6.8 (±11.3)	14.0 (±6.6)	21.4 (±12.5)	2.5 (±2.3	7.4 (±12.4)
Non-caregivers, not in paid work	14.9 (±7.5)	21.2 (±11.6)	2.5 (±2.4)	6.3 (±11.0)	13.9 (±8.1)	21.9 (±11.6)	2.5 (±2.1)	8.1 (±11.3)

Cortisol presented as nmol/L mean (±SD). Abbreviations: CAR = Cortisol Awakening Response

Table 3. Various aspects of informal caregiving and the association with differences in cortisol awakening response (CAR) and cortisol slopes

	Men (n=2,772)			Women(n=955)		
	Number (%)	**Difference in CAR (nmol/L) β (95%CI)**	**Difference in slope (log nmol/L/hour) β (95%CI)**	**Number (%)**	**Difference in CAR (nmol/L) β (95%CI)**	**Difference in slope (log nmol/L/hour) β (95%CI)**
Care recipient:						
Spouse	89 (3%)	-1.96 (-4.32;0.39)	0.007 (-0.006;0.019)	36 (4%)	4.53 (0.54;8.51)	-0.008 (-0.028;0.012)
Parent, relative or friend	197 (7%)	-1.32 (-2.94;0.30)	-0.002 (-0.010;0.007)	92 (10%)	0.15 (-2.47;2.78)	0.004 (-0.009;0.017)
Non-caregivers	2,486 (90%)	0	0	827 (87%)	0	0
Weekly caregiving hours:						
>100 hours	33 (1%)	-2.38 (-6.15;1.40)	-0.011 (-0.030;0.009)	28 (3%)	1.40 (-3.20;6.00)	0.017 (-0.006;0.040)
20-100 hours	40 (1%)	-3.17 (-6.60;0.25)	-0.015 (-0.033;0.003)	28 (3%)	1.68 (-2.75;6.10)	-0.005 (-0.027;0.017)
9-20 hours	60 (2%)	-1.46 (-4.27;1.35)	0.012 (-0.003;0.027)	21 (2%)	0.52 (-4.74;5.78)	0.006 (-0.020;0.031)
1-8 hours	128 (5%)	-0.37 (-2.36;1.63)	0.002 (-0.009;0.012)	40 (4%)	2.03 (-1.79;5.84)	-0.013 (-0.032;0.006)
Non-caregivers	2,485 (91%)	0	0	827 (88%)	0	0
Test for trend		p=0.03	p=0.44		p=0.29	p=0.45
Duration of caregiving:						
Long-term caregiving	142 (5%)	-0.87 (-2.74;1.00)	-0.008 (-0.018;0.003)	69 (8%)	2.04 (-0.95;5.02)	0.004 (-0.011;0.025)
Short-term caregiving	115 (4%)	-3.26 (-5.33;-1.18)	0.009 (-0.002;0.020)	39 (4%)	-0.24 (-4.07;3.59)	0.005 (-0.014;0.025)
Non-caregivers	2,421 (90%)	0	0	799 (88%)	0	0

