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Acute stress impairs recall after interference in older people, but not in young people

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ABSTRACT

Stress has been associated with negative changes observed during the aging process. However, very little research has been carried out on the role of age in acute stress effects on memory. We aimed to explore the role of age and sex in the relationship between hypothalamus–pituitary–adrenal axis (HPA-axis) and sympathetic nervous system (SNS) reactivity to psychosocial stress and short-term declarative memory performance. To do so, sixty-seven participants divided into two age groups (each group with a similar number of men and women) were exposed to the Trier Social Stress Test (TSST) and a control condition in a crossover design. Memory performance was assessed by the Rey Auditory Verbal Learning Test (RAVLT). As expected, worse memory performance was associated with age; but more interestingly, the stressor impaired recall after interference only in the older group. In addition, this effect was negatively correlated with the alpha-amylase over cortisol ratio, which has recently been suggested as a good marker of stress system dysregulation. However, we failed to find sex differences in memory performance. These results show that age moderates stress-induced effects on declarative memory, and they point out the importance of studying both of the physiological systems involved in the stress response together.

Introduction

Stress has been suggested as a main factor related to negative changes observed during the aging process. However, little is known about the role of age in acute stress effects on memory performance. Given that there are data suggesting age differences in the reactivity to stress, the need to obtain evidence to fill this gap in the literature seems clear.

Stress, particularly, provokes the activation of two systems: (i) the sympathetic nervous system (SNS) and (ii) the hypothalamus–pituitary–adrenal axis (HPA-axis). The fast SNS response includes the release of the catecholamines (adrenaline and noradrenaline), which are responsible for different physiological changes preparing the organism for a "fight-or-flight" response. Minutes after the onset of the stressor, HPA-axis activation occurs and, consequently, large amounts of glucocorticoids are secreted in the adrenal cortex. There are numerous glucocorticoid receptors in the brain areas involved in the memory process, such as the hippocampus, the frontal lobe and the amygdala (Lupien and Lepage, 2001; Lupien et al., 2009; Roozendaal, 2000), which also play an important role in the regulation of the HPA-axis (Herman et al., 2005; Lupien and Lepage, 2001). Thus, cortisol, the main glucocorticoid hormone in humans, would have important effects on memory, although the direction of these effects remains unclear. They can differ depending on several factors, some related to the task (such as the type of memory or the nature of the material, neutral or emotional) and others associated with characteristics of the individual (including age and sex). In addition, it has been well established that SNS activation can also affect memory performance through the influence of cathecolamines on the limbic brain structures. According to Roozendaal et al. (2009), the noradrenergic activation of the amygdala and the interactions between the amygdala and hippocampus are crucial to finding cortisol effects on hippocampus-dependent memory performance.

The majority of studies about the relationship between the exposure to an acute stressor and memory have been performed on declarative memory in young people, reporting mixed results. When subjects have to learn neutral material after stress induction, worsening effects (Jelicic et al., 2004; Kirschbaum et al., 1996; Payne et al., 2006, 2007; Smeets et al., 2006), enhancing effects (Espin et al., 2013; Schwabe et al., 2008), and even a lack of effects (Hidalgo et al., 2012; Wolf et al., 2001b) have been described in mixed-sex samples. When studying only one sex, enhancing effects were found in young men (Nater et al., 2007), but non-effects were detected in women when they were grouped without taking age into account (32–68 years) (Domes et al., 2002). Bohnen et al. (1990) compared two groups of women (41–49 vs. 61–69 years) exposed to a 4-hour mental task, finding no significant differences.

To our knowledge, only Wolf et al. (2001a) have investigated the pre-learning cortisol effects on short-term memory considering the role of age by directly comparing young and older people, specifically

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men. These authors reported that cortisol did not influence the recall of a list of neutral words learned after they injected a cortisol agonist (hydrocortisone). However, there are important differences between the glucocorticoid increases induced by pharmacological administration and those produced by exposure to stress. As mentioned above, in addition to the cortisol increase that occurs with drug administration, stress provokes other physiological (i.e. SNS activation) changes (Lupien and Schramek, 2006). Hence, the use of stress paradigms in the laboratory allows a more complete study of stress effects on memory performance. In recent years, SNS activation has been measured by means of the salivary alpha-amylase (sAA), an oral enzyme secreted by the salivary glands (mainly parotid glands) due to parasympathetic and sympathetic nerve stimulations innervating the salivary glands. sAA is involved in converting starch into glucose and maltose in the oral cavity (Baum, 1993), eliminating bacteria from the mouth, and preventing bacterial attachment to oral surfaces (Scannapieco et al., 1993). A growing body of literature considers sAA to be a sensitive biomarker for stress-related changes in the body reflecting sympathetic nervous system activation (Granger et al., 2007; Nater and Rohleder, 2009; Rohleder and Nater, 2009). Moreover, as it is readily accessible and easily obtained, sAA is a good surrogate for catecholamines in psychoneuroendocrinological research.

Reactivity to stress changes throughout the lifespan; while the role of age in the cortisol response has been investigated more extensively, with most studies reporting that older people have a higher cortisol response than young people (for a review see: Kudielka et al., 2009), for the sAA response, results are fewer and mixed (Almela et al., 2011b; Strahler et al., 2010). Thus, the HPA-axis and the SNS activity could influence memory performance differently as a function of age. Furthermore, since both the HPA-axis and the SNS work in alliance to generate the stress response, in addition to the action of each system separately, it seems logical to study the two systems concurrently. According to Bauer et al. (2002), to obtain an optimal adaptation to stress, a coordinated response of the two stress systems is necessary. Thus, an uncoordinated response could mean a maladaptive response related to health or behavior problems. Studies examining this relationship in children and adolescents have suggested its value in predicting individual differences in behavioral adjustments to stress (Allwood et al., 2011; El-Sheikh et al., 2008; Gordis et al., 2006, 2008; Vigil et al., 2010). Recently, a few studies have focused on the effects of stress on cognitive functioning and even academic achievement (Berry et al., 2012; Keller et al., 2012); however, as mentioned above, these interactions, and specifically their potential effects on cognitive performance, have not been studied in young and older people.

With all this in mind, the purpose of the present study is to investigate age-related differences in memory performance in response to acute psychosocial stress, taking into account the sex and the relationship between the two stress systems, the HPA-axis and the SNS. No previous studies have been published on the influence of an acute laboratory social stressor on declarative memory in young and older people of both sexes. Previously, we reported stress effects on declarative memory in older people, especially in post-menopausal women (Almela et al., 2011a), but not in young people (Hidalgo et al., 2012). Based on these results, in the present study we have directly compared two different age samples employing the same protocol, a statistically different approach, and both stress markers (cortisol and sAA), in order to examine the different effects of stress on declarative memory depending on age or sex. The present study compares sixty-seven healthy participants divided into two age groups, 35 young adults and 32 older adults, with a similar number of men and women in each group. All the older women were postmenopausal, and all the young women were in the early follicular phase of their menstrual cycle, that is, the period with lower sex hormone levels. In a crossover design, the participants were exposed to both psychosocial stress (Trier Social Stress Test, TSST; Kirschbaum et al., 1993) and a control condition. In each condition, declarative memory performance was measured after the task. Previous studies employing a limited age range (41–49 vs. 61–69 years) and a 4-hour mental stressor in women (Bohnen et al., 1990) or cortisol administration in men (Wolf et al., 2001a) did not find age-related differences in stress/cortisol effects on declarative memory. However, we think that with a broader age range and a psychosocial stress task as the stressor, age differences would appear in the stress effects on declarative memory. To test this, we directly compared two age groups (18–35 years vs. 54–78 years) containing men and women, and we employed the TSST, which provokes both HPA-axis and SNS activation. In addition, we investigated stress reactivity by combining the two main stress physiological systems, considering that the imbalance between the two systems (an uncoordinated response) could prejudice memory performance. Finally, since sex differences have been reported in the effects of stress on memory in older people, greater negative stress effects were expected in older women.

