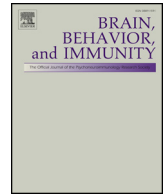




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# The long-term association of adverse childhood experiences with C-reactive protein and hair cortisol: Cumulative risk versus dimensions of adversity

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## ABSTRACT

**Background:** Exposure to adverse childhood experiences (ACEs) may lead to stress-induced upregulation of inflammatory and neuroendocrine processes. However, it remains unclear whether such effects persist into later life, and which dimensions of ACEs might have the strongest impact on these biological mechanisms. Therefore, this study investigated the effects of ACEs on C-reactive protein (CRP) and hair cortisol in a large sample of older adults, distinguishing between cumulative exposure and dimensions of ACEs.

**Methods:** We utilised data from the English Longitudinal Study of Ageing. ACEs were assessed through retrospective reports at wave 3(2006/07). CRP (N = 4198) was measured at waves 4(2008/09) and 6(2012/13), and hair cortisol (N = 3357) at wave 6. The effects of ACEs cumulative exposure were examined using linear and ordinal logistic regression analysis. ACEs dimensions (i.e. threat, household dysfunction, low parental bonding, and loss of an attachment figure) were identified using explorative and confirmatory factor analysis with cross-validation. All analyses were adjusted for relevant confounders.

**Results:** Participants with three or more ACEs had higher CRP levels at wave 4 and an elevated risk of high CRP concentrations across waves 4 and 6 compared with those who did not experience any ACEs. The four ACEs dimensions were all positively associated with both CRP outcomes and had similar effect sizes. In contrast, neither the cumulative score nor the dimensions of ACEs were significantly related to hair cortisol. However, there was a positive, yet small, interaction effect between ACEs and age on hair cortisol.

**Conclusion:** Older adults who retrospectively reported three or more ACEs had chronically elevated CRP levels and exhibited a slightly steeper increase in hair cortisol with age. Different dimensions of ACEs had similar associations with the biomarkers.

## 1. Introduction

A large body of research has made it clear that adverse childhood experiences (ACEs) play a pivotal role in shaping adult health outcomes throughout the life course (Felitti and Anda, 2009). ACEs such as abuse, neglect, and parental divorce are emerging as a critical public health issue across the world owing to their high prevalence and association with numerous causes of morbidity and mortality (NHS Highland Public Health, 2018; Sethi et al., 2018; Stoltenborgh et al., 2015). Different meta-analyses and systematic reviews have provided robust evidence for the link between ACEs and several mental and physical health conditions such as cardiovascular disease, diabetes, depression, and anxiety disorders (Hughes et al., 2017; Kalmakis and Chandler,

2015; Norman et al., 2012).

Maladaptive alterations in the function of the inflammatory response system and the hypothalamic–pituitary–adrenal (HPA)-axis might explain the long lasting effects of ACEs on health (Danese and Lewis, 2017; Danese and McEwen, 2012; Koss and Gunnar, 2018). Exposure to stressful circumstances activates sympathetic nervous system circuits and stimulates the release of corticotropin-releasing hormone (CRH) from the hypothalamus. This in turn triggers a cascade of hormonal reactions resulting in increased production of glucocorticoids from the HPA-axis and greater levels of proinflammatory cytokines in the brain and peripherally (Miller et al., 2009). Although glucocorticoids exert negative feedback to inflammatory processes, HPA-axis hyperactivity and elevated inflammation have been shown to

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be concurrent in the context of chronic stress (Pariante, 2017). Indeed, sustained activation of the HPA-axis can desensitise glucocorticoid receptors thereby reducing the inhibitory effects of glucocorticoids on inflammation (Chiang et al., 2015; Miller et al., 2009). The most fully investigated biomarkers of inflammation and HPA-axis function are C-reactive protein (CRP) and cortisol respectively. Elevated levels of these biomarkers have been implicated in the pathogenesis of several mental and physical health problems such as depression, cardiovascular disease, and diabetes (Acabchuk et al., 2017; Baumeister et al., 2014; Belvederi Murri et al., 2014; Girod and Brotman, 2004; Haapakoski et al., 2015; Hackett and Steptoe, 2017; Iob et al., 2019; Kivimäki and Steptoe, 2017; Stetler and Miller, 2011).

Several studies have examined the relationship of ACEs with inflammation and HPA-axis function providing evidence for elevated CRP and dysregulated cortisol levels in people exposed to ACEs (Baumeister et al., 2015; Danese and Lewis, 2017; Deighton et al., 2018; Kuhlman et al., 2017; Kumari et al., 2013; Pinto Pereira et al., 2019). However, not all studies have confirmed these effects, and some reported opposite associations. For instance, a meta-analysis of ACEs and inflammatory markers in children and adolescents did not find significant evidence for this link (Kuhlman et al., 2019). In relation to cortisol, a systematic review found evidence for increased cortisol reactivity in children exposed to adversity (Hunter et al., 2011). However, a more recent meta-analysis including samples of all ages indicated a moderate negative effect of ACEs on cortisol response to social stress (Bunea et al., 2017). Another meta-analytic synthesis did not find significant evidence for the relationship of early-life stress with the cortisol awakening response, baseline cortisol, or cortisol levels over the day (Fogelman and Canli, 2018). Noticeably, the majority of the included studies were cross-sectional and therefore unable to examine the relationship of ACEs with cortisol and inflammatory markers over time.

A number of gaps and limitations of the current evidence base could explain such inconsistent findings. Relatively little is known about how different dimensions of ACEs may influence CRP and cortisol levels. Indeed, most work has examined this link either focusing on specific types of adversity in isolation or using the cumulative risk approach which creates an overall risk score based on the total number of ACEs experienced by the participant (McLaughlin and Sheridan, 2016). Whereas the specific experience approach ignores the high co-occurrence of ACEs, the cumulative risk method can only take into account the overlap between different elements of stressful experiences thereby ignoring their potentially different physiological effects (Kuhlman et al., 2017; McLaughlin and Sheridan, 2016). A novel approach involves identifying dimensions of adversity that may influence stress physiology in different ways. McLaughlin and Sheridan (2016) have proposed two main dimensions of adversity, namely *threat* – adversities involving harm or threat of harm, and *deprivation* – experiences related to absence of expected cognitive and social input. Interestingly, there is some preliminary empirical evidence suggesting that experiences related to *threat* but not *deprivation* are associated with abnormal cortisol levels (Busso et al., 2017; LoPilato et al., 2019), as well as with other biological and cognitive processes (Machlin et al., 2019; Sumner et al., 2019). However, it is worth noting that this dimensional model may not capture all aspects of ACEs (e.g. parental substance abuse and separation), and certain *deprivation* exposures could actually belong to distinct sub-dimensions (e.g. parental loss and neglect) (Westermair et al., 2018; Zhang et al., 2019). Different lines of research suggest that childhood neglect is particularly important for cortisol dysregulation (Koss and Gunnar, 2018), whereas elevated CRP levels are primarily related to parental absence (Baumeister et al., 2016). Thus, it remains unclear which dimensions of ACEs might have stronger relationships with HPA-axis function and inflammation. Furthermore, childhood socioeconomic disadvantage (e.g. poverty) is often considered as an ACE. However, socioeconomic disadvantage is conceptually different from many types of psychosocial adversity and is likely to be an important predictor of ACEs rather than an ACE itself (Liming, 2018). There also is

a lack of evidence for the possible long-term effects of ACEs on HPA-axis and immune function in later life since most research has focused on young people or middle-aged adults (Li et al., 2015; Wielaard et al., 2017). Importantly, CRP and cortisol have been shown to increase in an age-dependent manner (Feller et al., 2014; Ferrari et al., 2001; Tang et al., 2017). Consequently, the relationship of ACEs with these biomarkers in older adults might be different from that observed in younger people. Moreover, older age is a phase of life particularly prone to the emergence of various long-term conditions which are linked to elevated inflammation and cortisol dysregulations such as cardiovascular disease, arthritis, and diabetes.