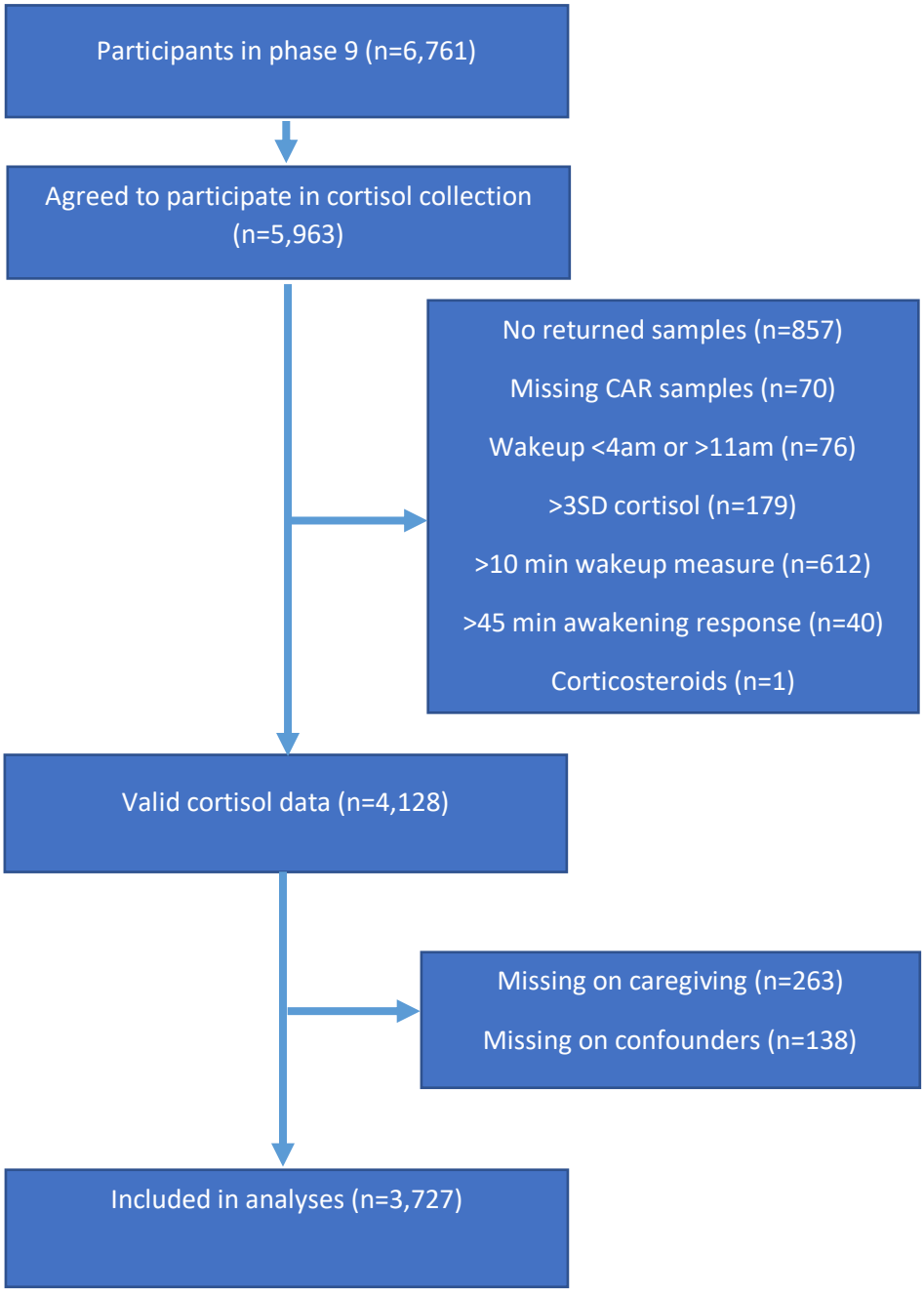
Multiple adjusted Age, ethnicity, married/cohabiting, gainful employment, income category, and wakeup time. For difference in the CAR, a negative β represents a blunted response, and for the difference in cortisol slope, a negative β represents a steeper slope.

Table 4. Joint effects of informal caregiving and paid work on differences in cortisol awakening response (CAR) and cortisol slopes