Method

Participants

This study is part of extensive research on the moderating role of age and sex in the effects of acute stress on memory. Partial results from the older (Almela et al., 2011a) and young (Espin et al., 2013; Hidalgo et al., 2012) participants have been previously published. Here, we employed a subsample to directly compare the stress effects on declarative memory, taking into account the age and sex factors.

The final sample employed was composed of sixty-seven participants divided into two age groups (older adults: N = 32; 16 men and 16 women; young adults N = 35; 18 men and 17 women). There were no differences between the two age groups with regard to sex, in subjective socioeconomic status (SES) or educational level, but there were differences in body mass index (BMI), with young men showing a higher BMI than young women ($p = 0.047$) (Table 1). SES was measured using the MacArthur Scale of Subjective Social Status (Adler et al., 2000). Subjects were asked to rate themselves according to their subjective socioeconomic status and compared to other people in Spain, on a scale ranging from 1 (people with the lowest education, income and worst jobs) to 10 points (people with the best education, income and jobs).

The older participants belonged to a study program at the University of Valencia for people over 50 years of age (NAU GRAN). We chose this University Program to increase the homogeneity of the sample and the likelihood of getting healthy volunteers to compare with young people.

Table 1
Descriptive statistics (mean ± SEM of young ($N = 35$) and older groups ($N = 32$).*SES: Subjective Socio-Economic Status Scale, ranging from 1 (lowest SES) to 10 (highest SES) (Adler et al., 2000). **Range: 0 = no studies, 1 = primary school, 2 = secondary education, 3 = university and higher education, 4 = postgraduate (Master, PhD).

	Young group			Older group		
	Total	Men	Women	Total	Men	Women
Age (years)	21.1 (0.7)	22.1 (1.2)	20.0 (0.7)	62.1 (0.8)	60.5 (1.2)	63.7 (1.1)
BMI (kg/m^2)	23.0 (0.5)	23.9 (0.7)	21.9 (0.7)	26.5 (0.5)	27.0 (0.5)	26.0 (1.0)
SES*	6.3 (0.1)	6.4 (0.2)	6.1 (0.2)	6.0 (0.2)	6.1 (0.3)	5.9 (0.3)
Education level**	2.3 (0.1)	2.5 (0.2)	2.2 (0.1)	2.8 (0.2)	2.7 (0.3)	2.9 (0.2)

Most of the young people were college students from different areas. The sample was recruited using informative talks and posters at the faculties of the University campus. Two hundred and seventy-two volunteers (113 older and 159 young subjects) were interviewed by phone and completed a general questionnaire to check whether they met the study prerequisites. The criteria for exclusion were: smoking more than 5 cigarettes a day, alcohol or other drug abuse, dental, visual or hearing problems, presence of cardiovascular, endocrine, neurological or psychiatric disease, and presence of a stressful life event during the past year. Participants were excluded if they were using any medication directly related to emotional or cognitive function, or one that was able to influence hormonal and sAA levels, such as glucocorticoids, β-blockers, antidepressants, benzodiazepines, asthma medication, thyroid therapies, psychotropic substances or contraceptives. Two hundred and five volunteers (81 older and 124 young volunteers) were eliminated for two reasons: (i) meeting the exclusion criteria, and/or (ii) incompatibility with the experiment's schedules.

All the older women were postmenopausal, having had their last menstrual period at least four years before, and none of them were receiving estrogen replacement therapy. All the young women were regular free-cycling and in the early follicular phase (2–5 days) of their menstrual cycle. The menstrual cycle phase was determined using a questionnaire (included in the general questionnaire) about the regularity and length of the menstrual cycle as well as the bleeding during the last year. Then, taking the day of onset of the last menstruation and the average length of the cycles as the reference, we estimated the day of onset of the next menstruation, and this was also verified by phone. Thus, we established the day of the appointment at the laboratory as the second to the fifth day after the onset of the new menstrual cycle.

The participants meeting the criteria were contacted by telephone and asked to attend two sessions that took place in a laboratory at the Faculty of Psychology. Before each session, participants were asked to maintain their general habits, sleep as long as usual, refrain from heavy physical activity the day before the session, and not consume alcohol since the night before the session. Additionally, they were instructed to drink only water, refrain from eating, smoking or taking any stimulants, such as coffee, cola, caffeine, tea or chocolate, two hours prior to the session, and not brush their teeth at least one hour prior to the session. The study was conducted in accordance with the Declaration of Helsinki, and the protocol and conduct were approved by the Ethics Research Committee of the University of Valencia. All the participants received verbal and written information about the study and signed an informed consent form.

Procedure

This study used a within-subject design with two completely randomized and counterbalanced conditions, a stress condition and a control condition, in two separate sessions with less than 10 days between them. The sessions consisted of several phases of equal durations in both conditions. Sessions took 1 h and 50 min to complete, and they were always held between 16.00 and 20.00 h. Each participant started his or her two sessions at the same hour. Upon arrival at the laboratory, the weight and height of the participants were measured (first session), and the experimenter checked to see whether they had followed the instructions given previously (both sessions).

Stress condition

To produce stress, we subjected the participants to the TSST. The stress tasks consisted of 5 min of free speech (job interview) and a 5 min arithmetic task, performed in front of a committee composed of a man and a woman. The participants remained standing at a distance of 1.5 m from the committee. Additionally, a video camera and a microphone were clearly visible. Both the speech and arithmetic tasks were filmed.

The protocol started with a habituation phase of 15 min to allow the participants to adapt to the laboratory setting. During this phase, the participants remained seated. After the habituation phase, the introduction phase started (duration 5 min). In this phase, the participants were informed about the procedure for the stress task. They received the instructions in front of the committee in the same room where the task took place. Next, the participants had 10 min to prepare for the task at hand. Following the preparation phase, the stress task was carried out. Then, subjects had 20 min to recover after the stress task. Each participant performed a standardized memory test consisting of 8 trials (Rey Auditory Verbal Learning Test, RAVLT), in order to measure declarative memory. The participants completed the first six trials between 30 to 40 min after the beginning of the TSST. After trial 6, they waited 30 min (delay period) before continuing with the memory test. After the delay period, they finished the memory test with trials 7 and 8 and, finally, were debriefed.

Taking into account the different time courses of the cortisol and sAA responses to stress induction, we collected the saliva samples for each of them at different moments. To measure cortisol, we collected four saliva samples, two before the stress task and two after the stress task. Specifically, the first saliva sample to measure cortisol was taken during the habituation phase, 10 min after the participant's arrival at the laboratory (−20 min pre-stress), and the second cortisol sample was taken during the preparation phase (−5 min pre-stress). The third and fourth cortisol samples were collected 15 (+15 min post-stress) and 40 (+40 min post-stress) min, respectively, after the onset of the stress task. To measure sAA, we collected five saliva samples, two before the task and three after it. Thus, the first saliva sample was collected 10 min before the onset of the stress task (−10 min pre-stress), and the second one was taken immediately before the onset of the speech (0 min). The third, fourth and fifth saliva samples were collected 5, 10 and 14 min after the onset of the stress task (after speech, +5 min; after arithmetic task, +10 min; +14 min post-stress, respectively).