Another limitation of the literature concerns the prevailing use of cross-sectional measures of CRP that are not sufficient to provide an accurate assessment of chronic inflammation. Likewise, most research has assessed HPA-axis function using single measures of cortisol in body fluids including saliva, blood, or urine. However, these specimens can only represent short-term cortisol levels and are strongly influenced by an array of situational and inter-individual fluctuations (Stalder and Kirschbaum, 2012). Recently, hair cortisol has emerged as a novel biomarker of psychosocial stress in both children and adults. The measurement of cortisol in hair offers the opportunity to reliably assess long-term exposure to cortisol over several weeks (Fuchs et al., 2018; Iob et al., 2018; Stalder et al., 2017; Staufenbiel et al., 2013). Hair cortisol has been shown to have high test–retest reliability, whereas cortisol measures from body fluids typically have low to moderate stability (Short et al., 2016; Stalder et al., 2012). However, correlations between concentrations of cortisol in hair and body fluids are typically low (Stalder et al., 2017), indicating that they may represent distinct aspects of HPA-axis function. An increasing number of studies have examined the relationship of ACEs with hair cortisol. Increased hair cortisol concentration has been found to be associated with exposure to psychosocial stress during prenatal or postnatal periods in one-year-old infants (Karlen et al., 2013; Palmer et al., 2013). Chronically elevated hair cortisol levels were also observed in a group of maltreated children aged three to eight years old (White et al., 2017). In contrast, psychosocial adversities such as lifetime exposure to trauma, maltreatment and maternal early-life adversity have been associated with diminished hair cortisol levels in late childhood and adolescence (Fuchs et al., 2018; Simmons et al., 2016; White et al., 2017). Accordingly, a recent meta-analysis of the relationship between adversity across all ages and hair cortisol found two classes of studies: the first was characterised by a moderate negative association, whereas the second larger set of studies had a small positive association (Khoury et al., 2019). However, no study to date has examined the association between ACEs and hair cortisol in older adults. Lastly, the majority of studies are based on small, non-representative samples thereby limiting the generalisability and reliability of the results.

The aim of this study was to investigate the association of multiple types of ACEs with hair cortisol and repeated measures of CRP in a large sample of older adults, distinguishing between cumulative risk and dimensions of ACEs. Using data from the English Longitudinal Study of Ageing, we tested three hypotheses. First, we expected that greater cumulative exposure to ACEs would be associated with elevated CRP and hair cortisol levels. Second, we hypothesised that using factor analysis the *threat-deprivation* theme would emerge along with other possible dimensions of ACEs. Third, we expected to find differential associations of distinct ACEs dimensions with CRP and hair cortisol.

## 2. Methods

### 2.1. Sample

The English Longitudinal Study of Ageing (ELSA) is an ongoing, multidisciplinary prospective cohort study of men and women living in England aged 50 years and over (Zaninotto and Steptoe, 2019). The study was initiated in 2002 with a nationally representative sample of

12,099 adults who were drawn from the Health Survey for England. Data collection is conducted every 2 years, with additional nurse visits every 4 years for the collection of biomedical data. Further details on the sample design and data collection methods can be found at [www.elsa-project.ac.uk](http://www.elsa-project.ac.uk). All respondents provided informed consent and ethical approval was obtained from the National Research Ethics Service (NatCen Social Research, 2018). The ELSA datasets can be accessed through the UK Data Service ([www.ukdataservice.ac.uk](http://www.ukdataservice.ac.uk)).

ACEs were assessed during the Life History Interview in wave 3 (2006/07), a one-off module that gathered retrospective information on the participants' early life experiences. Out of the 9,971 individuals who participated in the wave 3 main interview 7,855 participants completed the Life History module. CRP levels were assessed during the nurse visit in waves 4 (2008/09) and 6 (2012/13). Blood samples for the measurement of CRP were successfully collected from 6,439 participants in wave 4 and from 6,126 participants in wave 6. Study members with CRP values greater than 10 mg/L ( $N_{\text{wave4}} = 444$ ;  $N_{\text{wave6}} = 342$ ) were excluded from the analysis since this may reflect immune activation due to current infection or trauma rather than chronic inflammation (Pearson et al., 2003). Hair cortisol was assessed only once as part of the wave 6 nurse visit. Hair samples were successfully collected from 5,451 participants. The sample with hair cortisol measures was restricted to 4,761 participants due to the presence of some undetectable or extreme (greater than 3 standard deviations – i.e. 660 pg/mg) hormone values. For the purpose of this analysis, we created two analytical samples. The first sample (CRP) included all participants with available CRP data at either wave 4 or 6 who completed the Life History interview ( $N = 4,198$ ). The second sample (cortisol) was comprised of all participants with hair cortisol data in wave 6 who completed the Life History module ( $N = 3,357$ ).

## 2.2. Measures

### 2.2.1. Adverse childhood experiences (ACEs)

The Life History interview contains different items on ACEs experienced up to the age of 16 years. These include: sexual abuse, physical assault, physical abuse from parents, parent arguments, parent mental illness or substance abuse, parent separation or divorce, maternal bonding, paternal bonding, separation from mother for more than six months, parent death, foster care or adoption, and institutionalisation. For all items, except parental bonding, participants reported whether or not they ever experienced that particular event during childhood. Child-parent relationships were assessed using the seven-item Parental Bonding Instrument (PBI) (Parker et al., 1979). This questionnaire is designed to retrospectively assess adults' perceptions of their parents' parenting styles (see Appendix for further details). Total bonding scores were calculated separately for each parent figure and ranged from zero (highest bonding) to seven (lowest bonding). For the analysis, we derived two binary measures of low maternal/paternal bonding (total score  $\geq 3$ ). This threshold was selected since the majority of participants (around 83%) had a total bonding score lower than three. The ACEs cumulative score was calculated by adding up the 12 ACEs items described above. This score was then categorised into 4 groups (0, 1, 2, and 3 + ACEs) in order to examine the graded effect of ACEs. The individual ACEs items were also used in factor analysis to identify underlying dimensions of adversity.

### 2.2.2. Biomarkers

**Plasma C-reactive Protein (CRP).** High sensitivity plasma concentrations of CRP were assayed at the Royal Victoria Infirmary laboratory in Newcastle (UK) using the N Latex CRP mono Immunoassay on the Behring Nephelometer II Analyser (Dade Behring, Milton Keynes, UK) (Graig et al., 2006). Exclusion criteria for blood sampling included: clotting or bleeding disorders, history of fits or convulsions, and being on anticoagulant medication (NatCen Social Research, 2018). CRP concentrations were expressed in mg/L. For the purpose of this analysis,

we used two different CRP outcomes: a continuous measure of log-transformed CRP values at wave 4 (CRP w4), and a categorical variable for sustained high CRP levels indicating whether the participant had high CRP levels (i.e.  $\geq 3$  mg/L; Pearson et al., 2003) on zero, one, or two occasions across waves 4 and 6 (high CRP w4-w6).

**Hair cortisol.** Hair strands of approximately 3 cm were collected from the posterior vertex as close to the scalp as possible. Based on an average hair growth of approximately 1 cm per month (Kirschbaum et al., 2009), the 3 cm hair segment closest to the scalp is assumed to provide a measure of the average cortisol output over the three preceding months. Exclusion criteria for hair sampling included: pregnancy, breastfeeding, certain scalp conditions, having less than two cm of hair length, and inability to sit with head remaining still. Hair cortisol concentrations were analysed at the Technische Universität Dresden (Germany) in two separate phases (2015 and 2018) owing to financial constraints. Cortisol levels were assayed using high performance liquid chromatography-mass spectrometry (LC/MS) following a standard wash and steroid extraction procedure (Gao et al., 2013). Hair cortisol concentrations were expressed in pg/mg. Since their distribution was positively skewed, they were log-transformed for the statistical analyses.

