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# Rapid effects of acute stress on cognitive emotion regulation

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## ABSTRACT

Acute stress has been shown to either enhance or impair emotion regulation (ER) performances. Besides sex, strategy use and stimulus intensity, another moderating factor appears to be timing of the ER task relative to stress exposure. Whereas somewhat delayed increases in the stress hormone cortisol have been shown to improve ER performances, rapid sympathetic nervous system (SNS) actions might oppose such effects via cognitive regulatory impairments. Here, we thus investigated rapid effects of acute stress on two ER strategies: reappraisal and distraction. N = 80 healthy participants (40 men & 40 women) were exposed to the Socially Evaluated Cold-Pressor Test or a control condition immediately prior to an ER paradigm which required them to deliberately downregulate emotional responses towards high intensity negative pictures. Subjective ratings and pupil dilation served as ER outcomes measures. Increases in salivary cortisol and cardiovascular activity (index of SNS activation) verified successful induction of acute stress. Unexpectedly, stress reduced subjective emotional arousal when distracting from negative pictures in men indicating regulatory improvements. However, this beneficial effect was particularly pronounced in the second half of the ER paradigm and fully mediated by already rising cortisol levels. In contrast, cardiovascular responses to stress were linked to decreased subjective regulatory performances of reappraisal and distraction in women. However, no detrimental effects of stress on ER occurred at the group level. Yet, our findings provide initial evidence for rapid, opposing effects of the two stress systems on the cognitive control of negative emotions that are critically moderated by sex.

#### 1. Introduction

Emotions are fundamentally adaptive, but can also be harmful when occurring too intense and long-lasting or provoking maladaptive action tendencies (Gross and Jazaieri, 2014). Therefore, the ability to flexibly regulate upcoming emotions is a crucial need in everyday life. In stressful situations, emotion regulation (ER) competencies are probably needed the most helping the organism to adapt to and recover from emotionally challenging events. ER deficits have been repeatedly linked to chronic stress states (Ragen et al., 2016) increasing the risk for the development and maintenance of mental disorders (Berking and Wupperman, 2012). Given its clinical relevance, it is essential to shed light on the neuroendocrinological mechanisms of acute stress effects on ER processes.

Cognitive ER comprises all cognitive attempts to change the type, intensity or duration of a current emotional state (Gross, 2015). *Reappraisal* and *distraction* are amongst the most powerful strategies to downregulate negative emotions (Webb et al., 2012) differing in long-term adaptivity, recruitment of cognitive control resources and

effectiveness when dealing with high intensity emotions (for a review, see Sheppes, 2020). While reappraisal refers to a reinterpretation of a given stimulus to change the valence of the emotional meaning, distraction aims at redirecting the attention away from the stimulus (Gross, 2015). Cognitive ER relies on a neural network composed of prefrontal (PFC), inferior parietal and cingulate cortex regions inhibiting activity in the amygdala (e.g., Etkin et al., 2015). Importantly, these brain regions are primary targets of physiological stress mediators such as cortisol (McEwen et al., 2016) implying an interrelated relationship. Acute stress quickly activates the sympathetic nervous system (SNS) leading to the release of catecholamines (e.g., adrenaline and noradrenaline) and the somewhat slower-acting hypothalamus-pituitary adrenocortical (HPA) axis. Stimulation of the HPA axis prompts the secretion of glucocorticoids (GCs; cortisol in humans) reaching its peak ~25 min after stress onset (Joëls and Baram, 2009). Both, catecholamines and glucocorticoids modify brain activity via  $\alpha$ - and  $\beta$ -adrenergic receptors as well as mineralocorticoid (MR) and glucocorticoid receptors (GR; Ulrich-Lai and Herman, 2009), respectively, in a regionally specific and timing-dependent manner (Hermans et al., 2014). In doing

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so, stress hormones work in concert to compose adaptive changes in cognitive and affective functioning to optimize stress coping (de Kloet et al., 2005).

Existing evidence of stress effects on cognitive ER is still relatively sparse and somewhat inconsistent revealing either beneficial, impairing or null findings. One potential moderating factor appears to be the timing of the ER task relative to stress exposure suggesting the predominance of the respective stress system (fast-acting SNS vs. slowacting HPA axis) to play a crucial role for the direction of stress effects to occur. Research from our lab revealed stress to improve ER performances when specifically applying reappraisal 25 min after exposure to psychosocial stress (Kinner et al., 2014). These beneficial stress effects on reappraisal have been linked to increases in cortisol probably boosting the cognitive engagement during regulatory attempts (Langer et al., 2020). Supporting these findings, oral administration of hydrocortisone increased PFC activity during distraction and decreased amygdala activity during reappraisal (Jentsch et al., 2019) particularly enhancing regulatory performances when dealing with high intensity emotions (Langer et al., 2022, 2021a). In contrast, there is evidence for acute stress to reduce the effectiveness of reappraisal to downregulate negative emotions (Raio et al., 2013; Zhan et al., 2017). Interestingly, in these studies ER performances were tested somewhat earlier after stress (~15 min) as compared to studies reporting regulatory improvements. Moreover, reappraisal success was negatively linked to salivary alpha-amylase levels (sAA; indirect marker of noradrenergic activation; Nater and Rohleder, 2009) hinting at the SNS to impair ER performances. In favor of this idea, catecholaminergic actions have repeatedly been associated with dampened prefrontal control functioning (Arnsten, 2009) and increased emotion-related amygdala activity (for a review, see Hermans et al., 2014). Taken together, SNS actions in response to stress may interfere with cognitive attempts to downregulate negative emotions which might be counteracted by somewhat delayed starting HPA axis effects.