Control condition

The control condition was similar to the experimental condition, except that the stressful task was replaced by a control task. This task was designed to be similar to the stress task in mental workload and global physical activity (Het et al., 2009), but without the main components capable of provoking stress, such as evaluative threat and uncontrollability (Dickerson and Kemeny, 2004). The control task was composed of 5 min of reading aloud and 5 min of counting. In the preparation phase, the participants read a book with neutral content. The timing of the saliva samples and the phase durations were the same for the two conditions.

Memory

Declarative memory

To measure declarative memory, the Spanish version of Rey's Auditory-Verbal Learning Test (RAVLT) was used (Miranda and Valencia, 1997). This test has several versions, and for each participant a different version of the RAVLT was used in the second session to avoid learning effects. The order of the two versions was randomized and counter-balanced. The RAVLT is composed of different trials. In the first five trials the experimenter read aloud a target list of 15 neutral words, and each participant had to repeat as many words as possible in each of the five trials. The performance on these first five trials reflects the rate of learning (Trials 1 to 5: *Learning curve*). After Trial 5, the experimenter read aloud an interference list of 15 words and tested the retention of these new words. Following this step, participants were asked to recall the words from the target list (Trial 6: *Recall after interference*); after a delay of 30 min, they had to recall them a second time (Trial 7: *Delayed recall*). In Trial 8 (*Recognition*), participants had to recognize the memorized words from a verbally-presented list containing 15 new and 15 previously learned words. Trial 8 was divided

into two different scores: *Hits*, the number of words correctly recognized as being on the target list; and *False alarms*, the number of words incorrectly recognized as being on the target list. To analyze the effects on recognition (Trial 8), we used d-prime (d′), which is the difference between the standardized proportion of correct hits and the standardized proportion of false alarms. One older woman (due to problems in the application of the memory test) and one young man (an outlier for memory outcomes) were removed from the statistical analyses for memory.

Biochemical analyses

Cortisol

Participants provided four saliva samples by depositing 5 ml of saliva in plastic vials. They took no more than 5 min to fill each vial. The samples were frozen at −80 °C until the analyses were performed. The samples were analyzed by a competitive solid phase radioimmunoassay (tube coated), using the commercial kit Coat-A-Count C (DPC, Siemens Medical Solutions Diagnostics). Assay sensitivity was 0.5 ng/ml. For each subject, all the samples were analyzed in the same trial. The within and inter assay variation coefficients were all below 8%. Five people (one older man, two young men and two young women) were excluded from the statistical analyses for cortisol because they were multivariate outliers on the basis of the $p < 0.001$ criteria for the Mahalanobis distance in cortisol samples.

Alpha-amylase (sAA)

Saliva was collected using salivettes (Sarstedt, Nümbrecht, Germany). Participants were instructed to introduce the cotton swab into their mouths for exactly 1 min, not chew the cotton, and move the swab around in a circular pattern to collect saliva from all the salivary glands (Rohleder and Nater, 2009). The samples were frozen at −20 °C after the completion of the session, until the analyses took place. The samples were shipped to Dresden and analyzed at the Kirschbaum lab, Technical University of Dresden. Concentration of alpha-amylase in saliva was measured by an enzyme kinetic method, according to the protocol specified in Rohleder et al. (2006). The lowest detectable concentration in our assay was 1.56 U/ml. Inter- and intra-assay variation was below 10%. Analyses of sAA failed to detect the sAA concentrations in the samples of two men, one young and one older, and one older woman; therefore, these subjects were eliminated from the sAA statistical analyses.

Statistical analyses

Data were checked for normal distribution and homogeneity of variance using Kolmogorov–Smirnov and Levene's tests before the statistical procedures were applied. Since neither the cortisol nor the sAA data had a normal distribution, they were square root transformed. Student's *t*-tests were used to investigate age and sex differences in the demographic variables.

We used linear mixed modeling to assess the salivary cortisol and sAA responses in both the stress and control conditions. As an estimation method, we used the restricted maximum likelihood procedure, since this procedure deals with outliers better (Diggle, 1988). As the dependent variable, we included either sAA or cortisol levels. To allow for differences in patterns between and within participants, we included random components for moment (cortisol: 4 saliva samples, sAA: 5 saliva samples) and for each subject. To analyze salivary cortisol and sAA levels, we added the following factors: (i) Time (for cortisol: −20 min, −5 min, +15 min, +40, and for sAA: −10 min, 0 min, +5 min, +10 min, +14 min), (ii) Condition (control, stress), (iii) Sex (man, woman), and (iv) Age (old, young).

We also used linear mixed modeling to assess memory performance. We performed separate analyses for the following indices: (i) learning curve, (ii) total learning, (iii) recall after interference or retroactive interference, (iv) delayed recall performance, and (v) recognition. As the dependent variable, we included the number of words remembered. We included random components for trial (Trials 1–5) and for each subject. Furthermore, we included the following factors: (i) Trial (learning curve: Trial 1 to Trial 5, total learning: $\sum$ Trial 1 to Trial 5, recall after interference: Trial 6, delayed recall performance: Trial 7, and recognition: Trial 8), (ii) Condition (control; stress), (iii) Sex (man, woman), and (iv) Age (old, young).

For all linear mixed models, we started with the most complex model containing all possible interactions, and then progressively removed non-significant effects, starting with the most complex effects. After removing a factor, we investigated whether the model's fit improved according to Akaike's Information Criterion (AIC) and Schwarz's Bayesian Information Criterion (BIC). See Appendix A for the results of these analyses for salivary cortisol (see Table 1), sAA (see Table 2) and memory (see Tables 3, 4, 5, and 6). To calculate AIC and BIC, the maximum likelihood procedure in SPSS was used because it gives more reliable estimates than the restricted maximum likelihood procedure. A lower value of at least 2 on one or both criteria was considered a better model (Burnham and Anderson, 2004).

In order to find out the possible order effects of session (whether the stress or control condition was first), we included this variable in each linear mixed model described above. Results did not show order effects in any model (all $p > 0.113$).

We calculated the cortisol reactivity and sAA reactivity to stress by subtracting the baseline levels from the sample taken immediately after stress, and then we obtained the ratio variable of cortisol over sAA by dividing the cortisol reactivity to stress by the sAA reactivity to stress (RCA). Furthermore, the ratio of sAA over cortisol was calculated by dividing the sAA reactivity to stress by the cortisol reactivity to stress (RAC). Pearson's correlations were performed to assess the relationships between cortisol reactivity and sAA reactivity and the two ratios (RCA and RAC) with memory performance (Trial 6 outcome). In addition, Fisher's Z tests were used to test significant differences between correlation coefficients.

For post hoc planned comparisons, we employed the Bonferroni correction. All *p*-values reported are two-tailed, and the level of significance was marked at <0.05. When not otherwise specified, results shown are means ± standard error of means (SEM). We used SPSS 17.0 to perform the statistical analyses. For an easy interpretation of the figures, the values in the figures represent raw values and not square root transformed values.

Results

Stress response

Salivary cortisol

The model predicting cortisol levels showed main effects for Condition ($F_{1,\ 174.490} = 87.842$, $p < 0.001$), Time ($F_{3,\ 132.927} = 8.225$, $p < 0.001$) and their interaction Condition × Time ($F_{3,\ 132.890} = 36.480$, $p < 0.001$). There were no baseline differences in cortisol levels between conditions ($p = 0.856$). In the stress condition, cortisol levels increased, reaching their peak immediately after the stress task ($p < 0.001$), and then starting to decrease but without recovering baseline levels in the last saliva sample ($p = 0.001$). In the control condition, cortisol levels decreased across time, but the differences were only significant between the −5 min and +15 min samples and the −20 min and +40 min samples (both $p \leq 0.001$). Cortisol levels were higher in the stress condition than in the control condition in both samples provided after the task (both $p < 0.001$).