### 2.2.3. Covariates

The statistical analyses were adjusted for relevant confounding variables. These included: sex, age, marital status, childhood socioeconomic position, adult wealth, body mass index (BMI), smoking status, physical activity, alcohol drinking, use of anti-inflammatory or antihypertensive drugs (CRP only), steroid medications (cortisol only), and hair-related characteristics - i.e. whether hair was dyed, season of hair collection, hair colour, and phase of hair analysis (cortisol only). A detailed description of the measurement and coding of the covariates can be found in the Appendix.

## 2.3. Statistical analyses

The associations of the ACEs cumulative score with the biomarkers were estimated using linear regression (CRP w4, hair cortisol) and ordinal logistic regression (high CRP w4-w6). For each outcome, we tested three different models: Model 1 – adjusted for age, sex, marital status, medication use, and hair characteristics; Model 2 – Model 1 + childhood socioeconomic position and adult wealth; Model 3 – Model 2 + BMI, smoking status, alcohol drinking, and physical activity. We also tested the interaction effects of the ACEs cumulative score with age and sex. In addition, we calculated E-values and least extreme confidence limits for all statistically significant effects of the ACEs score. This allowed us to determine the average effect required for any possible unmeasured confounder to fully explain the associations between the ACEs score and the biomarkers (VanderWeele and Ding, 2017).

The ACEs dimensions were extracted using explorative factor analysis (EFA) and confirmatory factor analysis (CFA) adopting a 5-fold cross-validation approach (Cohen-Cline et al., 2019; Knafelz and Grey, 2007). This method offers the opportunity to test how the results of one analysis will generalise to an independent dataset. Thus, it is particularly useful to validate the factorial validity of a model and avoid risk of overfitting. The factor analysis of the ACEs items was conducted using the entire sample of participants who completed the Life History interview ( $N = 7855$ ). This sample was randomly split into five equally sized groups: four training datasets and one test dataset. EFA with geomin rotation was conducted in the four training datasets to define the number of underlying factors and identify the most stable and consistent factor structure. In each training set, extraction of factors was informed by inspection of scree plots and eigenvalues. CFA was finally performed on the test dataset. The results of EFA were used to inform the specification of the number of latent factors and item loadings in CFA. Since the ACEs items were categorical, all models were fitted

using a robust weighted least squares estimator (WLSMV). Model fit was evaluated using Root Mean Square Error of Approximation (RMSEA), Comparative Fit Index (CFI), and Tucker Lewis Index (TLI). RMSEA is a measure of absolute fit which should be smaller than 0.06. CFI and TLI are comparative fit indices which should be greater than 0.90 (Byrne, 2012). In a second step, full structural equation models (SEM) were tested to evaluate the association of each ACEs dimension with the biomarkers. For each outcome (i.e. CRP w4, high CRP w4-w6, and hair cortisol) we tested 2 models. The first examined the effect of each ACEs dimension on the biomarkers separately with adjustment for all covariates. In the second model, the associations between the ACEs dimensions and the biomarkers were instead mutually adjusted in order to test their independent effects.

Missing data on all ACEs items, outcome variables, and covariates were estimated using multiple imputation by chained equations (MI). The proportion of missing data in the analytical samples ranged from 0.1% to 25%. All variables included in the analysis were used as predictors in the imputation models in addition to [supplementary data](#) on physical and mental health, survey weights, and covariates from other waves of the study. MI estimates missing information under the Missing at Random (MAR) assumption (Little and Rubin, 2002). Therefore, missing values can be reliably imputed if all variables associated with the missing data generation process are included in the imputation models (Ploubidis et al., 2014). Such assumption is likely to be met in our study since all main drivers of attrition in ELSA - i.e., age, socioeconomic position, and health (Steptoe et al., 2013), were used as predictors in the imputation models. Twenty imputed datasets were created and the pooled estimates from regression, EFA, CFA, and SEM are reported. A comparison of the observed and imputed data demonstrated that these values were broadly similar (Appendix, Tables A1, A2), suggesting that the imputation process was conducted appropriately. Data management and MI analysis were performed in Rstudio 3.4.4 using the R package 'mice' (van Buuren, 2007). Regression analysis was conducted in R using the base functions (linear regression) and the R package 'MASS' (ordinal logistic regression) (Ripley et al., 2019). The EFA, CFA, and SEM models were estimated in Mplus 7. The imputed datasets were exported to Mplus using the R package 'MplusAutomation' (Hallquist and Wiley, 2018). Sensitivity analyses were conducted to explore possible differences in socioeconomic and lifestyle characteristics between the analytical sample and participants excluded from the analysis due to missing ACEs or biomarker data. All models were rerun in the samples of participants with complete data on all measures. Another sensitivity analysis tested the associations between ACEs and the biomarkers controlling for depressive symptoms which were ascertained using the 8-item Center for Epidemiological Studies Depression Scale (CESD-8) at wave 3. In addition, we further examined the relationship of ACEs with CRP excluding only participants with CRP values higher than 20 mg/L (N = 4,782) rather than 10 mg/L.

### 3. Results

#### 3.1. Descriptive statistics

Descriptive statistics of the study participants are reported in [Table 1](#). The average age was 70 years in both the CRP (56% female) and cortisol (68% female) samples. The majority of participants were married. There was a higher proportion of participants in the highest compared to the lowest wealth quintiles. The prevalence of ACEs ranged from 1.6% (e.g. institutionalisation) to 20% (e.g. parent arguments). Around 18% of participants reported one ACE. The percentage of participants with 2 or 3 + ACEs was 9% in the CRP and cortisol samples. Spearman's rank coefficients of the correlations between the ACEs items are reported in the Appendix (Tables A3, A4). Most items were positively correlated, with coefficients ranging from 0.05 (e.g. foster care/adoption and parent mental illness/substance abuse) to 0.45 (e.g. Maternal and Paternal Bonding). There was a positive albeit small

**Table 1**  
Descriptive statistics of the CRP and cortisol samples (observed data).

Variables	Levels	CRP sample (N = 4198)		Cortisol sample (N = 3357)	
		Mean(sd)/ %	Missing (%)	Mean(sd)/%	Missing (%)
<b>Adverse Childhood Experiences (ACEs)</b>					
Physical abuse		3.2	15	3.5	14.9
Sexual abuse		5.3	15.1	6.1	15
Physical assault		2.7	15	2.4	14.9
Parent arguments		20.7	15.9	21.5	15.8
Low maternal bonding		17.1	19.5	18	20.1
Low paternal bonding		17.4	22.1	18	22.3
Institutionalisation		1.6	0.1	1.6	0.1
Separation from mother		14.6	0.2	15	0.1
Foster care/Adoption		1.8	0.1	1.8	0.1
Parent death		5.1	0.2	5.1	0.2
Parent mental illness/ substance abuse		6.5	15.3	7.1	15.1
Parent separation/divorce		5.9	0.2	6.4	0.2
ACEs cumulative score		0 = 64%, 1 = 18%, 2 = 9%, 3+ = 9%		0 = 65%, 1 = 17%, 2 = 9%, 3+ = 9%	
<b>Demographic and socioeconomic characteristics</b>					
Age		69.67(8.72)	0	69.79(9.10)	0
Sex: Female		56	0	67.9	0
Marital status	Married	69.3	0.1	68.3	1.2
	Separated/ Divorced	11.7		11.9	
Wealth quintiles	Windowed	13.4		14.6	
	Single	5.7		5.3	
	1 (lowest)	15.3	1.7	15.2	3
	2	18.2		17.8	
	3	20.4		19.7	
	4	22.2		22.3	
	5 (highest)	23.9		24.9	
<b>Childhood socioeconomic indicators</b>					
Overcrowding		19.2	3.8	19.2	4.1
No books when aged 10		25	4.3	22.6	4.7
Manual occupation (father)		1.1	0.2	1.1	0.3
Financial hardship		6.8	15.3	7.9	15.3
Parent unemployment		8	15.3	7.9	15.3
<b>Biomarkers</b>					
CRP log (w4) (< 10 mg/L)		0.481(0.94)	20		
CRP log (w6) (< 10 mg/L)		0.393(0.93)	15		
High CRP (w4 + w6) (≥ 3mg/L)		0	64.2		
	1	25.6			
	2	10.2			
Hair cortisol log (w6) (pg/mg)				0.896(0.576)	0
<b>Lifestyle indicators</b>					
BMI		28.21(5.09)	9	28.3(5.26)	5
Smoking	Current smoker	16.1	0.2	15.9	1.4
	Smoked in the past	43.3		42	
	Never smoked	40.6		42.1	
Physical activity	Light	14.7	1.1	16.2	2.2
	Moderate	51.1		50.2	
	Vigorous	34.2		33.2	
Alcohol consumption	5-7 days a week	24.4	10.7	23.5	11.4
	1-4 days a week	40		38	
	Less than weekly	35.5		38.5	
<b>Medications</b>					
Anti-inflammatory /antihypertensive drugs		48.2	0		
Steroids				4.8	0
<b>Hair characteristics</b>					
Hair colour	Blonde/ginger			16	0.6
	Brunette			23.5	
	Grey/white			60.5	