A growing body of work suggests sex differences in stress reactivity (Kudielka and Kirschbaum, 2005), ER effectivity and flexibility (Goubet and Chrysikou, 2019; McRae et al., 2008) as well as stress effects on cognitive and emotional functioning (Jentsch et al., 2022; Shields et al., 2016; ter Horst et al., 2012). Complementary, previous studies from our lab showed stress effects on ER effectivity to depend on sex (Kinner et al., 2014; Langer et al., 2020). More specifically, stress improved reappraisal capacities in men but not in women possibly mediated via larger cortisol increases in men (Langer et al., 2020). In addition, men and women appear to differ in the excitability of the locus coeruleus (major source of catecholamines in the brain; Roosevelt et al., 2006) hinting at possible sex-specific SNS-driven stress effects on cognitive ER.

Besides timing of ER after stress, previous studies differ in the used stress induction protocol (e.g., psychosocial vs. physical), emotional material (e.g., pictures vs. stories), ER outcome measures (e.g., subjective vs. physiological vs. neural) and sample characteristics. As stated before, sex (Kinner et al., 2014; Langer et al., 2020) but also intensity of the emotional material (Langer et al., 2022, 2021a) were identified as two critical moderators. Thus, it is still not clear whether acute stress indeed compromise ER capacities in early time windows after stress. Furthermore, regulatory success of distraction immediately after stress never had been studied so far. To fill in these gaps, we examined rapid stress effects on the effectiveness of two ER strategies (reappraisal & distraction) in men and women using a similar methodological approach as previous studies from our lab revealing ER improvements at least 25 min after stress onset (Langer et al., 2022, 2021b, 2020). Eighty men (n = 40) and women (n = 40) were either exposed to stress or a control condition immediately prior to an ER paradigm. Ratings of emotional arousal, valence and regulatory success at the end of each trial served as subjective ER outcomes measures. Similar to previous studies of our lab (Langer et al., 2021b, 2020), pupil sizes were recorded as a physiological proxy of ER. Besides evidence for the pupil to dilate as a function of emotional arousal (Bradley et al., 2008), recent research showed that pupil diameters also enlarge with increasing cognitive effort required for ER (Kinner et al., 2017; Langer et al., 2021b, 2020). Collectively, pupil dilation provides information on both, changes in emotional arousal and the cognitive regulatory effort.

With respect to previous findings (e.g., Raio et al., 2013), we expected stress to reduce ER performances primarily when applying reappraisal which should be reflected by enhanced arousal, reduced valence and success ratings. Given that the pupil dilates in dependence of prefrontal regulatory control (Urry, 2006) and evidence for rapid detrimental effects of stress on PFC activity (Arnsten, 2009), we predicted reduced pupil sizes during ER after stress. In addition, we hypothesized heightened cardiovascular reactivity (SNS biomarker) but not cortisol responses (HPA axis biomarker) to be related to reduced ER performances. Given evidence for stronger stress effects on cognitive ER (e.g., Langer et al., 2020) in men relative to women, we expected the effects to be particularly pronounced in male participants.

## 2. Materials and methods

## 2.1. Participants

To determine the required sample size we conducted an a-priori power analysis using G\*Power 3.1 (Faul et al., 2009). With respect to previous studies from our lab (Langer et al., 2020, 2021a), we assumed a small-to-medium-sized sex-dependent effect (d=0.3) of stress on ER outcomes. Analysis revealed a sample size of 80 participants to detect a significant interaction between stress, sex and ER condition with  $\alpha$  = 0.05, an assumed correlation of r = 0.4 for repeated measurements and a power of  $1-\beta \ge 0.95$ . Thus, 80 healthy participants (40 men and 40 women) aged between 18 and 35 years (M=24.40, SD=4.45) and a mean Body Mass Index (BMI) between 18 and 29 (M=23.33 kg/m<sup>2</sup>  $SD=2.76 \text{ kg/m}^2$ ) were recruited via online advertisements, mailing lists and notice boards throughout the Ruhr University Bochum. Volunteers were excluded from participation if they reported any chronic or acute illnesses, history or current psychological treatment, hormonal contraception, irregular menstrual cycle, drug use including smoking, previous experiences with the current stress protocol or the ER paradigm and corrected-to-normal vision more than  $\pm$  1.5 diopters due to pupillary recordings. All naturally cycling women were exclusively tested in the luteal phase defined as three to nine days prior to the next menses (Schoofs and Wolf, 2009). An equal number of male and female participants was randomly assigned to the stress and the control condition which did not differ in BMI, age, habitual use of reappraisal and distraction as assessed via the emotion regulation inventory (ERI) or flexibility in the use of different ER strategies in daily life (Flex-ER; all ps > .05). The present study was not preregistered. The experimental procedure was in accordance with the Declaration of Helsinki and approved by the ethics committee of the psychological faculty at the Ruhr University Bochum (n. 604).

#### 2.2. Experimental procedure

All participants were instructed to refrain from sports, drugs and alcohol 24 h prior to experimental testing as well as food and any drinks except for water two hours before. All testing took place between 10.30 a.m. and 6.00 p.m. In order to avoid confounding effects of the cortisol awakening response (Pruessner et al., 1997), participants were asked to wake up at least two hours before the start of the experimental testing. To check this requirement, awakening time was assessed prior to testing. In addition, groups were matched for time ensuring no systematic differences in testing time between stressed men, stressed women, control men and control women ( $\chi^2$ -test: p > .05). The procedure started with study information, written informed consent and some questionnaires (demographic data, ERI, Flex-ER, Brief Symptom Inventory (BSI)) after which participants were prepared for pupillary recordings and instructed as well as familiarized with the ER paradigm. After baseline

cardiovascular measurement, participants underwent the stress or control protocol followed by a subjective stress questionnaire. The experimenter then shortly reminded all participants of the ER strategy instructions to ensure correct task comprehension. The ER paradigm started directly after the stress / control manipulation as soon as calibration of pupillary recordings has been finished. At several time points across the experiment participants provided saliva samples and rated their current affective state (baseline,t.<sub>2</sub>,t<sub>+2</sub>,t<sub>+30</sub>; see Fig. 1). At the end of each testing procedure, participants were debriefed and reimbursed with 15  $\in$ .