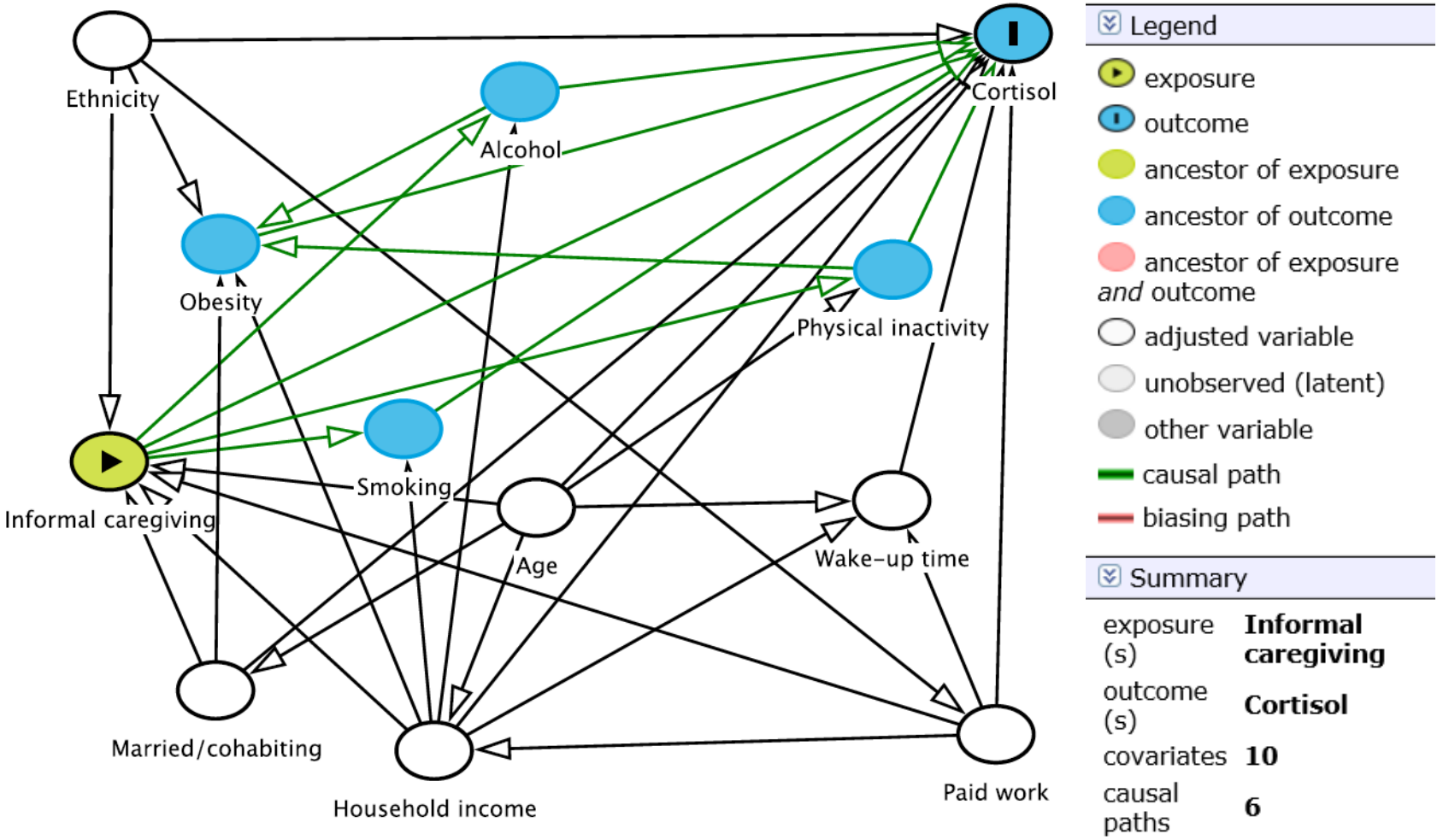
	Men (n=2,772)			Women (n=955)		
	Number (%)	**Difference in CAR (nmol/L) β (95%CI)**	**Difference in slope (log nmol/L/hour) β (95%CI)**	**Number (%)**	**Difference in CAR (nmol/L) β (95%CI)**	**Difference in slope (log nmol/L/hour) β (95%CI)**
Caregivers, in paid work	86 (3%)	-3.23 (-5.73;-0.72)	0.004 (-0.003;0.009)	30 (3%)	-0.27 (-4.79;4.26)	-0.014 (-0.036;0.009)
Caregivers, not in paid work	200 (7%)	-0.71 (-2.33;0.91)	0.000 (-0.008;0.009)	98 (10%)	1.41 (-1.16;3.98)	0.009 (-0.003;0.022)
Non-caregivers, in paid work	805 (29%)	0.23 (-0.86;1.31)	0.002 (-0.008;0.018)	206 (22%)	-1.79 (-3.97;0.40)	0.014 (0.004;0.025)
Non-caregivers, not in paid work	1,681 (61%)	0	0	621 (65%)	0	0

Multiple adjusted Age, ethnicity, married/cohabiting, gainful employment, income category, and wakeup time. CAR interaction: men p=0.07, women p=0.97. Slope interaction: men p=0.84, women p=0.01. For difference in the CAR, a negative β represents a blunted response. For the difference in cortisol slope, a negative β represents a steeper slope.