The main effect of Age was not significant ($p = 0.486$), but the Condition × Age ($F_{1,\ 216.607} = 9.404$, $p = 0.002$) interaction was significant. Both age groups had higher cortisol levels in the stress condition than in the control condition (both $p \leq 0.001$). In addition, in the stress condition both age groups had similar cortisol levels ($p = 0.680$),

but, as a trend, the older group had lower cortisol levels than the younger group in the control condition ($p = 0.057$). The interaction between Time and Age was also significant ($F_{3,\ 111.853} = 13.868$, $p < 0.001$), with older participants showing lower baseline cortisol levels than young participants ($p = 0.018$).

Finally, the factor Sex ($F_{1,\ 67.246} = 9.056$, $p = 0.004$) and the interactions Condition × Sex ($F_{1,\ 216.607} = 13.894$, $p < 0.001$) and Time × Age × Sex ($F_{3,\ 111.853} = 2.856$, $p = 0.040$) were also significant. Men showed higher cortisol levels than women in the experimental condition ($p < 0.001$), but not in the control condition ($p = 0.110$). With respect to Time, we observed that in older people, men only showed higher pre-task levels of cortisol than women in the −5 min sample ($p < 0.015$). However, in young people, men presented significantly higher cortisol levels than women in the +15 min and +40 min samples (both $p < 0.020$), and as a trend, in the −5 min sample ($p = 0.053$). There were no age differences between men and women in any of the four samples (all $p > 0.080$) (see Fig. 1). Model fit did not improve when adding other main effects or interaction effects (see Table 1 in Appendix A).

Salivary alpha-amylase (sAA)

The model predicting sAA levels showed main effects of Condition ($F_{1,\ 453.476} = 64.348$, $p < 0.001$), Time ($F_{4,\ 206.281} = 35.838$, $p < 0.001$), and their interaction, Condition × Time ($F_{4,\ 206.034} = 4.940$, $p = 0.001$). There were no baseline differences between conditions ($p = 0.942$); however, the sAA concentrations were higher in the stress condition than in the control condition in the rest of the samples (all $p \leq 0.001$). In the stress condition, sAA levels were similar to baseline in the 0 min sample ($p = 0.123$), higher in the +5 min and +10 min samples (both $p \leq 0.001$), and decreased until reaching baseline levels in the last sAA sample (+14 min) ($p = 0.423$). In the control condition, a similar sAA profile was found.

The factor Age ($F_{1,\ 61.431} = 3.239$, $p = 0.077$) and the interaction Condition × Age ($F_{1,\ 458.180} = 3.503$, $p = 0.062$) were marginally significant, whereas the interaction Time × Age ($F_{4,\ 173.901} = 3.164$, $p = 0.015$) was significant. Older adults had higher sAA concentrations than younger adults, with this difference being significant in the control condition ($p = 0.035$), but not in the stress condition ($p = 0.172$). Both age groups had higher sAA concentrations in the stress condition than in the control condition (both $p \leq 0.001$). Comparing the two age-groups, the older participants showed higher sAA levels than the younger participants in the +5 min ($p = 0.036$) and +10 min ($p = 0.008$) samples, but not in the rest of the samples (all $p > 0.190$). Finally, the factor Sex and its interactions were not significant (all $p > 0.116$) (see Fig. 2). Model fit did not improve when adding other main effects or interaction effects (see Table 2 in Appendix A).

Memory performance

Learning curve (Trials 1 to 5)

The model predicting the learning curve showed that there was a main effect of Trial ($F_{4,\ 211.477} = 336.876$, $p < 0.001$) and Age ($F_{1,\ 62.296} = 31.178$, $p < 0.001$). All the participants showed a positive learning curve across the first five trials. In every consecutive trial, more words were remembered (all $p < 0.002$). Moreover, older participants had lower performance across the learning curve than young

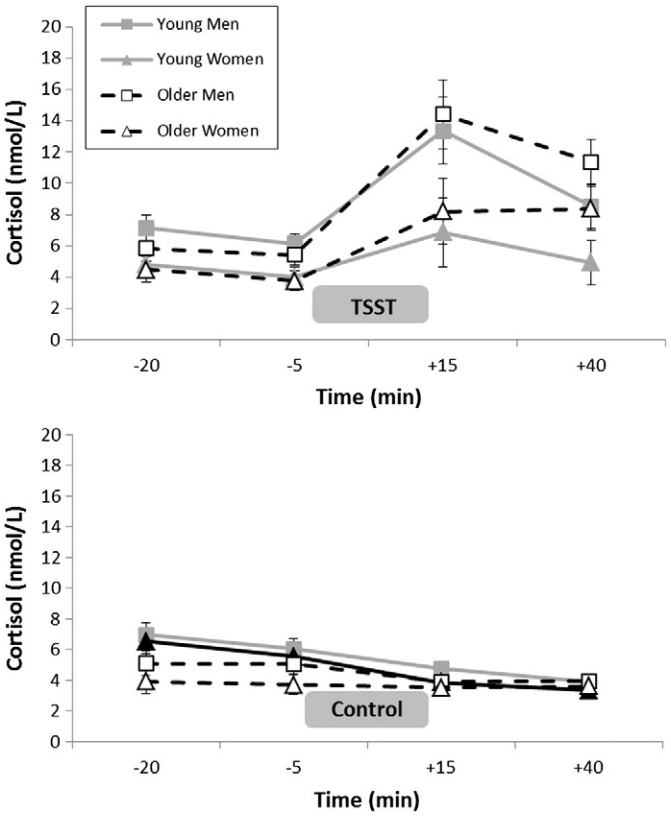

Fig. 1. Means of salivary cortisol concentrations (±SEM) in the TSST (up) and the control (down) conditions in both age groups (young: N = 31, older: N = 31). In the stress condition, all participants increased their cortisol levels immediately after the stress task ($p < 0.001$), with men having higher cortisol levels than women ($p < 0.001$). In the control condition, all participants decreased their cortisol levels across time, according to the normal cortisol circadian rhythm.

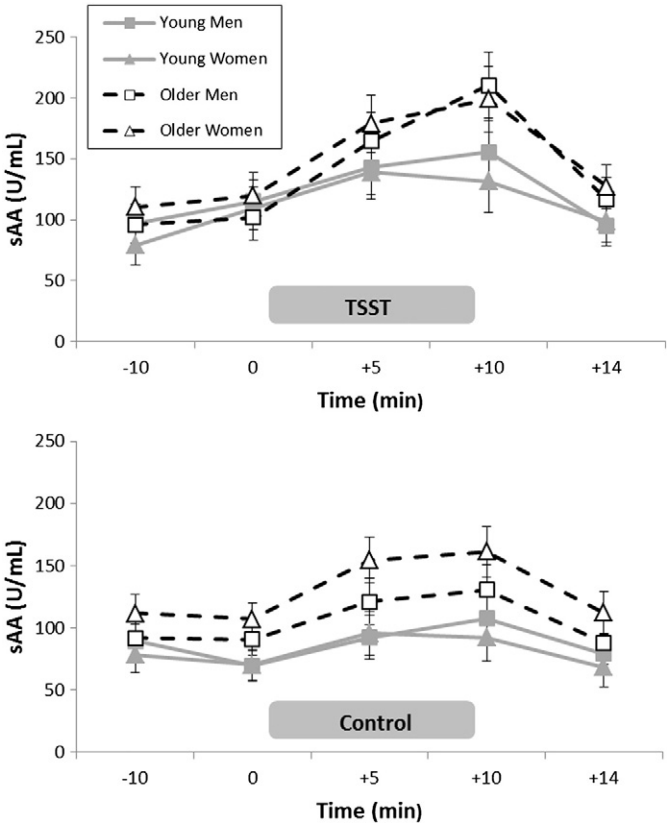

Fig. 2. Means of salivary alpha-amylase (sAA) concentrations (±SEM) in the TSST (up) and control (down) conditions in both age groups (young: N = 34, older: N = 30). Except on baseline sAA concentrations ($p = 0.942$), all participants had higher sAA concentrations in the stress condition than in the control condition (all $p < 0.001$). In addition, the older group had higher sAA concentrations than the young adults, although this difference was only significant in the control condition ($p = 0.035$).