#### 2.3. Stress and control manipulation

Half of male and female participants were exposed to the Socially Evaluated Cold-Pressor Test (SECPT; Schwabe et al., 2008), whereas the other half underwent a warm water control condition. In the SECPT, participants were asked to immerse their non-dominant hand including the wrist into ice cold water (0–2  $^{\circ}$ C). At the same time participants were videotaped as well as observed and corrected by a reserved experimenter of the opposite sex who did not provide any facial or social feedback. Participants were instructed to look into the camera and sit upright while keeping their hand in cold water as long as possible. After three minutes participants were asked to remove their hand from the water. In the warm water control condition, participants were required to put their hand in warm water (37  $^{\circ}$ C) for three minutes without being observed or videotaped.

To validate cardiovascular reactivity (SNS marker) in response to the SECPT, systolic and diastolic blood pressure (BP) as well as heart rate (HR) was recorded via an Omron M700 Intelli IT device (Omron Healthcare Co., Kyoto, Japan) immediately before stress onset (baseline), during the stress / control procedure (peak) and eight minutes after stress onset (post) for three times within three minutes, respectively. For each time point, BP and HR data were averaged across three measurements. Due to technical failure, cardiovascular data of five participants could not be recorded (2 stress, 3 control). Activation of the HPA axis was checked by collection of saliva samples using Salivette® sampling devices (Sarstedt, Nümbrecht, Germany) at multiple time points across the experiment (baseline,t.2,t+2,t+30; see Fig. 1). Salivettes $\mathbb{R}$  were stored at -20 °C and subsequently analyzed with a timeresolved fluorescence immunoassay (IBL; Hamburg, Germany) to determine the amount of free, unbound salivary cortisol. Due to an insufficient amount of saliva, cortisol levels of one female participant could not be determined. All intra- and inter-assay coefficients of variance were below 5.41%. The Differential Affect Scale (DAS; Merten and Krause, 1993) was used to assess subjective stress responses via mean summary scores of negative (sadness, anger, disgust, contempt, anxiety, shame, guilt) and positive affect factor values (joy, surprise, interest).

Immediately after having completed the stress/control procedure participants additionally evaluated the experienced situation in terms of difficulty, stressfulness, painfulness and unpleasantness on a visual analog scale ranging from 0 ("not at all") to 100 ("very much" adopted from Schwabe et al., 2008).

### 2.4. Emotion regulation paradigm

A slightly adapted version of the emotion regulation paradigm implemented in previous experiments of our lab was used (Kinner et al., 2017; Langer et al., 2021a, 2020) to increase comparability of the results. It is a well-established paradigm in laboratory research on emotion regulation (Sheppes, 2020) and has been shown to reliably induce emotional activation which can be influenced by deliberate regulatory attempts following strategy instructions (e.g., Kanske et al., 2011; Kinner et al., 2014; Schönfelder et al., 2013). In this task, participants were asked to simply view neutral and negative pictures (control conditions) or to deliberately downregulate emotional responses towards negative pictures via reappraisal and distraction (regulation conditions). In the reappraisal condition, participants were asked to reframe the presented situation by imagining it to either happen in a positive context or with a positive ending. In the distraction condition, participants were instructed to think about a completely unrelated, neutral situation while watching the picture to provoke a self-monitored attentional shift. In the view condition, participants were required to watch and respond naturally to the presented picture. To ensure correct strategy application, the experimenter went through all instructions together with the participants and then practiced each strategy with sample pictures. Furthermore, six computer-based practice trials (two trials for each regulation condition and one for each view condition) served to familiarize participants with the procedure and timing of the paradigm.

At the beginning of each trial, a 750 ms instructional cue (view, reappraisal, distraction) was presented. Next, a white fixation cross was displayed on a gray luminance-matched background for 2500 ms prior to picture presentation which introduced either the regulation phase or the view control condition lasting for 5000 ms. Afterwards, participants were asked to rate their emotional responses on a 9-point visual analog scale with respect to arousal (ranging between 1 =emotionally quiet to 9 =emotionally active) and valence (ranging between 1 =unpleasant to 9 =pleasant). In addition, participants rated how successful they were in responding naturally or applying the two regulatory strategies on a 5-point scale (ranging from 1 =not successful at all to 5 =very good). Each rating scale was presented for 5000 ms followed by an inter-trial interval of 2000 ms depicting a black screen. Presentation of the stimuli and behavioral recordings were controlled by MATLAB R2020a (MathWorks Inc. Natick, MA).