(continued on next page)

Table 1 (continued)

Variables	Levels	CRP sample (N = 4198)		Cortisol sample (N = 3357)	
		Mean(sd)/ %	Missing (%)	Mean(sd)/%	Missing (%)
Season hair collection	Autumn			43.4	0
	Spring			6.3	
	Summer			23.9	
	Winter			26.4	
Hair dyed/chemically treated			31.3	0	
Phase of hair analysis	2 (2018)			39.7	0
<b>Depressive symptoms</b>					
CESD-8 (mean)		1.31(1.85)	0.1	1.36(1.87)	0.1

Note. Sample = ELSA, w3-w6. Sd = standard deviation. ACEs = adverse childhood experiences. CRP = C-reactive protein. BMI = body mass index.

correlation ( $r = 0.04$ ,  $p = 0.076$ ) between hair cortisol and CRP in the subsample of participants with both measures available ( $N = 2,886$ ).

### 3.2. ACEs cumulative score

#### 3.2.1. Associations with CRP w4 and sustained high CRP w4-w6

The marginal effects of the ACEs cumulative score on CRP w4 and sustained high CRP w4-w6 are reported in Table 2 and illustrated in Fig. 1.a and 1.b. The first model indicated that the group of participants with 3 + ACEs was estimated to have a log CRP value of 0.19 points

Table 2

Associations of the ACEs cumulative score with CRP w4 and sustained high CRP w4 + w6.

Ref: ACE(0)	B	SE	P-Value	95% CI	E-value (lower CI)
<b>Outcome: CRP (w4)</b>					
<b>Model 1</b>					
ACE(1)	0.006	0.041	0.892	-0.074;0.086	
ACE(2)	-0.042	0.054	0.436	-0.148;0.064	
ACE(3 + )	0.190	0.056	0.001	0.080;0.300	1.69(1.38)
<b>Model 2</b>					
ACE(1)	0.000	0.040	0.999	-0.078;0.078	
ACE(2)	-0.043	0.054	0.426	-0.149;0.063	
ACE(3 + )	0.166	0.055	0.003	0.058;0.274	1.63(1.31)
<b>Model 3</b>					
ACE(1)	0.002	0.037	0.961	-0.071;0.075	
ACE(2)	-0.002	0.049	0.964	-0.098;0.094	
ACE(3 + )	0.151	0.052	0.003	0.049;0.253	1.58(1.28)
Ref: ACE(0)	OR	SE	P-Value	95% CI	E-value (lower CI)
<b>Outcome: High CRP (w4 + w6)</b>					
<b>Model 1</b>					
ACE(1)	1.117	0.016	< 0.001	1.081;1.153	1.30(1.24)
ACE(2)	1.087	0.021	< 0.001	1.044;1.132	1.25(1.17)
ACE(3 + )	1.494	0.020	< 0.001	1.437;1.553	1.74(1.69)
<b>Model 2</b>					
ACE(1)	1.082	0.017	< 0.001	1.047;1.118	1.24(1.18)
ACE(2)	1.049	0.021	0.023	1.007;1.092	1.18(1.06)
ACE(3 + )	1.385	0.020	< 0.001	1.332;1.442	1.63(1.58)
<b>Model 3</b>					
ACE(1)	1.049	0.017	0.005	1.014;1.085	1.18(1.09)
ACE(2)	1.071	0.022	0.001	1.027;1.118	1.22(1.13)
ACE(3 + )	1.352	0.021	< 0.001	1.297;1.409	1.60(1.54)

Note. CRP Sample = ELSA, w3-w6 (N = 4198). Pooled estimates from linear regression (CRP w4) and ordinal logistic regression (high CRP w4 + w6) with 20 multiple imputed datasets. Model 1: adjusted for sex, age, marital status, and use of anti-inflammatory/anti-hypertensive medications; Model 2: Model 1 + childhood and adult socioeconomic factors; Model 3: Model 2 + BMI, smoking, alcohol consumption, and physical activity. B = linear regression coefficient. OR = odds ratio. SE = standard error. CI = confidence interval.

higher than the reference group (i.e. those who did not experience any ACEs) independently of demographic characteristics and use of anti-inflammatory or anti-hypertensive medications (Model 1:  $b = 0.19$ , 95% CI = 0.08,0.30). Models 2 and 3 demonstrated that this association was robust to adjustment for childhood socioeconomic position and adult wealth (Model 2:  $b = 0.17$ , 95%CI = 0.06,0.27) as well as for lifestyle indicators including BMI, smoking, physical activity, and alcohol drinking (Model 3:  $b = 0.15$ , 95%CI = 0.05,0.25). In contrast, the predicted differences in CRP levels between participants with 1 or 2 ACEs and the reference group were not statistically significant in any of the models (Table 2, Fig. 1.a).

For sustained high CRP w4-w6, the marginal effects of the ACEs cumulative score were larger and statistically significant across all ACEs groups (Table 2, Fig. 1.b). In Model 1, the ordered odds of sustained high CRP across waves 4 and 6 for participants with 1, 2, or 3 + ACEs were respectively 1.12 (95%CI = 1.08,1.15), 1.09 (95%CI = 1.04,1.13), and 1.49 (95%CI = 1.44,1.55) times larger than for the reference group of no ACEs. These effects were also independent of childhood and adult socioeconomic characteristics (Model 2:  $b_{ACE(1)} = 1.08$ , 95%CI = 1.05,1.12;  $b_{ACE(2)} = 1.05$ , 95%CI = 1.01,1.09;  $b_{ACE(3+)} = 1.39$ , 95%CI = 1.33;1.44) and lifestyle indicators (Model 3:  $b_{ACE(1)} = 1.05$ , 95%CI = 1.01,1.09;  $b_{ACE(2)} = 1.07$ , 95%CI = 1.03,1.12;  $b_{ACE(3+)} = 1.35$ , 95%CI = 1.30,1.41). For both CRP outcomes, the interaction effects of the ACEs cumulative score with sex and age were almost null and did not reach statistical significance. Hence, they were not included in the analysis. The E-values suggested that very modest confounder associations could explain away the observed effects of the ACE(1) and ACE(2) groups. The effects of the ACE(3 + ) group were instead more robust since relatively larger confounder associations would be needed to fully explain them.