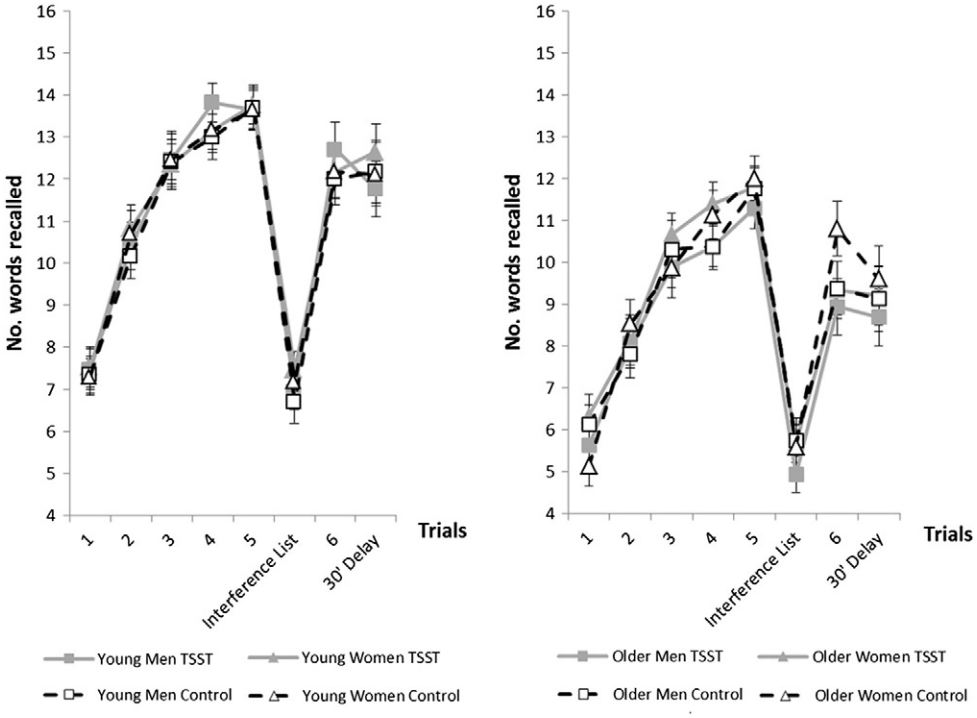

Fig. 3. Number of words recalled in each trial of the RAVLT by (left) young (N = 34) and (right) older (N = 31) groups, divided into men and women in the TSST and control conditions. Among the young participants, no stress effects were found on memory; however, we found an interaction between Condition and Age in the trial 6 outcome. Older people have poorer performance on this trial in the stress condition than in the control condition. Depicted values are means, and error bars represent the SEM.

participants (see Fig. 3). Model fit did not improve when adding other main effects or interaction effects (see Table 3 in Appendix A).

Total learning ($\sum$Trial 1 to Trial 5)

The model predicting total learning showed that there was only a main effect of Age ($F_{1,\ 63} = 31.775$, $p < 0.001$); older people had worse total learning performance than young people. Model fit did not improve when adding other main effects or interaction effects (see Table 4 in Appendix A).

Recall after interference (Trial 6)

The model predicting immediate recall performance showed that there was a main effect of Age ($F_{1,\ 61.995} = 21.103$, $p < 0.001$). Older participants recalled fewer words than young participants. Although the main effect of Condition was not significant ($p = 0.319$), the Condition × Age ($F_{1,63} = 4.935, p = 0.030$, Cohen's $d = 0.32$) interaction was significant. Older participants recalled fewer words after the stress condition than in the control condition ($p = 0.029$); therefore, the stressor only impaired older participants' performance. However, young participants had a similar performance in both conditions ($p = 0.382$), and their performance was better than that of the older participants in both conditions (both $p < 0.002$) (see Fig. 3). Model fit did not improve when adding other main effects or interaction effects (see Table 5 in Appendix A).

Delayed recall (Trial 7)

The model predicting short-term delayed recall performance showed that there was only a main effect of Age ($F_{1,\ 126.314} = 37.284$, $p < 0.001$). Thus, older participants recalled fewer words than young participants (see Fig. 3). Model fit did not improve when adding other main effects or interaction effects (see Table 6 in Appendix A).

Recognition (Trial 8)

The model predicting recognition performance did not show main effects for condition, age or sex, nor were there interactions among these factors (all $p > 0.377$).

The relationship between the stress response and memory performance

The correlations among biomarker indexes and memory performance were analyzed only for Trial 6, due to the significant effect found in the older group. In this group, no significant correlations were found between recall after interference and cortisol reactivity, sAA reactivity or RCA (ratio of cortisol over sAA) (all $p > 0.167$). However, a negative relationship was observed between recall after interference and the RAC (ratio of sAA over cortisol) ($r = -0.507$, $p = 0.006$). Therefore, the older people who had a predominance of sAA response over cortisol response had poorer memory performance.

In the young group, no significant correlations were found between the Trial 6 outcome and cortisol reactivity, sAA reactivity, RCA or RAC (all $p > 0.337$). Significance testing using Fisher's Z tests revealed marginal differences between the older and young groups in the correlation between RAC and Trial 6 outcome ($z = 1.6$, $p = 0.054$).

Discussion

The purpose of this study was to examine the role of age and sex in the relationship between stress and memory performance. To do so, we compared the effect of acute stress on memory in young and older, healthy and non-stressed adults. In a crossover design in which each subject participated in a stress condition and a control condition, we induced stress in the participants by exposing them to an acute psychological stressor (TSST). After both the stress and control tasks, we evaluated their declarative memory performance. Our results confirm that the experimental procedure induced stress, since the TSST provoked an increase in cortisol and sAA responses in the total sample. Although we failed to find stress-induced changes in learning, delayed recall or recognition, the exposure to the TSST impaired immediate recall after interference, but only in older people. In addition, among older people, this effect was negatively related to the ratio of sAA over cortisol. No sex differences were found in the stress effects on memory performance.

The experimental procedure was indeed able to induce stress, since both stress systems (i.e. HPA-axis and SNS) were activated, as reflected

in the cortisol and sAA responses (see Figs. 1 and 2, respectively). However, when we studied the role of age and sex in the stress response, we observed that they each had a different role in the response of each stress biomarker. Thus, we found sex differences in the cortisol response, but not in the sAA response. Men had a higher cortisol response to the TSST than women, regardless of the age. This result coincides with previous studies in young (Childs et al., 2010; Kirschbaum et al., 1999) and older people (Kudielka et al., 1998, 2004). In contrast, we failed to find age differences in the cortisol response, but we found that older adults had higher sAA concentrations than younger adults, and these differences were significant in the control condition, but not in the stress condition. This result confirms the idea that there is increased basal sympathoneural activity among older people (Seals and Dinenno, 2004).