The ER paradigm consisted of four ER conditions (view neutral, view

**Fig. 1.** Study procedure. Participants provided four saliva samples together with affective state ratings (Differential Affect Scale; DAS) at different time points across the experiment marked with dark blue boxes (baseline,  $t_{.2}$ ,  $t_{+20}$ ,  $t_{+30}$ ). After exposure to the Socially Evaluated Cold-Pressor Test (SECPT) or the warm water control condition ( $t_{+2}$ ) participants received an additional subjective stress questionnaire. Cardiovascular recordings of blood pressure (BP) and heart rate (HR) were scheduled directly prior to, during and after the SECPT / control condition. The emotion regulation paradigm started as soon as pupil calibration has been finished.

negative, reappraisal, distraction) presented in sets of five trials in a pseudorandomized order, once in the first and once in the second half of the paradigm (overall 40 trials). All pictures were taken from the Nencki Affective Picture System (NAPS; Marchewka et al., 2014) and were presented only once. Three sets of 10 high intensity negative pictures (overall 30 negative pictures; valence: M=2.27, SD=0.52; arousal: M=7.36, SD=0.29) were matched for content and complexity and randomly assigned to the reappraisal, distraction and view negative condition. In addition, a set of 10 neutral pictures (valence: M=4.92, SD=0.35; arousal: M=4.53, SD=0.18) was used in the *view neutral* condition. Negative pictures were normatively rated as significantly more arousing (t(38) = 14.92, p < .001) and less pleasant (t(38) = -28.47, p < .001) than neutral pictures. All pictures were displayed in grayscale and matched for mean luminosity using the MATLAB R2016a SHINE toolbox (Willenbockel et al., 2010).

## 2.5. Pupillometry

Pupil diameter was recorded using the Eyelink® Portable Duo eye tracker (SR Research Ltd., Mississauga, Ontario, Canada) connected to an EyeLink recording device (ThinkPad T470 W10DG, Lenovo Notebook). The eye tracker was permanently located 50 cm in front of participant's head below the PC screen. It is equipped with a high-speed USB camera on the left side and an infrared illuminator on the right side for dark pupil detection assessing retinal and corneal reflections to obtain participants' pupil sizes of both eyes. A double ten-point calibration procedure ensured correct tracking of the pupil. During the ER paradigm, pupil data were continuously recorded at a binocular sampling rate of 250 Hz in arbitrary units while participant's head was permanently stabilized via a chin rest. To control for variation in luminosity, all testing took place in a moderately lit room without any daylight incidence.

Preprocessing of pupillary data was conducted according to previous studies from our lab (Kinner et al., 2017; Langer et al., 2022, 2020). Pupil diameter was averaged across both eyes and smoothed with a finite impulse response filter at 6 Hz. We removed dilation speed outliers with a cutoff threshold of 15 median absolute deviations at most (MAD; Kret and Sjak-Shie, 2018). A MATLAB-based algorithm was used to discard trials with major gaps resulting from eye blinks (>10 samples) and to correct trials with smaller gaps using linear interpolation. Pupil sizes recorded during the 300 ms prior to each picture onset for each participant was subtracted from mean pupil diameters during each picture presentation to control for individual differences. The area under the curve with respect to ground (AUCg) from 2 s to 5 s after picture onset (cf. Langer et al., 2022, 2020) served as a measure of pupillary responses to picture presentation. Pupillary data were averaged across all trials of each ER condition and across five trials per condition in each half of the paradigm for exploratory purposes.

#### 2.6. Statistical analysis

To analyze rapid stress effects on ER outcomes, we used a  $2 \times 2 \times 4$  mixed study design with the between-subject factors *Stress* (SECPT vs. control) and *Sex* (men vs. women) and the within-subject factor ER *Condition* (view neutral vs. view negative vs. reappraisal vs. distraction). All statistical analyses were conducted with IBM SPSS Statistics 20 (Armonk, USA) for Windows with a significance level of  $\alpha = .05$ . Kolmogorov-Smirnov tests served to check for normal distribution of all outcome variables. Since salivary cortisol and affect ratings were skewed (both  $ps \leq .035$ ), statistical analyses were conducted with log-transformed data. In addition, we checked all dependent variables for homogeneity of variance via Levene-tests and reported Greenhouse-Geisser corrected *p*-values and degrees of freedom if sphericity was violated. Partial eta square ( $\eta^2$ ) are reported as estimations of effect sizes.

All analyses of variance (ANOVAs) included the between-subject

factors Stress (SECPT vs. control) and Sex (males vs. females). Significant interactions were solved using appropriate (Bonferroni-corrected) post-hoc tests. To verify successful stress induction, salivary cortisol, affect ratings, systolic (BPsys), diastolic blood pressure (BPdia) and HR were analyzed using mixed-design ANOVAs with the repeated measures factor Time (tbaseline vs. t-2 vs. t+ 2 vs. t+ 30 for cortisol & affect ratings; baseline vs. peak vs. post for BP & HR). Differences in the subjective stress experience at  $t_{+2}$  between the SECPT and the control group were analyzed via multivariate ANOVA with difficulty, stressfulness, painfulness, unpleasantness as dependent variables. To verify successful induction of negative emotions and emotional downregulation via reappraisal and distraction as well as to investigate stress effects on ER outcomes, we conducted mixed-design ANOVAs with the repeated measures factor Condition (view neutral vs. view negative vs. reappraisal vs. distraction) for all ER outcome measures (arousal, valence, success ratings and pupil dilations).

Examining the link between physiological stress mediators and ER outcomes, we calculated delta scores of stress biomarkers ( $\Delta$  BP<sub>svs</sub>,  $\Delta$  $BP_{dia}$ ,  $\Delta$  HR,  $\Delta$  cortisol) subtracting the baseline sample from the respective peak sample (BP & HR:  $t_{+2}$ ; cortisol:  $t_{+30}$ ) and correlated them with mean subjective ratings and pupillary data for reappraisal and distraction specifically in the stress group using Pearson productmoment correlations. To test whether stress effects on ER were predominantly mediated by activation of one of the two major stress systems (SNS vs. HPA) and whether this mediation is further modulated by sex, we conducted moderated mediation analyses using the PROCESS 3.2 macro model 14 for SPSS (Hayes, 2013) with stress as the predictor X (control=0, stress=1), ER outcomes as the outcome variable Y, increases in stress biomarkers ( $\Delta$  BP<sub>sys</sub>,  $\Delta$  BP<sub>dia</sub>,  $\Delta$  HR,  $\Delta$  cortisol) as possible mediators M and Sex as the moderator W (male=0, female=1). Bootstrap tests served to test the significance of the different paths. Direct and indirect effects were examined via calculation of 5000 biascorrected and accelerated (BCa) bootstrap 95% confidence intervals (CI). P-values for each pathway and the BCa CI for significance of the indirect effects are reported.