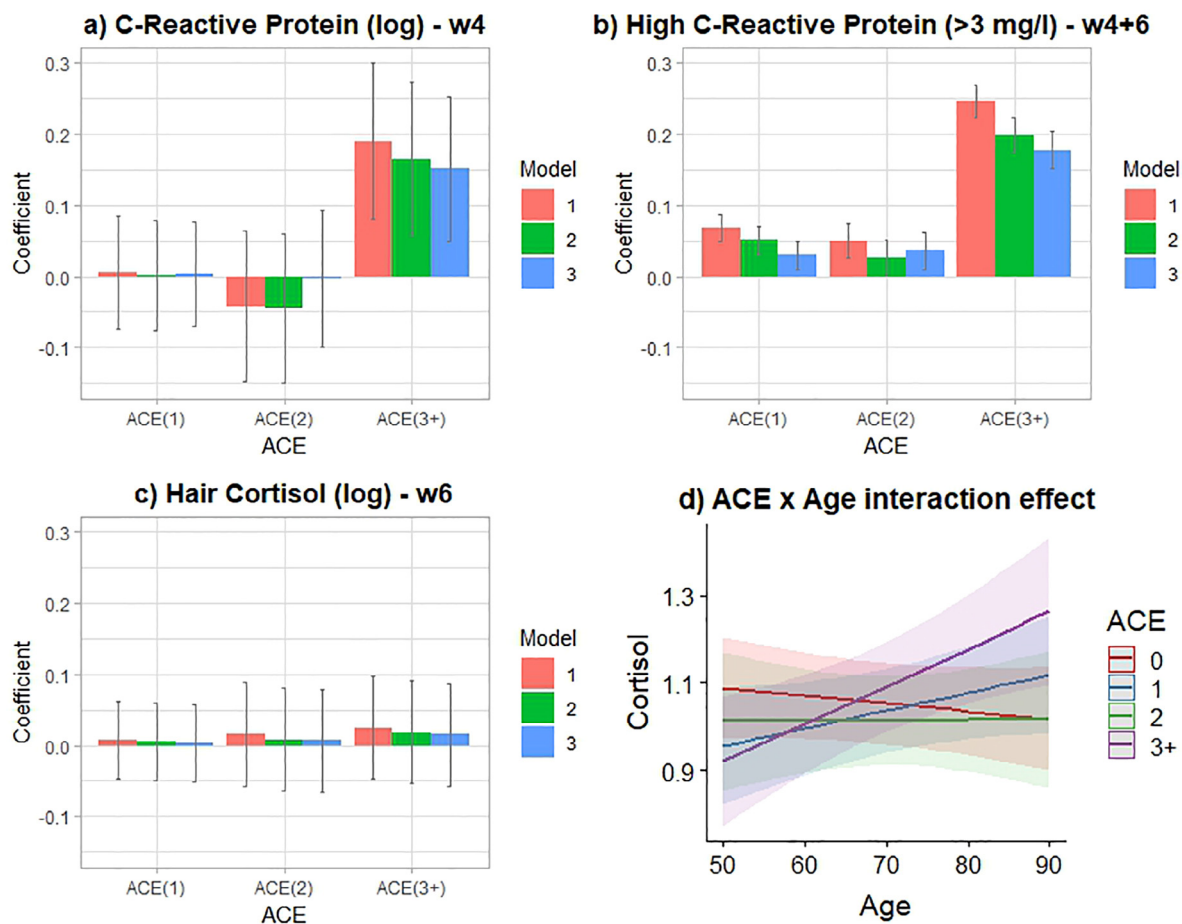
#### 3.2.2. Associations with hair cortisol

The marginal effects of the ACEs cumulative score on hair cortisol are reported in Table 3 and illustrated in Fig. 1.c. The expected differences in hair cortisol concentrations between participants without ACEs and those with 1, 2, or 3 + ACEs were non-significant across all models. However, we found a positive and statistically significant interaction effect between the ACE(3 + ) group and age. In contrast, there was no significant evidence for the interaction effect with sex. The interaction effect between the ACEs score and age remained statistically significant across all models. This result suggests that hair cortisol levels were expected to have a steeper increase with age in the group of participants with 3 + ACEs compared to those who did not experience any ACEs, independently of all covariates (Model 3:  $b = 0.01$ , 95%CI = 0.00,0.02) (Table 3, Fig. 1.d). Nevertheless, this effect was quite small, and the E-value indicated that very small unmeasured confounding would be necessary to fully explain it (Table 3).

### 3.3. ACEs dimensions

#### 3.3.1. EFA and CFA with cross-validation

The results of EFA of the ACEs items in the four training datasets are described in the Appendix and reported in Table A5. The results of the final CFA model fitted in the test dataset are illustrated in Fig. 2. The model fit the test dataset very well, RMSEA = 0.031, CFI = 0.954, TLI = 0.936. Standardised factor loadings ranged from medium (0.44) to high (0.86). All factors had good discriminant validity since their correlations were < 0.85 (Kenny, 2016). The first dimension (THREAT) was characterised by experiences related to threat, namely sexual abuse, physical abuse from parents, and physical assault. The second (HOUSEHOLD DYSFUNCTION) was related to dysfunctional household experiences including parent arguments, mental illness or substance abuse, and divorce or separation. A third dimension (LOW PARENTAL BONDING) emerged for maternal and paternal bonding experiences. The fourth dimension (LOSS) represented events related to loss of an



**Fig. 1.** Plots of the associations of the ACEs cumulative score with log CRP (w4), high CRP (w4 + w6), and hair cortisol (w6). Plots 1.a, 1.b, 1.c: ELSA, w3-w6 (CRP sample: N = 4198; Cortisol sample: N = 3357). Pooled estimates from linear regression (CRP w4, hair cortisol) and ordinal logistic regression (high CRP w4 + w6) with 20 multiple imputed datasets. Model 1: adjusted for sex, age, marital status, hair characteristics (cortisol only), and medication use; Model 2: Model 1 + childhood and adult socioeconomic factors; Model 3: Model 2 + BMI, smoking, alcohol consumption, and physical activity. Plot 1.d: Interaction effect on hair cortisol from linear regression with 20 multiple imputed datasets. Adjusted for all covariates (Model 3).

attachment figure including parent death, maternal separation, foster care or adoption, and institutionalisation. The adequacy of this dimensional model was finally evaluated across the twenty imputed datasets of the CRP and cortisol samples. The model had good discriminant validity and fit the data well in both samples (CRP sample: RMSEA = 0.038, CFI = 0.946, TLI = 0.926; Cortisol sample: RMSEA = 0.039, CFI = 0.949, TLI = 0.930).

### 3.3.2. Associations with CRP w4, high CRP w4-w6, and hair cortisol

The associations of the ACEs dimensions with CRP w4, high CRP w4-w6, and hair cortisol are reported in Table 4 and illustrated in Fig. 3. In Model 1, the four ACEs dimensions were all positively associated with CRP w4 independently of all covariates ( $\beta_{\text{Threat}} = 0.05$ ,  $p = 0.034$ ;  $\beta_{\text{HouseholdDysfunction}} = 0.06$ ,  $p = 0.022$ ;  $\beta_{\text{Bonding}} = 0.04$ ,  $p = 0.043$ ;  $\beta_{\text{Loss}} = 0.08$ ,  $p = 0.004$ ) (Table 4, Fig. 3). The same pattern of results was observed for the risk of high CRP levels across waves 4 and 6 ( $\beta_{\text{Threat}} = 0.07$ ,  $p = 0.003$ ;  $\beta_{\text{HouseholdDysfunction}} = 0.07$ ,  $p = 0.001$ ;  $\beta_{\text{Bonding}} = 0.07$ ,  $p = 0.001$ ;  $\beta_{\text{Loss}} = 0.09$ ,  $p < 0.001$ ), with larger effect sizes than for CRP w4 (Table 4). The Loss dimension exhibited the largest associations with both CRP outcomes. However, none of the observed differences amongst the distinct effects of the ACEs dimensions was statistically significant (Appendix, Table A12). The mutually adjusted effects of the ACEs dimensions were tested in Model 2. (Table 4). These results revealed that only the Loss dimension remained significantly associated with CRP w4 ( $\beta = 0.07$ ,  $p = 0.047$ ) and sustained high CRP w4-w6 ( $\beta = 0.09$ ,  $p = 0.009$ ) independently of

the other ACEs dimensions and the covariates. This result could be explained by the medium-sized correlations between the Threat, Dysfunctional Household, and Bonding dimensions. In contrast, the Loss dimension had much weaker correlations with the other ACEs dimensions (Fig. 2). As for the main effect of the cumulative score, the associations of the ACEs dimensions with hair cortisol were almost null and did not reach statistical significance (Table 4, Fig. 3).