To our knowledge, this is the first study to compare acute stress effects on the memory performance of young and older men and women. The results show that, in general, older people had poorer declarative memory performance than young people, as they recalled fewer words than young participants on all trials of the RAVLT, except the recognition task (see Fig. 3). This result agrees with a previous review on this topic (Park et al., 2003). According to these authors, there is an age-related decline in some types of memory, including declarative and working memory; however, non-declarative and recognition memory performance were maintained, or even improved, across the lifespan.

It is worth noting that the exposition to an acute stressful event tends to enhance learning of new information in adult male animals (for a review on this topic see: Shors, 2006). It is important to note that the direction of stress effects on memory depends on several factors, such as the memory phase assessed (i.e. acquisition, consolidation or retrieval), the type of memory studied, the magnitude of the stress-induced cortisol reactivity, and the sex of the subjects.

We found a very specific, negative effect of the stressor on memory. Specifically, the stressor impaired immediate recall (Trial 6) only in older people. Why did the stressor selectively affect the memory performance of older people? One explanation could be that, although the RAVLT assesses declarative memory, the effect obtained on Trial 6 may fall under the domain of working memory. On this trial the participants had to recall, after an interference list, as many words as possible from the target list, but without its previous presentation as occurred in the first five trials. This new word list interferes with the recall of the previously-learned target list, resulting in retroactive interference (Dewar et al., 2007). According to Hedden and Park (2001), older people show greater retroactive interference effects compared to young adults, so that they seem to be more vulnerable to this interference than young people. Difficulties in deleting irrelevant information from the working memory could hinder their performance. Moreover, both working memory (Galloway et al., 2008) and retroactive interference (Dewar et al., 2007) may be related to prefrontal cortex functioning. In addition to the hippocampus, this brain area seems to be sensitive to glucocorticoid effects during human aging. Several studies suggest that stress exacerbates the aging process (Lupien et al., 2007; Piazza et al., 2010) and, consequently, age-related changes such as memory impairment.

Previous studies by our group and others have suggested that older people may be less sensitive to the effects of acute stress on long-term memory retrieval (Pulopulos et al., 2013) and to the effects of pharmacologically-induced acute cortisol increases on working memory tasks involving the maintenance and manipulation of information (i.e. Digit Span and Letter-Number Sequencing tasks) (Wolf et al., 2001a; Yehuda et al., 2007). An age-related dysregulation of the HPA-axis activity (Mizoguchi et al., 2009) and functional changes in the amygdala and hippocampus (Mather, 2006; Murthy et al., 2010; St. Jaques et al., 2009) have been proposed as possible explanations for the lack of cortisol effects on the performance of these kinds of tasks. Together with our results, these studies indicate that older people may be sensitive to the effect of stress on retroactive interference, a cognitive ability that involves the activation of the prefrontal cortex to control irrelevant information and that has shown a greater age-related decline (Hedden and Park, 2001), but not on other kinds of memory abilities, such as long-term memory retrieval or working memory tasks, which involve maintenance and manipulation of information. However, it should be noted that previous studies investigating the effects of cortisol on working memory in older people have used a pharmacological approach (Wolf et al., 2001a; Yehuda et al., 2007); therefore, the lack of SNS activation in these studies may also account for the absence of cortisol effects observed. Thus, more research is needed to investigate the effects of acute stress on other kind of tasks that specifically measure working memory. Moreover, we found a negative relationship between the ratio of sAA over cortisol and recall after interference only in older people. It should be pointed out that even after considering the Bonferroni correction for multiple analyses, the critical α level would be 0.00625 (0.05/8); therefore, this correlation would remain significant. As we outlined above, the immediate recall of wordlists not only reflects declarative memory processes, but also working memory functions (Lezak et al., 2004; Tops et al., 2004). On the one hand, it has been well established that declarative memory depends on hippocampal functioning (Scoville and Milner, 2000), and working memory depends on prefrontal cortex functioning (Galloway et al., 2008). On the other hand, these two brain structures are affected by the glucocorticoid action and noradrenergic activation in response to stress, respectively (Patel et al., 2000; Schoofs et al., 2008). Therefore, this trial will be affected by the activation of both stress systems related to each type of memory. Taking this into account, we considered it appropriate to examine whether the impairing effects found in the recall after interference were related not only to the HPA-axis or SNS action separately, but also to the relationship between them, expressed as the ratio of one biomarker over the other and vice versa. The hormonal ratio method has been widely implemented in research as a reliable index for a variety of health and behavioral outcomes (Adlercreutz et al., 1986; Ostroff et al., 1982, 1985; Terburg et al., 2009). Recently, the sAA over cortisol ratio has been suggested as a good marker of stress system dysregulation, positively related to subjective indexes of stress and depression (Ali and Pruessner, 2012). We tried to extend this relationship into the cognitive domain, as has been initiated in other stages of the life span (Berry et al., 2012).

Sex differences have previously been reported among older people (Almela et al., 2011a), showing impaired declarative memory, but only related to higher cortisol response to stressors in older women. However, we failed to find sex differences in the relationship between acute stress and memory performance. The small sample size may be the underlying explanation for this lack of significant effects. Further studies are needed to investigate whether the sex affects the relationship between acute stress and retroactive interference, specifically in older people.

Some other limitations have to be considered in the current study. We collected a homogeneous and cognitively and physically healthy sample, using exclusion criteria that have contributed to obtaining a very restricted sample. This fact may limit the ability to detect effects and generalize the results. Further studies are needed to extend this research to a more general population, including older people with age-related diseases and medication use, young women in other phases of the menstrual cycle, and oral contraceptive users. In this study, several outcomes were examined (e.g. different dependent variables from the same memory task), which can lead to an increase in the type I error. However, we found a correlation between the Trial 6 outcome and the RAC in older people and, although as a trend, differences between the correlations in older and young people, in line with the results shown with linear mixed modeling. Taken together, these consistent results do not seem to be due to chance, but they must be considered tentative and confirmed in further studies with other and more extensive samples.

In conclusion, we have studied the role of age in the effects of acute psychosocial stress on declarative memory, considering sex. Our results show a very specific effect associated with the worse consequence of the interference derived from very similar and neutral stimuli in healthy, non-stressed older people. They confirm that age moderates this specific stress-induced effect on memory, providing new knowledge about the importance of studying both physiological systems involved in the stress response together.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.yhbeh.2013.12.017.