#### 3. Results

## 3.1. Stress induction

## 3.1.1. The physiological stress response

Exposure to the SECPT caused significant increases in BP<sub>sys</sub> (Stress x Time: F(2,140)=30.37, p < .001;  $\eta^2=0.303$ ; Fig. 2a), BP<sub>dia</sub> (Stress x Time: F(1.76,123.36)=33.73, p < .001;  $\eta^2=0.325$ ; Fig. 2b), HR (Stress x Time: F(1.35,94.30)=3.59, p = .049;  $\eta^2=0.049$ ; Fig. 2c) and salivary cortisol levels (Stress x Time: F(1.52,114.32)=36.50, p < .001;  $\eta^2=0.327$ ; Fig. 2d) verifying successful induction of physiological stress. *Post-hoc* pairwise comparisons confirmed that groups did not differ in BP<sub>sys</sub>, BP<sub>dia</sub>, HR and cortisol at baseline (all ps > .05). During the SECPT, however, stressed participants showed significant higher values of BP<sub>sys</sub> (t(73)=-3.29, p = .002) and BP<sub>dia</sub> (t(73)=-4.30, p < .001) than controls. As expected, 30 min after SECPT onset, stressed participants exhibited significant larger salivary cortisol levels than controls (t(77)=-3.21, p = .002). There were no significant differences in physiological stress responses between men and women (all ps > .05).

#### 3.1.2. The subjective stress response

No significant Stress x Time interaction for affect ratings (DAS) occurred (p > .05). However, in response to the SECPT participants reported significant larger increases in negative affect compared to the warm water control condition ( $\Delta$  DAS; t(76) = -2.11, p = .038). Furthermore, participants rated the SECPT as significantly more difficult, stressful, painful and unpleasant than the control procedure (main effects of Stress: all ps < .001) verifying successful induction of subjective stress. A significant Stress x Sex interaction (F(1,76) = 4.43, p = .039;  $\eta^2 = 0.055$ ) indicated that women rated the SECPT as



Fig. 2. Biomarkers of the stress response. Mean (± SEM) systolic (a) and diastolic (b) blood pressure (in mmHg) as well as heart rate (in beats/ minute, c) and mean ( $\pm$  SEM) salivary cortisol levels (in nmol/l, d) for participants in the stress (Socially Evaluated Cold-Pressor Test, SECPT) and the control group (warm water condition). Exposure to the SECPT caused significant increases in systolic, diastolic blood pressure and heart rate rapidly returning to baseline after stress offset. Moreover, the SECPT caused significant increases in salivary cortisol 30 min after stress onset. The time point of stress manipulation (SECPT / control) and the emotion regulation (ER) paradigm is highlighted by shaded areas. Significant differences between the stress and the control group after Bonferroni-corrected post-hoc t-tests are marked as follows: \*\*\* p < .001; \*\* p < .01.

significantly more difficult than men (F(1,38) = 4.56, p = .039;  $\eta^2 = 0.107$ ).

## 3.2. Emotion induction and regulation

## 3.2.1. Subjective ratings

ANOVAs revealed significant differences in arousal, valence and success ratings between the ER conditions (main effects of Condition; arousal: F(2.0,151.67) = 106.62, p < .001;  $\eta^2 = 0.584$ , Fig. 3a; valence: F (3,228)=178.35, p < .001;  $\eta^2 = 0.701$ , Fig. 3b; success: F (1.97,149.71) = 53.60, p < .001;  $\eta^2 = 0.414$ ). Post-hoc pairwise comparisons showed that negative pictures were rated as significantly more arousing and less pleasant than neutral pictures (both ps < .001) confirming successful induction of negative emotions via NAPS. In addition, arousal and valence ratings were further modulated by ER attempts. When applying distraction, participants rated negative pictures as significantly less arousing compared to just viewing them (p = .01). However, distraction did not cause significant changes in valence ratings (p > .05). When downregulating emotions via reappraisal participants rated negative pictures as significantly more pleasant relative to simply viewing them (p < .001) while no changes in subjective emotional arousal occurred (p > .05). Participants rated their regulatory performances in all ER conditions as similarly successful (all ps > .05). There were no significant differences between men and women in emotional reactivity and general ER performances (all ps > .05).

#### 3.2.2. Pupil diameter

Analyses of pupillary data showed significant differences in pupil dilations between the ER conditions (F(3,183)=20.49, p < .001;  $\eta^2 = 0.251$ ; Fig. 3c). Pupil size enlargements in response to negative compared to neutral pictures (p < .001) verified that the pupil was modulated by emotional stimulation. In addition, when applying reappraisal to downregulate negative emotions, pupil sizes were significantly increased compared to distracting from (p = .022) and simply viewing (p = .001) negative pictures. This finding suggests that the pupil further enlarged as a function of cognitive effort required for regulatory attempts. No difference in pupil sizes between men and women were found (all ps > .05).

#### 3.3. Stress effects on emotion regulation outcomes

#### 3.3.1. Subjective ratings

Analyses of arousal ratings resulted in a significant three-way interaction between stress, sex and ER condition (F(3,228)=2.88, p = .037;  $\eta^2 = 0.037$ ). *Post-hoc* repeated measures ANOVAs for men and women separately showed that stressed men rated negative pictures as significantly less emotional arousing when applying distraction than controls (Stress x Condition: F(1.94,73.81)=3.96, p = .024;  $\eta^2 = 0.094$ ; t(38) = 2.58, p = .014; Fig. 4a). However, no such stress effect was found in women (p > .05; for a figure, see Supplementary Information A). Moreover, there were no significant stress effects on ER for valence and success ratings (all ps > .05; Fig. 4b+c).