### 3.4. Sensitivity analyses

The marginal effects of the covariates on the biomarkers can be found in the Appendix (Tables A6, A7). Sensitivity analyses indicated significant differences in socioeconomic and lifestyle characteristics between ELSA participants included in the analysis and those excluded due to missing ACEs or biomarker data. Nevertheless, none of these effects exceeded 0.2% (Appendix, Table A8). All models were rerun in the samples of participants with complete data on all measures. The results revealed very similar associations to those observed in the imputed datasets (Appendix, Tables A9, A10, A11). Likewise, the relationships between ACEs and the biomarkers did not change substantially when excluding only participants with CRP levels greater than 20 mg/L or when controlling for depressive symptoms in addition to the other covariates (Appendix, Tables A13, A14).

**Table 3**  
Associations of the ACEs cumulative score with hair cortisol (w6).

	B	SE	P-Value	95% CI	E-value (lower CI)
<b>Model 1</b>					
ACE(1)	0.007	0.028	0.797	−0.048;0.062	
ACE(2)	0.016	0.037	0.666	−0.057;0.089	
ACE(3+)	0.025	0.037	0.500	−0.048;0.098	
<b>Model 2</b>					
ACE(1)	0.005	0.028	0.851	−0.050;0.060	
ACE(2)	0.010	0.037	0.777	−0.063;0.083	
ACE(3+)	0.019	0.037	0.598	−0.054;0.092	
<b>Model 3</b>					
ACE(1)	0.003	0.028	0.904	−0.052;0.058	
ACE(2)	0.008	0.037	0.824	−0.065;0.081	
ACE(3+)	0.016	0.037	0.671	−0.057;0.089	
<b>Interaction effect: ACE × Age</b>					
<b>Model 1</b>					
ACE(1)*Age	0.003	0.003	0.354	−0.003;0.009	
ACE(2)*Age	0.001	0.004	0.894	−0.007;0.009	
ACE(3+)*Age	0.010	0.005	0.031	0.001;0.020	1.14 (1.02)
<b>Model 2</b>					
ACE(1)*Age	0.003	0.003	0.359	−0.003;0.009	
ACE(2)*Age	0.000	0.004	0.991	−0.008;0.008	
ACE(3+)*Age	0.010	0.005	0.032	0.000;0.020	1.14 (1.02)
<b>Model 3</b>					
ACE(1)*Age	0.003	0.003	0.350	−0.003;0.009	
ACE(2)*Age	0.000	0.004	0.956	−0.008;0.008	
ACE(3+)*Age	0.010	0.005	0.029	0.000;0.020	1.14 (1.02)

Note. Cortisol Sample = ELSA, w3-w6 (N = 3357). Pooled estimates from linear regression with 20 multiple imputed datasets. Model 1: adjusted for sex, age, marital status, hair characteristics, and use of steroids; Model 2: Model 1 + childhood and adult socioeconomic factors; Model 3: Model 2 + BMI, smoking, alcohol consumption, and physical activity. B = linear regression coefficient. SE = standard error. CI = confidence interval.

## 4. Discussion

### 4.1. Summary of main findings

Our results highlight a number of important findings. In relation to the ACEs cumulative score, we found elevated CRP values at wave 4 and higher CRP levels across waves 4 and 6 in participants reporting three or more ACEs compared with those who did not experience any ACEs, independently of demographic, socioeconomic, lifestyle, and medication confounders. Although the associations of the ACEs cumulative score with hair cortisol were not statistically significant, we found evidence for a positive interaction effect between ACEs and age on hair cortisol. This suggests that the predicted increase in hair cortisol with age was greater amongst participants with three or more ACEs compared to those without. All ACEs dimensions (i.e. Threat, Dysfunctional Household, Low Parental Bonding, and Loss of an Attachment Figure) were positively associated with CRP at wave 4 and risk of high CRP levels across waves 4 and 6. These associations were independent of all included confounders and had similar effect sizes. In contrast, none of the ACEs dimensions was significantly associated with hair cortisol.

### 4.2. Explanation of results in relation to previous findings

#### 4.2.1. ACEs cumulative score

The findings for CRP are in line with previous studies of adults indicating that cumulative exposure to ACEs may contribute to a pro-inflammatory state with higher levels of CRP and other inflammatory markers (Baumeister et al., 2016; Chen and Lacey, 2018; Pinto Pereira et al., 2019). Our results extend the current evidence base since they

demonstrate that the relationship between ACEs and the inflammatory system is likely to persist into later life. The majority of previous studies were based on younger samples whose average age did not exceed 50 years. In contrast, our sample had a mean age of 70 years thereby providing a more accurate representation of inflammatory processes in older people. We have also shown that the effects of ACEs are particularly strong when considering the risk of elevated inflammation over time. In addition, they are partly independent of both childhood and adult socioeconomic factors, as well as of relevant lifestyle factors. Elevated levels of inflammatory markers have been linked to numerous physical and mental health conditions (Acabchuk et al., 2017; Baumeister et al., 2014; Belvederi Murri et al., 2014; Girod and Brotman, 2004; Haapakoski et al., 2015; Hackett and Steptoe, 2017; Kivimäki and Steptoe, 2017). Therefore, taken together these different lines of evidence support the idea that the immune system might be one of the main psychobiological mechanisms underlying the pathogenesis of mental and physical illnesses in the context of early-life stress. Nevertheless, it is important to note that the evidence for elevated inflammation in children and adolescents exposed to ACEs is weak (Kuhlman et al., 2019). Levels of inflammation are typically low during childhood and have been shown to progressively increase with age through biological ageing processes that hinder effective regulation (Franceschi and Campisi, 2014; Ribeiro, 1997). Hence, the association between ACEs and elevated inflammation might not emerge before adulthood (Kuhlman et al., 2019).

To our knowledge, this is the first study to investigate the effects of ACEs on hair cortisol in a large sample of older adults. Results from previous studies of young people and middle-aged adults suggest that individuals exposed to ACEs tend to exhibit higher hair cortisol concentrations, although some studies have also reported negative associations (Khoury et al., 2019). In our analysis, we did not find evidence for the relationship between cumulative exposure to ACEs and hair cortisol. However, the positive interaction effect of ACEs with age suggests that having at least three ACEs is associated with a greater increase in hair cortisol with age. As for CRP, concentrations of cortisol in hair have been shown to increase in an age-dependent fashion and are related to a number of biological changes underlying the ageing process (Feller et al., 2014). Thus, exposure to multiple ACEs could accelerate biological ageing and lead to a greater increase in cortisol levels, which in turn may augment risk of poor mental and physical health (Belvederi Murri et al., 2014; Girod and Brotman, 2004). Nevertheless, this result must be interpreted with caution since the interaction effect between ACEs and age was small and particularly prone to possible unmeasured confounding. On the other hand, two studies found evidence for lower morning salivary cortisol levels in older adults who experienced childhood abuse (Gerritsen et al., 2010; Wielaard et al., 2018). Thus, ACEs could have larger associations with other aspects of HPA-axis function than with total cortisol output over time in older adults. The small association between ACEs and hair cortisol could also be explained in view of evidence suggesting that recent adversities might produce greater alternations in hair cortisol than more distal events (Khoury et al., 2019).