Appendix A. Flowchart, Whitehall II



Appendix B. Directed acyclic graph of the causal effect of informal caregiving on cortisol levels



Appendix C. Sub- and sensitivity analyses

C1. Joint effects of informal caregiving and paid work on cortisol awakening response and diurnal cortisol slope

	Men (n=2,772)		Women (n=955)	
	Difference in CAR Unadjusted, β (95%CI)	**Difference in CAR Multiple adjusted, β (95%CI)**	**Difference in CAR Unadjusted, β (95%CI)**	**Difference in CAR Multiple adjusted, β (95%CI)**
Caregivers, workers	-3.227 (-5.657;-0.797)	-3.225 (-5.729;-0.720)	0.602 (-3.856;5.059)	-0.267 (-4.790;4.257)
Caregivers, non-workers	-0.814 (-2.433;0.805)	-0.709 (-2.328;0.911)	1.179 (-1.379;3.737)	1.411 (-1.159;3.980)
Non-caregivers, workers	0.339 (-0.593;1.271)	0.226 (-0.858;1.309)	-0.632 (-2.515;1.251)	-1.786 (-3.967;0.396)
Non-caregivers, non-workers	0 (ref.)	0 (ref.)	0 (ref.)	0 (ref.)
	Difference in cortisol slope Unadjusted, β (95%CI)	**Difference in cortisol slope Multiple adjusted, β (95%CI)**	**Difference in cortisol slope Unadjusted, β (95%CI)**	**Difference in cortisol slope Multiple adjusted, β (95%CI)**
Caregivers, workers	-0.000 (-0.013;0.013)	0.005 (-0.008;0.018)	-0.015 (-0.038;0.007)	-0.014 (-0.036;0.009)
Caregivers, non-workers	-0.000 (-0.009;0.009)	0.000 (-0.008;0.009)	0.005 (-0.008;0.017)	0.009 (-0.003;0.022)
Non-caregivers, workers	-0.001 (-0.006;0.004)	0.003 (-0.003;0.009)	0.004 (-0.005;0.014)	0.014 (0.004;0.025)
Non-caregivers, non-workers	0 (ref.)	0 (ref.)	0 (ref.)	0 (ref.)

CAR: Cortisol Awakening Response. Slopes were analyzed using log(cortisol) with hours as the underlying time axis. Multiple adjusted: Age, ethnicity, married/cohabiting, gainful employment, income category, wakeup time. CAR: Interaction, men p=0.07, women p=0.97. Slope: Interaction, men p=0.84, women p=0.01. For difference in the CAR a negative β represents a blunted response, and for the difference in cortisol slope a negative β represents a steeper slope.

C2. Associations between various aspects of informal caregiving and cortisol awakening response

	Men (n=2,772)		Women (n=955)	
	Difference in CAR Unadjusted, β (95%CI)	**Difference in CAR Multiple adjusted, β (95%CI)**	**Difference in CAR Unadjusted, β (95%CI)**	**Difference in CAR Multiple adjusted, β (95%CI)**
Informal caregiving	-1.633 (-2.991;-0.275)	-1.392 (-2.740;-0.044)	1.205 (-1.034;3.444)	1.329 (-0.857;3.514)
Care recipient:				
Spouse	-2.208 (-4.539;0.123)	-1.965 (-4.316;0.386)	3.676 (-0.246;7.598)	4.526 (0.537;8.514)
Parent, relative or friend	-1.368 (-2.982;0.246)	-1.320 (-2.938;0.298)	0.171 (-2.442;2.783)	0.153 (-2.473;2.778)
Weekly caregiving hours:				
1-8 hours	-0.416 (-2.411;1.579)	-0.367 (-2.361;1.628)	1.983 (-1.855;5.821)	2.026 (-1.792;5.844)
9-20 hours	-1.596 (-4.406;1.214)	-1.462 (-4.270;1.346)	0.489 (-4.744;5.722)	0.520 (-4.738;5.778)
20-100 hours	-3.184 (-6.611;0.244)	-3.173 (-6.601;0.254)	1.518 (-2.926;5.962)	1.677 (-2.749;6.103)
>100 hours	-2.565 (-6.333;1.203)	-2.375 (-6.148;1.398)	0.862 (-3.745;5.468)	1.401 (-3.198;6.000)
Test for trend	p=0.026	p=0.023	p=0.392	p=0.287
Duration of caregiving:				
Long-term caregiving	-1.015 (-2.880;0.850)	-0.921 (-2.786;0.944)	1.834 (-1.118;4.786)	2.069 (-0.895;5.032)
Short-term caregiving	-3.295 (-5.343;-1.247)	-3.090 (-5.142;-1.037)	-1.097 (-4.747;2.552)	-0.834 (-4.465;2.797)