Fig. 3. Emotional ratings and trajectories of pupil diameter with respect to each emotion regulation condition. Box plots depict subjective arousal (a) and valence ratings (b) as well as mean changes in pupil diameter relative to baseline in thousands of arbitrary units (c) over the course of picture presentation for each emotion regulation condition. Medians are marked by black horizontal lines within boxes that range from the first (bottom: Q1) to third quartile (top: Q3). Whiskers that extend from the boxes indicate the minimum and maximum surrounded by outliers defined as 1.5 > interquartile range (Q3) Q1) below Q1 or above Q3. Successful emotion induction was indicated by increased arousal (a) and reduced valence ratings (b) as well as pupil size enlargements (c) after viewing negative compared to neutral pictures. Participants rated negative pictures as significantly less arousing when distracting from the pictures and more pleasant when downregulating negative emotions via reappraisal relative to simply viewing them. Moreover, reappraisal led to significant increases in pupil dilations compared to distracting from and just viewing negative pictures. Significant effects after Bonferronicorrected post-hoc t-tests are marked as follows: \*\*\* *p* < .001; \* *p* < .05.

Given that timing of the ER paradigm relative to stress has been discussed to moderate stress effects on ER outcomes (Langer et al., 2020; Sandner et al., 2021), we reran the reported analyses for each half of the ER paradigm separately (first block: 10–20 min after stress onset; second block: 20–30 min after stress onset) for exploratory purposes. Whereas no main or interaction effects of stress were found in the first half (arousal, valence, success: all *ps* > .05), analyses resulted in a significant three-way interaction between stress, sex and ER condition for arousal ratings in the second half of the ER paradigm (*F*(2.27,173.06)= 3.07, *p* = .043;  $\eta^2$ = 0.039). *Post-hoc* repeated measures ANOVAs showed that stressed males rated negative pictures as significantly less emotional arousing when applying distraction than controls (Stress x Condition: *F* (2.14,81.41)= 4.10, *p* = .018;  $\eta^2$ = 0.097; *t*(38)= 2.64, *p* = .012). No such stress effects were found in women or with respect to other ER conditions or rating scales (all *ps* > .05).

#### 3.3.2. Pupil diameter

Stressed and control participants did not significantly differ in pupil dilations irrespective of the ER condition (p > .05). There were no main effects or interactions with sex or block of the ER paradigm (all ps > .05).

3.4. The relationship between stress biomarkers and emotion regulation outcomes

Overall, correlation analyses with the stress group showed no significant link between cardiovascular responses and ER outcomes of reappraisal and distraction (all ps > .05). To test for possible sexdependent associations, we subsequently conducted correlation analyses separately for men and women. Increases in BPsys were related to heightened subjective emotional arousal after distraction in women (r = 0.543, p = .016). Moreover, HR increases were associated with reduced valence ratings when applying reappraisal (r = -0.590, p = .008) and distraction (r = -0.582, p = .009) indicating a negative association between SNS reactivity and regulatory performances in women but not in men (all ps > .05). In contrast, stress-induced cortisol increases were associated with reduced subjective emotional arousal when applying distraction in men (r = -0.454, p = .045). No such association with cortisol was found in women (p > .05). Exploratory follow-up analyses of each half of the ER paradigm revealed that cardiovascular responses were related to decreased regulatory performances of reappraisal and distraction in women especially in the first half of the paradigm. In contrast, the link between cortisol increases and reduced emotional arousal when men applied distraction was particularly pronounced in the second half of the paradigm (for more details, see Supplementary Information B). No significant correlations between stress biomarkers and ER were found with respect to other outcome



Fig. 4. Stress effects on emotion regulation outcomes in men. Box plots show subjective arousal (a), valence (b), success (c) ratings and mean changes in pupil sizes relative to baseline indexed by the area under the curve with respect to ground (AUCg, d) for each emotion regulation condition (view neutral, view negative, reappraisal, distraction) in stressed (dark blue) and control male participants (light blue). Medians are marked by black horizontal lines within boxes that range from the first (bottom: Q1) to third quartile (top: Q3). Whiskers that extend from the boxes indicate the minimum and maximum surrounded by outliers defined as 1.5 > interguartile range (O3 - O1) below O1 or above O3. Stressed men reported significantly reduced subjective emotional arousal when distracting from negative pictures than controls (a). Significant effects after Bonferroni-corrected pairwise comparisons are highlighted as follows: \* *p* < .05.

measures (valence, success, pupil dilation; all ps > .05).