References

Adler, N.E., Epel, E.S., Castellazzo, G., Ickovics, J.R., 2000. Relationship of subjective and objective social status with psychological and physiological functioning: preliminary data in healthy, white women. Health Psychol. 19, 586–592.
Adlercreutz, H., Harkonen, M., Kuoppasalmi, K., Naveri, H., Huhtaniemi, I., Tikkanen, H., Remes, K., Dessypris, A., Karvonen, J., 1986. Effect of training on plasma anabolic and catabolic steroid hormones and their response during physical exercise. Int. J. Sports Med. 7 (Suppl. 1), 27–28.
Ali, N., Pruessner, J.C., 2012. The salivary alpha amylase over cortisol ratio as a marker to assess dysregulations of the stress systems. Physiol. Behav. 106, 65–72.
Allwood, M.A., Handwerger, K., Kivlighan, K.T., Granger, D.A., Stroud, L.R., 2011. Direct and moderating links of salivary alpha-amylase and cortisol stress-reactivity to youth behavioral and emotional adjustment. Biol. Psychol. 88, 57–64.
Almela, M., Hidalgo, V., Villada, C., Espín, L., Gómez-Amor, J., Salvador, A., 2011a. The impact of cortisol reactivity on memory: sex differences in middle-aged people. Stress 14, 117–127.
Almela, M., Hidalgo, V., Villada, C., van der Meij, L., Espin, L., Gomez-Amor, J., Salvador, A., 2011b. Salivary alpha-amylase response to acute psychosocial stress: the impact of age. Biol. Psychol. 87, 421–429.
Bauer, A.M., Quas, J.A., Boyce, W.T., 2002. Associations between physiological reactivity and children's behavior: advantages of a multisystem approach. J. Dev. Behav. Pediatr. 23, 102–113.
Baum, B.J., 1993. Principles of saliva secretion. Ann. N. Y. Acad. Sci. 694, 17–23.
Berry, D., Blair, C., Willoughby, M., Granger, D.A., Family Life Project Key Investigators, 2012. Salivary alpha-amylase and cortisol in infancy and toddlerhood: direct and indirect relations with executive functioning and academic ability in childhood. Psychoneuroendocrinology 37, 1700–1711.
Bohnen, N., Houx, P., Nicolson, N., Jolles, J., 1990. Cortisol reactivity and cognitive performance in a continuous mental task paradigm. Biol. Psychol. 31, 107–116.
Burnham, K.P., Anderson, D.R., 2004. Multimodel inference: understanding AIC and BIC in model selection. Sociol. Methods Res. 33, 261–304.
Childs, E., Dlugos, A., De Wit, H., 2010. Cardiovascular, hormonal, and emotional responses to the TSST in relation to sex and menstrual cycle phase. Psychophysiology 47, 550–559.
Dewar, M.T., Cowan, N., Sala, S.D., 2007. Forgetting due to retroactive interference: a fusion of Muller and Pilzecker's (1900) early insights into everyday forgetting and recent research on anterograde amnesia. Cortex 43, 616–634.
Dickerson, S.S., Kemeny, M.E., 2004. Acute stressors and cortisol responses: a theoretical integration and synthesis of laboratory research. Psychol. Bull. 130, 355–391.
Diggle, P.J., 1988. An approach to the analysis of repeated measurements. Biometrics 44, 959–971.
Domes, G., Heinrichs, M., Reichwald, U., Hautzinger, M., 2002. Hypothalamic–pituitary–adrenal axis reactivity to psychological stress and memory in middle-aged women: high responders exhibit enhanced declarative memory performance. Psychoneuroendocrinology 27, 843–853.
El-Sheikh, M., Erath, S.A., Buckhalt, J.A., Granger, D.A., Mize, J., 2008. Cortisol and children's adjustment: the moderating role of sympathetic nervous system activity. J. Abnorm. Child Psychol. 36, 601–611.
Espin, L., Almela, M., Hidalgo, V., Villada, C., Salvador, A., Gomez-Amor, J., 2013. Acute pre-learning stress and declarative memory: impact of sex, cortisol response and menstrual cycle phase. Horm. Behav. 63, 759–765.
Galloway, E.M., Woo, N.H., Lu, B., 2008. Persistent neural activity in the prefrontal cortex: a mechanism by which BDNF regulates working memory? Prog. Brain Res. 169, 251–266.
Gordis, E.B., Granger, D.A., Susman, E.J., Trickett, P.K., 2006. Asymmetry between salivary cortisol and alpha-amylase reactivity to stress: relation to aggressive behavior in adolescents. Psychoneuroendocrinology 31, 976–987.
Gordis, E.B., Granger, D.A., Susman, E.J., Trickett, P.K., 2008. Salivary alpha amylase–cortisol asymmetry in maltreated youth. Horm. Behav. 53, 96–103.
Granger, D.A., Kivlighan, K.T., el-Sheikh, M., Gordis, E.B., Stroud, L.R., 2007. Salivary alpha-amylase in biobehavioral research: recent developments and applications. Ann. N. Y. Acad. Sci. 1098, 122–144.
Hedden, T., Park, D., 2001. Aging and interference in verbal working memory. Psychol. Aging 16, 666–681.
Herman, J.P., Ostrander, M.M., Mueller, N.K., Figueiredo, H., 2005. Limbic system mechanisms of stress regulation: hypothalamo-pituitary-adrenocortical axis. Prog. Neuropsychopharmcol. Biol. Psychiatry 29, 1201–1213.
Het, S., Rohleder, N., Schoofs, D., Kirschbaum, C., Wolf, O., 2009. Neuroendocrine and psychometric evaluation of a placebo version of the "Trier Social Stress Test". Psychoneuroendocrinology 34, 1075–1086.
Hidalgo, V., Villada, C., Almela, M., Espin, L., Gomez-Amor, J., Salvador, A., 2012. Enhancing effects of acute psychosocial stress on priming of non-declarative memory in healthy young adults. Stress 15, 329–338.
Jelicic, M., Geraerts, E., Merckelbach, H., Guerrieri, R., 2004. Acute stress enhances memory for emotional words, but impairs memory for neutral words. Int. J. Neurosci. 114, 1343–1351.
Keller, P.S., El-Sheikh, M., Granger, D.A., Buckhalt, J.A., 2012. Interactions between salivary cortisol and alpha-amylase as predictors of children's cognitive functioning and academic performance. Physiol. Behav. 105, 987–995.
Kirschbaum, C., Pirke, K.M., Hellhammer, D.H., 1993. The 'Trier Social Stress Test'—a tool for investigating psychobiological stress responses in a laboratory setting. Neuropsychobiology 28, 76–81.
Kirschbaum, C., Wolf, O.T., May, M., Wippich, W., Hellhammer, D.H., 1996. Stress- and treatment-induced elevations of cortisol levels associated with impaired declarative memory in healthy adults. Life Sci. 58, 1475–1483.
Kirschbaum, C., Kudielka, B.M., Gaab, J., Schommer, N.C., Hellhammer, D.H., 1999. Impact of gender, menstrual cycle phase, and oral contraceptives on the activity of the hypothalamus–pituitary–adrenal axis. Psychosom. Med. 61, 154–162.
Kudielka, B.M., Hellhammer, J., Hellhammer, D.H., Wolf, O.T., Pirke, K.M., Varadi, E., Pilz, J., Kirschbaum, C., 1998. Sex differences in endocrine and psychological responses to psychosocial stress in healthy elderly subjects and the impact of a 2-week dehydroepiandrosterone treatment. J. Clin. Endocrinol. Metab. 83, 1756–1761.
Kudielka, B.M., Schommer, N.C., Hellhammer, D.H., Kirschbaum, C., 2004. Acute HPA axis responses, heart rate, and mood changes to psychosocial stress (TSST) in humans at different times of day. Psychoneuroendocrinology 29, 983–992.
Kudielka, B.M., Hellhammer, D.H., Wüst, S., 2009. Why do we respond so differently? Reviewing determinants of human salivary cortisol responses to challenge. Psychoneuroendocrinology 34, 2–18.
Lezak, M.D., Howleson, D.B., Loring, D.W., 2004. Neuropsychological Assessment. Oxford University Press, New York.
Lupien, S.J., Lepage, M., 2001. Stress, memory, and the hippocampus: can't live with it, can't live without it. Behav. Brain Res. 127, 137–158.
Lupien, S.J., Schramek, T.E., 2006. The differential effects of stress on memory consolidation and retrieval: a potential involvement of reconsolidation? Theoretical comment on Beckner et al. (2006). Behav. Neurosci. 120, 735–738.
Lupien, S.J., Maheu, F., Tu, M., Fiocco, A., Schramek, T.E., 2007. The effects of stress and stress hormones on human cognition: implications for the field of brain and cognition. Brain Cogn. 65, 209–237.
Lupien, S.J., McEwen, B.S., Gunnar, M.R., Heim, C., 2009. Effects of stress throughout the lifespan on the brain, behaviour and cognition. Nat. Rev. Neurosci. 10, 434–445.
Mather, M., 2006. Why memories may become more positive as people age. In: Uttl, B., Ohta, A.L. (Eds.), Memory and Emotion: Interdisciplinary Perspectives. Blackwell, Malden, MA, pp. 135–157.
Miranda, J.P., Valencia, R.R., 1997. English and Spanish versions of a memory test: word-length effects versus spoken-duration effects. Hisp. J. Behav. Sci. 19, 171–181.
Mizoguchi, K., Ikeda, R., Shoji, H., Tanaka, Y., Maruyama, W., Tabira, T., 2009. Aging attenuates glucocorticoid negative feedback in rat brain. Neuroscience 159, 259–270.
Murthy, V.P., Sambataro, F., Das, S., Tan, H., Callicott, J.H., Golberg, T.E., et al., 2010. Age-related alterations in simple declarative memory and the effect of negative stimulus valence. J. Cogn. Neurosci. 21, 1920–1933.
Nater, U.M., Rohleder, N., 2009. Salivary alpha-amylase as a non-invasive biomarker for the sympathetic nervous system: current state of research. Psychoneuroendocrinology 34, 486–496.
Nater, U.M., Moor, C., Okere, U., Stallkamp, R., Martin, M., Ehlert, U., Kliegel, M., 2007. Performance on a declarative memory task is better in high than low cortisol responders to psychosocial stress. Psychoneuroendocrinology 32, 758–763.
Ostroff, R., Giller, E., Bonese, K., Ebersole, E., Harkness, L., Mason, J., 1982. Neuroendocrine risk factors of suicidal behavior. Am. J. Psychiatry 139, 1323–1325.
Ostroff, R.B., Giller, E., Harkness, L., Mason, J., 1985. The norepinephrine-to-epinephrine ratio in patients with a history of suicide attempts. Am. J. Psychiatry 142, 224–227.