#### 4.2.2. ACEs dimensions

Our results are partially consistent with the existing literature on the dimensionality of ACEs. As expected, the *threat-deprivation* theme emerged also in our analysis. However, we found evidence for other distinct dimensions of psychosocial adversity which align closely with the results of other studies using data reduction techniques (Germine et al., 2015; Westermair et al., 2018; Zhang et al., 2019). A number of investigations demonstrated that ACEs involving *threat* but not *deprivation* are associated with salivary cortisol (Busso et al., 2017; LoPilato et al., 2019), as well as with other physical and mental health outcomes (Machlin et al., 2019; Sumner et al., 2019). However, other studies have suggested that childhood neglect is particularly important for cortisol abnormalities (Koss and Gunnar, 2018; White et al., 2017),

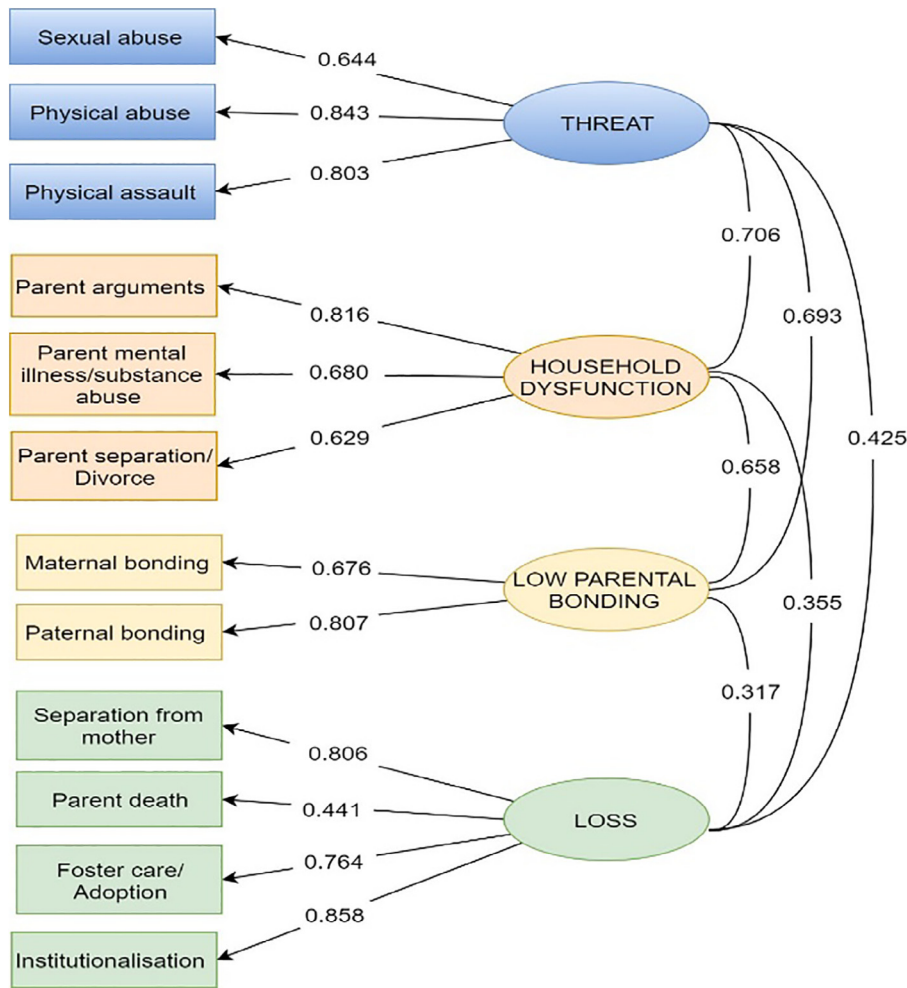


Fig. 2. CFA of the ACEs items in the training dataset. Sample = ELSA, Life History Interview w3 (N = 1571). Estimator = WLSMV. All coefficients are standardised and statistically significant (P-value < 0.001).

whereas elevated CRP levels might be primarily related to parental absence (Baumeister et al., 2016). In our study, all ACEs dimensions were positively associated with CRP and had similar effect sizes. These results do not support our hypothesis for differential associations

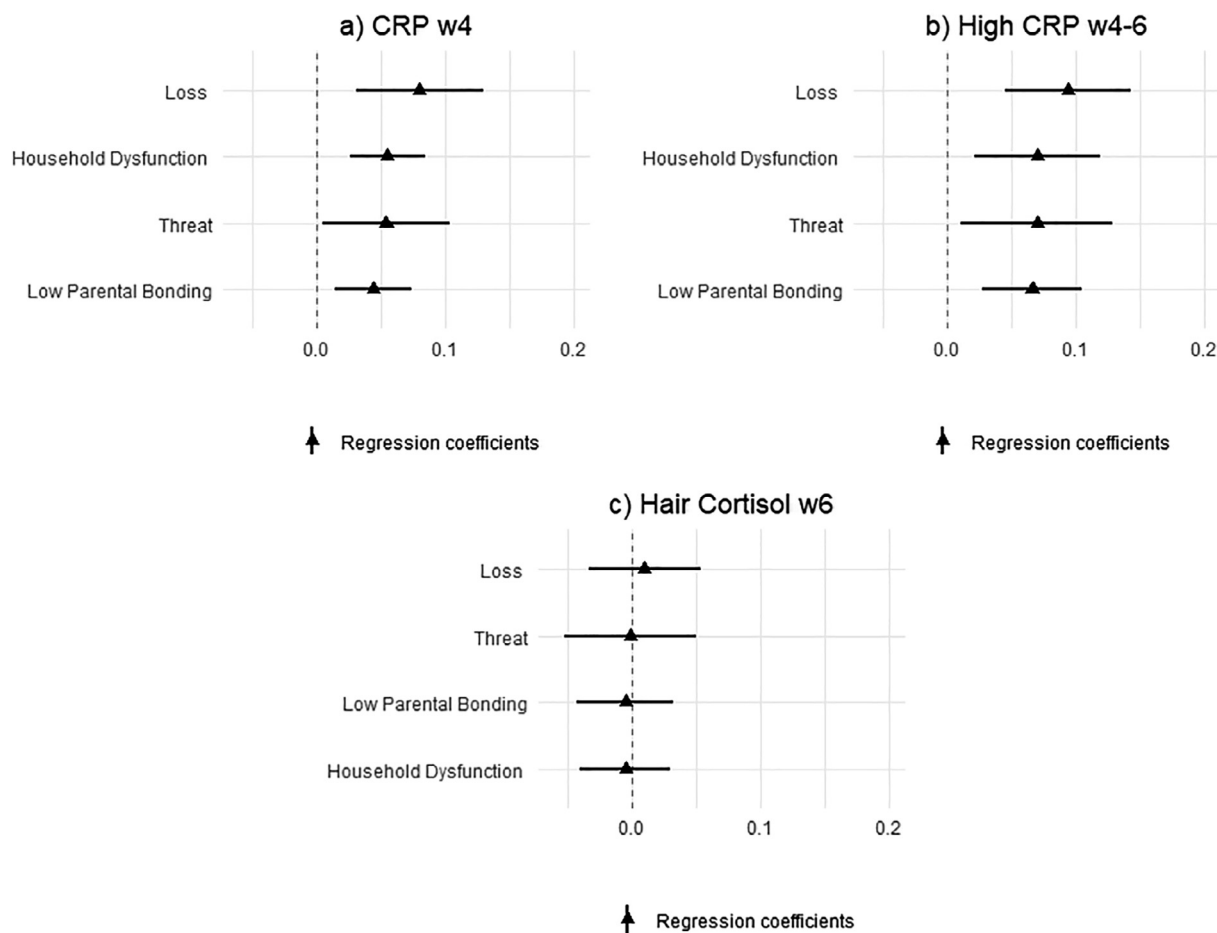
between distinct ACEs dimensions and the biomarkers. Notwithstanding this, our ACEs dimensional model makes an important contribution to the current ACEs measurement approach suggesting that all types of ACEs assessed in this study are related to elevated