To examine which stress system primarily drives differences in distraction outcomes between stressed and control participants (reported in Section 3.3.1), we conducted moderated mediation analyses between stress (predictor X), sex (moderator W) and emotional arousal when applying distraction (dependent variable Y) subsequently adding each physiological stress biomarker as possible mediators to the model (see Fig. 5 for paths and statistics). Stress exposure significantly predicted increases in  $BP_{sys}$  (path a=17.932, p < .001),  $BP_{dia}$  (path a=14.438, p < .001), HR (path a=5.063, p = .043) and cortisol (path a=6.811, p < .001). Increases in BP<sub>sys</sub> were positively linked to arousal ratings when applying distraction in women only (path  $w_{my}$ =0.083, p = .011; Fig. 5a). A significant moderated mediation effect of stress on arousal ratings via BP<sub>svs</sub> (a x b x w=1.394, BCa Cl [0.394, 2.476]; Fig. 5b) revealed that stress-induced SNS reactivity predicted enhanced arousal ratings after distraction in women (a x b=1.417, BCa Cl [0.305, 2.540]) but not in men (a x b=-0.037, BCa Cl [-0.868, 0.609]). No direct or indirect effects of stress on distraction outcomes were found when adding BP<sub>dia</sub> or HR as mediators to the model (Fig. 5b-c). In contrast, cortisol increases did significantly relate to reduced subjective emotional arousal when applying distraction (*path* b=-0.115, p = .007; Fig. 5d) which again was moderated by Sex (path  $w_{my}$ =0.132, p = .012). A significant moderated mediation effect (a x b x w=0.898, BCa Cl [0.481, 1.991]) indicated that the negative relationship between stress and arousal after distraction was fully mediated by cortisol increases in men (*a x b*=-0.781, BCa Cl [-1.664, -0.334]) but not in women (*a x* b=0.112, BCa Cl [-0.148, 1.211]). The direct effect of stress on arousal

ratings was no longer significant (path c=0.226, p > .05).

#### 4. Discussion

In the present study, we investigated rapid effects of acute stress on the ability to downregulate negative emotions via reappraisal and distraction in men and naturally cycling women. Stress reduced subjective emotional arousal when men distracted themselves from negative pictures. This effect was critically mediated by increasing cortisol levels suggesting beneficial stress effects on distraction to be predominantly driven by glucocorticoids. In contrast, cardiovascular reactivity was related to reduced regulatory performances of reappraisal and distraction in women. In particular, stress was indirectly linked to heightened emotional arousal when applying distraction via increases in blood pressure in women suggesting the SNS to be associated with regulatory impairments. However, at the group level no detrimental stress effects on ER could be found.

In contrast to our hypothesis, present data provide further evidence for stress to promote regulatory performances particularly when men sought to downregulate emotional arousal via distraction. Importantly, this effect was especially pronounced in the second half of the ER paradigm (20–30 min after stress onset) during which HPA-driven actions become superior via rising cortisol levels. In fact, moderated mediation analyses identified cortisol as a specific mediator of beneficial stress effects on arousal ratings after distraction in men while other physiological stress mediators appeared to be less engaged. These results corroborate with previous studies in which participants were either



Fig. 5. Moderated mediation models of stress effects on arousal after distraction. Mediation graphs depict the relationship between stress (predictor: X), increases in systolic blood pressure (BP<sub>sys</sub>; a), diastolic blood pressure (BP<sub>dia</sub>, b), heart rate (HR, c) and cortisol (d) as potential mediators (M), sex (moderator: W) and subjective emotional arousal when distracting from negative pictures (outcome: Y). Stress caused significant increases in BPsys, BPdia, HR and cortisol (path a effects). Stress was related to enhanced arousal ratings via increases in BPsys in women (a, significant  $w_{my}$  effect, significant moderated mediation: a x b x w effect), whereas BPdia and HR did not act as a significant mediator  $(\mathbf{b} + \mathbf{c})$ . In contrast, cortisol increases significantly mediated dampening stress effects on arousal ratings in men (d, significant path b and  $w_{my}$  effect, significant moderated mediation: a x b x w effect). Significant paths are marked as follows: \* *p* < .05; \*\*\* *p* < .001.

stressed (Langer et al., 2021b) or administered to hydrocortisone (Langer et al., 2021a) prior to an ER task pointing at cortisol to cause regulatory improvements especially for distraction. Imaging data additionally showed cortisol to increase PFC activity during distraction (Jentsch et al., 2019) implying boosted cognitive regulatory capacities under elevated stress hormone levels. It is worth mentioning that stress did not generally benefit ER performances but mainly improved the downregulation of emotional arousal via distraction. Our data thus contradict previous findings showing stress to specifically improve reappraisal performances (Kinner et al., 2014) via boosted cognitive regulatory engagement (Langer et al., 2020). One critical moderator of stress effects on ER is the intensity of the emotional material (Langer et al., 2022, 2021a). Whereas previous studies used stimuli of moderate intensities (Kinner et al., 2014; Langer et al., 2020), we here included high intensity negative pictures. Typically, distraction is more successful than reappraisal when dealing with high intensity emotions (Shafir et al., 2015) and requires less cognitive resources (Strauss et al., 2016). Stress has been shown to favor the choice of low demanding, though efficient cognitive strategies (e.g., Schwabe and Wolf, 2013) such as distraction when downregulating high intensity emotions (Langer et al., 2022). This stress-induced shift towards low demanding strategies has been argued to rescue regulatory performances especially under stress states when prefrontal control resources are limited. One may therefore hypothesize that stressed participants were more motivated to put effort in distracting from high intensity stimuli than reappraising the presented situation ultimately leading to better regulatory outcomes. However, no statistically meaningful differences in pupil dilations during distraction between stressed and control participants occurred. Importantly, pupil sizes not only fluctuate as a function of regulatory effort but also of emotional arousal (Bradley et al., 2008). Dampening effects of stress on emotional arousal might thus have counteracted effort-driven increases in pupil dilations particularly when applying distraction. To overcome this ambiguity, ER research may benefit from

electroencephalography (EEG) recordings providing distinct indices of emotional activation and cognitive regulatory effort (Shafir et al., 2015). Together, future EEG studies varying the intensity of the emotional material are warranted to explore its role for stress effects on the effectiveness of specific ER strategies.