CAR=Cortisol Awakening Response (nmol/L). Non-caregivers is the reference group in all analyses. Multiple adjusted: Age, ethnicity, married/cohabiting, gainful employment, income category, wakeup time. For difference in the CAR a negative β represents a blunted response.

C3. Associations between various aspects of informal caregiving and diurnal cortisol slopes

	Men (n=2,772)		Women (n=955)	
	Difference in Slope Unadjusted, β (95%CI)	**Difference in Slope Multiple adjusted, β (95%CI)**	**Difference in Slope Unadjusted, β (95%CI)**	**Difference in Slope Multiple adjusted, β (95%CI)**
Informal caregiving	0.000 (-0.007;0.008)	0.001 (-0.006;0.008)	-0.001 (-0.012;0.010)	0.001 (-0.011;0.012)
Care recipient:				
Spouse	0.008 (-0.005;0.020)	0.007 (-0.006;0.020)	-0.005 (-0.025;0.015)	-0.008 (-0.028;0.012)
Parent, relative or friend	-0.003 (-0.012;0.006)	-0.002 (-0.010;0.007)	0.000 (-0.013;0.013)	0.004 (-0.009;0.017)
Weekly caregiving hours:				
1-8 hours	-0.000 (-0.011 ;0.010)	0.002 (-0.009;0.012)	-0.016 (-0.035;0.004)	-0.013 (-0.032;0.006)
9-20 hours	0.010 (-0.005;0.025)	0.012 (-0.003;0.027)	-0.002 (-0.028;0.023)	0.006 (-0.020;0.031)
20-100 hours	-0.012 (-0.031;0.006)	-0.015 (-0.033;0.003)	-0.001 (-0.023;0.021)	-0.005 (-0.027;0.017)
>100 hours	-0.009 (-0.028;0.011)	-0.011 (-0.030;0.009)	0.016 (-0.007;0.039)	0.017 (-0.006;0.040)
Test for trend	p=0.476	p=0.443	p=0.546	p=0.448
Duration of caregiving:				
Long-term caregiving	-0.009 (-0.019;0.002)	-0.008 (-0.018;0.003)	0.001 (-0.014;0.016)	0.004 (-0.011;0.025)
Short-term caregiving	0.008 (-0.003;0.020)	0.009 (-0.002;0.020)	0.004 (-0.016;0.023)	0.005 (-0.014;0.025)

Non-caregivers is the reference group in all analyses. The underlying time axis is hours. Slopes are analysed using log(cortisol). Multiple adjusted: Age, ethnicity, married/cohabiting, gainful employment, income category, wakeup time. For difference in cortisol slope a negative β represents a steeper slope.

C4. Sensitivity analyses for the association between informal caregiving and various analyses on cortisol awakening response

	Men (n=2,772)		Women (n=955)	
	Difference in CAR **Multiple adjusted, β (95%CI)**	**P-value**	**Difference in CAR** **Multiple adjusted, β (95%CI)**	**P-value**
Informal caregiving, main analysis	-1.523 (-2.882;-0.163)	p=0.028	1.436 (-0.805;3.678)	p=0.209
Adjusted for morning cortisol	-1.480 (-2.784;-0.177)	p=0.026	1.363 (-0.781;3.506)	p=0.213
CAR/time difference	-0.049 (-0.092;-0.006)	p=0.026	0.044 (-0.027;0.114)	p=0.227

CAR: Cortisol Awakening Response. Multiple adjusted: Age, ethnicity, married/cohabiting, gainful employment, income category, wakeup time. For difference in the CAR a negative β represents a blunted response.