Park, H.L., O'Connell, J.E., Thomson, R.G., 2003. A systematic review of cognitive decline in the general elderly population. Int. J. Geriatr. Psychiatry 18, 1121–1134.
Patel, P.D., Lopez, J.F., Lyons, D.M., Burke, S., Wallace, M., Schatzberg, A.F., 2000. Glucocorticoid and mineralcorticoid receptor mRNA expression in squirrel monkey brain. J. Psychiatr. Res. 34, 383–392.
Payne, J., Jackson, E., Ryan, L., Hoscheidt, S., Jacobs, J., Nadel, L., 2006. The impact of stress on neutral and emotional aspects of episodic memory. Memory 14, 1–16.
Payne, J.D., Jackson, E.D., Hoscheidt, S., Ryan, L., Jacobs, W.J., Nadel, L., 2007. Stress administered prior to encoding impairs neutral but enhances emotional long-term episodic memories. Learn. Mem. 14, 861–868.
Piazza, J.R., Almeida, D.M., Dmitrieva, N.O., Klein, L.C., 2010. Frontiers in the use of biomarkers of health in research on stress and aging. J. Gerontol. B Psychol. Sci. Soc. Sci. 65, 513–525.
Pulopulos, M.M., Almela, M., Hidalgo, V., Villada, C., Puig-Perez, S., Salvador, A., 2013. Acute stress does not impair long-term memory retrieval in older people. Neurobiol. Learn. Mem. 104, 16–24.
Rohleder, N., Nater, U.M., 2009. Determinants of salivary alpha-amylase in humans and methodological considerations. Psychoneuroendocrinology 34, 469–485.
Rohleder, N., Wolf, J.M., Maldonado, E.F., Kirschbaum, C., 2006. The psychosocial stress-induced increase in salivary alpha-amylase is independent of saliva flow rate. Psychophysiology 43, 645–652.
Roozendaal, B., 2000. 1999 Curt P. Richter award. Glucocorticoids and the regulation of memory consolidation. Psychoneuroendocrinology 25, 213–238.
Roozendaal, B., McEwen, B., Chattarji, S., 2009. Stress, memory and the amygdala. Nat. Rev. Neurosci. 10, 423–433.
Scannapieco, F.A., Torres, G., Levine, M.J., 1993. Salivary alpha-amylase: role in dental plaque and caries formation. Crit. Rev. Oral Biol. Med. 4, 301–307.
Schoofs, D., Preuss, D., Wolf, O.T., 2008. Psychosocial stress induces working memory impairments in an n-back paradigm. Psychoneuroendocrinology 33, 643–653.
Schwabe, L., Bohringer, A., Chatterjee, M., Schachinger, H., 2008. Effects of pre-learning stress on memory for neutral, positive and negative words: different roles of cortisol and autonomic arousal. Neurobiol. Learn. Mem. 90, 44–53.
Scoville, W.B., Milner, B., 2000. Loss of recent memory after bilateral hippocampal lesions. J. Neuropsychiatry Clin. Neurosci. 12, 103–113.
Seals, D.R., Dinenno, F.A., 2004. Collateral damage: cardiovascular consequences of chronic sympathetic activation with human aging. Am. J. Physiol. Heart Circ. Physiol. 287, H1895–H1905.
Shors, T.J., 2006. Stressful experience and learning across the lifespan. Annu. Rev. Psychol. 57, 55–85.
Smeets, T., Jelicic, M., Merchelbach, H., 2006. The effect of acute stress on memory depends on word valence. Int. J. Psychophysiol. 62, 30–37.
St. Jaques, P.L., Dolcos, F., Cabeza, R., 2009. Effects of aging on functional connectivity of the amygdala for subsequent memory of negative pictures: a network analysis of functional magnetic resonance imaging data. Psychol. Sci. 20, 74–84.
Strahler, J., Mueller, A., Rosenloecher, F., Kirschbaum, C., Rohleder, N., 2010. Salivary alpha-amylase stress reactivity across different age groups. Psychophysiology 47, 587–595.
Terburg, D., Morgan, B., van Honk, J., 2009. The testosterone–cortisol ratio: a hormonal marker for proneness to social aggression. Int. J. Law Psychiatry 32, 216–223.
Tops, M., van der Pompe, G., Wijers, A.A., Den Boer, J.A., Meijman, T.F., Korf, J., 2004. Free recall of pleasant words from recency positions is especially sensitive to acute administration of cortisol. Psychoneuroendocrinology 29, 327–338.
Vigil, J.M., Geary, D.C., Granger, D.A., Flinn, M.V., 2010. Sex differences in salivary cortisol, alpha-amylase, and psychological functioning following Hurricane Katrina. Child Dev. 81, 1228–1240.
Wolf, O.T., Convit, A., McHugh, P.F., Kandil, E., Thorn, E.L., De Santi, S., McEwen, B.S., de Leon, M.J., 2001a. Cortisol differentially affects memory in young and elderly men. Behav. Neurosci. 115, 1002–1011.
Wolf, O.T., Schommer, N.C., Hellhammer, D.H., McEwen, B.S., Kirschbaum, C., 2001b. The relationship between stress induced cortisol levels and memory differs between men and women. Psychoneuroendocrinology 26, 711–720.
Yehuda, R., Harvey, P.D., Buchsbaum, M., Tischler, L., Schmeidler, J., 2007. Enhanced effects of cortisol administration on episodic and working memory in aging veterans with PTSD. Neuropsychopharmacology 32, 2581–2591.