**Table 4**  
Associations of the ACEs dimensions with CRP w4, sustained high CRP w4 + w6, and hair cortisol w6.

	Model 1				Model 2			
	B	SE	P-Value	$\beta$	B	SE	P-Value	$\beta$
<b>Outcome: log CRP w4 (N = 4198)</b>								
Threat	0.105	0.050	0.034	0.054	-0.099	0.134	0.462	-0.051
Household Dysfunction	0.061	0.027	0.022	0.055	0.057	0.056	0.312	0.050
Low Parental Bonding	0.059	0.029	0.043	0.044	0.013	0.062	0.835	0.010
Loss of an attachment figure	0.097	0.034	0.004	0.080	0.087	0.044	0.047	0.071
<b>Outcome: Sustained high CRP w4 + w6 (N = 4198)</b>								
Threat	0.098	0.033	0.003	0.070	-0.159	0.098	0.102	-0.115
Household Dysfunction	0.056	0.017	0.001	0.070	0.033	0.039	0.397	0.042
Low Parental Bonding	0.062	0.019	0.001	0.066	0.080	0.044	0.069	0.085
Loss of an attachment figure	0.082	0.022	0.000	0.094	0.074	0.028	0.009	0.085
<b>Outcome: log Hair Cortisol w6 (N = 3357)</b>								
Threat	-0.001	0.026	0.976	-0.001	0.065	0.077	0.401	0.066
Household Dysfunction	-0.003	0.018	0.843	-0.005	-0.038	0.043	0.387	-0.053
Low Parental Bonding	-0.004	0.019	0.842	-0.005	-0.022	0.042	0.592	-0.028
Loss of an attachment figure	0.008	0.022	0.708	0.010	0.020	0.028	0.477	0.025

Note. ELSA, w3-w6; CRP sample: N = 4198; Cortisol sample: N = 3357. Pooled estimates from SEM with 20 multiple imputed datasets. Model 1: adjusted for sex, age, marital status, hair characteristics (cortisol only), medication use, childhood and adult socioeconomic factors, BMI, smoking, alcohol consumption, and physical activity. Model 2: Model 1 + mutual adjustment of ACEs dimensions. B = regression coefficients. SE = standard error. CI = confidence interval.  $\beta$  = standardised regression coefficient. Estimator = WLSMV.





**Fig. 3.** Associations of the ACEs dimensions with log CRP (w4), high CRP (w4 + w6), and hair cortisol (w6). ELSA, w3-w6 (CRP sample: N = 4198; Cortisol sample: N = 3357). Pooled standardised estimates from linear regression (CRP w4, hair cortisol) and ordinal logistic regression (high CRP w4 + w6) with 20 multiple imputed datasets. Model 1: adjusted for sex, age, marital status, hair characteristics (cortisol only), medication use, childhood and adult socioeconomic factors, BMI, smoking, alcohol consumption, and physical activity.

inflammation in older adults.

#### 4.3. Strengths and limitations

Our analysis has several strengths. These include: 1) a population-based sample of older adults not selected on the basis of adversity exposure or health problems and therefore more representative of the general population; 2) a sample size that is substantially larger than most previous studies – e.g. the recent *meta-analysis* of ACEs and hair cortisol included 28 studies with a total sample of only 3,397 participants (Khoury et al., 2019); 3) a more robust assessment of pro-inflammatory state and long-term HPA-axis activity owing to the use of repeated measures of CRP and quantification of cortisol in human hair; 4) the assessment of the effects of cumulative exposure versus dimensions of ACEs; and 5) a more refined measurement model of ACEs dimensions tested using both EFA and CFA and evaluated through cross-validation.

Notwithstanding this, a number of limitations should be noted. First, our ACEs measures were based on retrospective reports which are particularly prone to measurement error due to motivation of participants, personality styles, and memory biases, particularly at older ages (Hardt and Rutter, 2004). Accordingly, comparisons of prospective and retrospective measures of ACEs have indicated low to modest agreement (Baldwin et al., 2019; Patten et al., 2015). Additionally, we did not have any information of the frequency or timing of adversity exposure. Second, we only considered a single biomarker for each biological system. Nevertheless, CRP and cortisol are the biological

measures that have been more consistently linked to adversity exposure. Moreover, other biological processes such as neurocognitive and epigenetic changes are also likely to be involved in the relationship between ACEs and health. Third, data on hair cortisol, CRP, and ACEs was not available for a large number of ELSA participants who did not take part in the Life History interview or the nurse visits. Importantly, these participants had worse socioeconomic and lifestyle characteristics compared to those included in the analytical samples. Fourth, despite the use of cross-validation, our empirically driven dimensional model of ACEs could be specific to this dataset. Fifth, there was a considerable time gap between the collection and the analysis of the hair samples (3–6 years). This could affect the relationship of hair cortisol with other variables since previous research has reported a negative association between length of storage and hair cortisol concentration (Abell et al., 2016). Nevertheless, we accounted for this possibility by including a covariate representing the phase of hair cortisol analysis. Lastly, it is important to note that the observational design of our study does not provide evidence for the causal effect of ACEs on inflammation owing to possible unmeasured confounding effects.

#### 4.4. Suggestions for further research

Further research is required to measure ACEs dimensions in different datasets and test their effects on CRP and cortisol levels using repeated measures of the biomarkers. In addition, future studies should replicate these results using prospective data on ACEs and taking into account the timing of adversity exposure. Given the relatively robust

associations of ACEs with CRP, it will also be important to formally test the plausible mediational role of CRP in the pathways leading from ACEs to poor physical and mental health. Moreover, further research should seek to determine how the interplay between ACEs and genetic factors might influence trajectories of biomarkers and other health outcomes across the life course.

#### 4.5. Conclusion

To conclude, our study suggests that the relationship between ACEs and pro-inflammatory responses is likely to persist into later life. Older adults with three or more ACEs had an elevated risk of high CRP levels both cross-sectionally and across a 4-year period. All ACEs dimensions (i.e. Threat, Dysfunctional Household, Low Parental Bonding, and Loss of an Attachment Figure) were associated with increased CRP levels and had similar effect sizes. Given the important role of inflammation in the pathogenesis of several mental and physical health conditions, the immune system might represent one of the main psychobiological mechanisms underlying the relationship of early-life stress with poor health and disease in adulthood. In contrast, the evidence for the relationship of the cumulative score or dimensions of ACEs with hair cortisol was particularly weak. However, ACEs were associated with a slightly greater increase in cortisol levels linked to biological ageing. These findings have important implications for childhood policies and interventions. In particular, they highlight the importance of improving family and mental health services for the prevention and treatment of the adverse effects of ACEs on mental and physical health (Step toe et al., 2019). In addition, psychosocial interventions to manage the impact of ACEs on children's lives and reduce stress levels in families may have beneficial effects on the individual's susceptibility to poor health and disease throughout the lifespan.

#### 5. Contributors

All authors contributed significantly to the conception, design, analysis or interpretation of data and were involved in revising it critically for intellectual context. The final submission of this paper was approved by all authors.

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#### Conflict of interest

None of the authors has any conflict of interest to declare related to the findings of this study.

#### Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.bbi.2019.12.019>.

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