In the expected direction, cardiovascular responses to stress were related to reduced regulatory performances of reappraisal and distraction in women hinting at the SNS to drive regulatory impairments. These findings corroborate with previous studies showing stress to rapidly impair cognitive attempts to downregulate anger (Zhan et al., 2017) and conditioned fear (Raio et al., 2013) critically associated with sAA increases. Suggesting possible underlying mechanisms, there is pharmacological evidence for the noradrenergic system to boost amygdala responses to fear signals (Onur et al., 2009). As such, the SNS may increase emotional reactivity that in turn might impede cognitive regulatory attempts immediately after stress exposure. In line with this idea, stress has been shown to impair PFC functioning (Arnsten, 2009) probably due to strengthened inhibitory input from emotion-related limbic regions (Hermans et al., 2011). In contrast to our expectations, however, we did not find any detrimental stress effects on ER outcomes at the group level suggesting other stress mediators to act in the opposite direction. As mentioned before, the stress hormone cortisol has repeatedly been linked to enhanced reappraisal and distraction performances (Langer et al., 2021a, 2021b, 2020) implying dominance of the HPA axis to guide ER improvements. Here, ER performances were assessed in a time window from 10 to 30 min after stress onset. Stimulation of the HPA axis provokes a gradual increase in cortisol levels reaching its peak about 20-30 min after stress onset (Allen et al., 2014). Hence, it is reasonable to assume that GC actions may already have come into play when ER was measured. Accordingly, null effects of stress on ER might result from concurrent activation of both stress systems (i.e., SNS and HPA axis) in this specific time window, allowing potential opposing actions of catecholamines and GCs on the emotion regulatory network to cancel out any group differences. Alternatively, the remaining SNS activation might have been too low during the ER paradigm to dominate beginning HPA actions. In favor of this idea, cardiovascular activity was already back at baseline when the ER paradigm started. It has to be noted though that noradrenergic actions on cognitive functions result from stimulation of the locus coeruleus (LC) in the brainstem projecting to numerous (sub)cortical structures (Roosevelt et al., 2006). However, both pathways (central and peripheral) rely on vagus nerve stimulation and are thus interrelated (Capilupi et al., 2020). Therefore, one may assume that catecholaminergic actions in the brain were at least somewhat flattened at paradigm onset and gradually reduced over time. Along this line, it can be speculated that noradrenergic activity was either too weak per se to hamper cognitive regulatory processes or not sufficiently powerful anymore when ER was measured. Future studies may benefit from a pharmacological suppression of the HPA axis in order to isolate and boost SNS reactivity to stress (Ali et al., 2020). Alternatively, study protocols in which the ER task is scheduled during or in anticipation of stress exposure may ensure that the SNS is still predominantly active.

As hypothesized, we found sex differences in stress effects on ER and their association to neuroendocrine responses. In accordance with previous studies (Langer et al., 2020; Ma et al., 2017), stress improved ER performances via cortisol increases in men but not in naturally cycling women. Importantly, women were tested in the mid-luteal phase of the menstrual cycle only in which progesterone peaks and estradiol levels are typically moderate (Allen et al., 2016). Elevated levels of progesterone have been linked to reduced receptor affinity (Turner, 1997) and sensitivity to glucocorticoids (Rohleder et al., 2001). Smaller stress effects on ER in women may thus result from gonadal steroids reducing sensitivity and/or binding capacities of GC receptors to rising cortisol levels. These sex differences in turn might affect the duration and power of SNS dominance. Of note, the female hormone progesterone was shown to increase amygdala reactivity to threatening stimuli (Van Wingen et al., 2008). Moreover, women are more emotionally reactive (Bradley et al., 2001) and exhibit greater activation of the LC arousal system after stress (Bangasser et al., 2019) than men. Together, these findings indicate a larger proportion of SNS to HPA-driven actions in women probably increasing their sensitivity to detrimental stress effects on ER performances. Given large fluctuations in reproductive hormones over the course of the menstrual cycle (Allen et al., 2016), future studies could compare women in different cycle phases to shed light on stress-sex hormone interactions on ER processes.

Some limitations are worth mentioning. First, reappraisal did not significantly reduce subjective emotional arousal but increased valence ratings relative to simply viewing negative pictures, while distraction did not lead to significant increases in picture valence but succeeded in downregulating emotional arousal. These inconsistencies are most probably due to differences in the potency of each strategy to exert its effects on each rating scale. Whereas instructions to positively reappraise negative pictures might have changed arousal in a positive direction, distraction asked participants to shift their attention towards neutral thoughts thereby being less potent to influence picture valence. Second, although being frequently used in laboratory ER research, this paradigm is somewhat artificial and not fully comparable with emotional trigger and regulatory requirements in everyday life. Third, despite measurement of cardiovascular activity as a valid and wellestablished marker of SNS activation, we did not directly assess levels of catecholamines such as adrenaline and noradrenaline.

In conclusion, this study showed stress to rapidly improve the ability to downregulate emotional arousal via distraction in men which was fully mediated by cortisol. In contrast, SNS reactivity was linked to decreased regulatory performances in women. Even though direct stress effects on ER were smaller than expected and our findings call for future replication, present data tentatively indicate opposing rapid effects of the two major stress systems on the cognitive control of negative emotions that are critically moderated by sex. This study contributes to a better understanding of the neuroendocrinological mechanisms of stress effects on ER that may help to develop adequate preventive and curative interventions of stress- and emotion-related disorders.

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## **Author Contributions**

KL designed the work, acquired, analyzed and interpreted data, drafted the manuscript, prepared figures and edited the manuscript. OTW and VLJ designed the work, interpreted data, edited and revised the manuscript.

#### Conflict of interest statement

The DFG has no role in study design, collection, analysis and interpretation of data, writing of the manuscript or in the decision to submit the paper for publication. All authors reported no biomedical financial interests or potential conflicts of interest.

# Data Availability

The data that support the findings of this study are available at the Open Science Framework (OSF) under https://osf.io/x95sj/.

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#### Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.psyneuen.2023.106054.

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