Biological Psychology 167 (2022) 108212

Contents lists available at ScienceDirect

Biological Psychology

journal homepage: www.elsevier.com/locate/biopsycho

Emotion regulation and diurnal cortisol: A longitudinal study of early adolescents

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ARTICLE INFO

Keywords:
Diurnal cortisol
Adolescence
Stress
Emotion regulation
COVID-19
Longitudinal

ABSTRACT

Aberrant patterns of diurnal cortisol, a marker of stress reactivity, predict adverse physical and mental health among adolescents. However, the mechanisms underlying aberrant diurnal cortisol production are poorly understood. Thus, the objective of this study was to investigate, for the first time, whether the core emotion regulation (ER) strategies of rumination (brooding, reflection), reappraisal, and suppression were prospectively associated with individual differences in diurnal cortisol during the COVID-19 pandemic, a period of significant stress. A community sample of 48 early adolescents (M_{age} = 13.45; 60% males) was recruited from British Columbia, Canada. Participants completed ER measures before the pandemic, and diurnal cortisol was assessed by collecting eight saliva samples over two days during the first COVID-19-related lockdown in the region. As expected, brooding predicted elevated waking cortisol and a blunted cortisol awakening response (CAR), whereas reflection predicted lower waking cortisol and suppression predicted a steeper CAR. Unexpectedly, reappraisal was not associated with diurnal cortisol production. Results indicate that ER strategies may represent a mechanism underlying individual differences in biological markers of wellbeing during stress.

1. Introduction

Adolescence is a period of rapid development characterized by neural and physiological plasticity before biological responses become stable patterns in adulthood (e.g., Ganzel, Kim, Gilmore, Tottenham, & Temple, 2013). Given that this is a period when vulnerabilities become embedded, adolescence is a key developmental stage for understanding aberrant patterns of biological stress reactivity, particularly dysregulated diurnal cortisol production. Among youth, dysregulated diurnal cortisol production can emerge during times of stress (Stroud, Vrshek-Shallhorn, Norkett, & Doane, 2019) and can have profound and long-term implications on physical and mental health (Adam, Quinn, Tavernier, McQuillan, Dahlke, & Gilbert, 2017; LeMoult, 2020). Importantly, significant individual differences in diurnal cortisol trajectories during stress are evident among youth (Starr, Dienes, Li, & Shaw, 2019). Thus, it is critical to identify factors that make adolescents more or less prone to the development of aberrant cortisol secretion in the context of life stress.

One intriguing possibility is that emotion regulation (ER) influences diurnal cortisol production (LeMoult, 2020). ER comprises strategies individuals use to modulate their emotional responses to stressors and, as such, ER has a significant influence on emotional wellbeing (Gross, 1998). There is also emerging evidence that ER is associated with acute physiological stress responses during laboratory stress-inductions (e.g., Balzarotti, Biassoni, Colombo, & Ciceri, 2017). However, ER strategies vary markedly in their effectiveness, and adaptive as well as adverse affective and physiological outcomes have been documented (Webb, Miles, & Sheeran, 2012). This suggests that some strategies may represent markers of risk, whereas others may represent markers of resilience to stress.

Despite mounting evidence for the impact of ER on emotional and physiological outcomes, only a handful of studies have examined whether specific ER strategies are associated with indices of diurnal cortisol. In a sample of adolescent girls, responding to interpersonal stress with active coping – a construct similar to ER – was cross-sectionally associated with adaptive diurnal cortisol production, evidenced by lower total cortisol output, a lower cortisol awakening response (CAR), and a steeper daytime slope (Sladek, Doane, & Stroud, 2017). Similarly, Gilbert, Mineka, Zinbarg, Craske, & Adam, 2017 found specific cross-sectional associations of various ER strategies (i.e., problem solving, disengagement, and emotional expression/support seeking) with diurnal cortisol rhythms using experience sampling in young

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https://doi.org/10.1016/j.biopsycho.2021.108212
Received 13 April 2021; Received in revised form 20 October 2021; Accepted 20 October 2021
Available online 23 October 2021

adults. Furthermore, Otto, Sin, Almeida, and Sloan (2018) examined the association of suppression (i.e., inhibiting emotionally expressive behavior) and reappraisal (i.e., redefining the meaning of a situation to change its emotional impact) with the CAR and diurnal slope in a sample of adults. They found that greater suppression was cross-sectionally associated with a steeper CAR and flatter diurnal slope, whereas reappraisal did not predict diurnal cortisol parameters. Researchers have also documented associations between rumination, another central ER strategy, and diurnal cortisol. For example, Sladek, Doane, and Breitenstein (2020) documented that greater rumination on a given day was associated with higher waking cortisol the following day, and greater than usual rumination on a low stress day predicted a flatter daytime slope the following day in a sample of young adults. In contrast, Hilt, Sladek, Doane, and Stroud (2017) reported that greater than usual rumination was associated with a lower CAR the following day, and trait rumination was associated with *lower* average waking cortisol and a flatter daytime slope. Mixed findings have also been reported for the association of rumination with the CAR (see Zoccola & Dickerson, 2012). Importantly, studies have collapsed across distinct subtypes of rumination, which may contribute to inconsistent results. Given that the brooding subtype of rumination involves passively and repetitively comparing one's situation to an unachieved standard, whereas the reflection subtype encompasses a purposeful and active turning inward to gain insight into problems, examining rumination as a unitary construct may obscure differential effects of brooding versus reflection, as has been found in mood outcomes research (e.g., Cox, Funasaki, Smith, & Mezulis, 2012).

Although emerging evidence suggests that ER may be associated with diurnal cortisol production, there are several major gaps in the literature. First, a paucity of research has examined the relative association of core ER strategies with diurnal cortisol. Given that individuals use a range of ER strategies both within and across contexts (Szasz, Coman, Curtiss, Carpenter, & Hofmann, 2018), it is important to take multiple strategies into account to assess the relative and unique contribution of central ER strategies on stress reactivity. Second, outside of experience sampling designs, the association of core ER strategies with diurnal cortisol has not been assessed longitudinally, which is essential for investigating longer-term associations and establishing temporal precedence. Third, the relative and longitudinal association of core ER strategies with diurnal cortisol has not been examined among adolescents. Identifying factors associated with diurnal cortisol when patterns of stress reactivity are still malleable represents a valuable opportunity for informing prevention efforts aimed at making lasting impacts on health. Fourth, associations of these ER strategies with diurnal cortisol rhythms have not been investigated during a time of marked stress. When assessed during an ongoing stressor, diurnal cortisol represents an important marker of stress-related HPA-axis activation (Miller, Chen, & Zhou, 2007), suggesting that it is crucial to identify individual differences in diurnal cortisol patterns during stressful periods, when diurnal cortisol patterns may be a particularly strong predictor of health outcomes.

The ongoing severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2, or COVID-19) pandemic represents an ideal stressor to assess diurnal cortisol in adolescents. Physical distancing restrictions have resulted in major changes to education, recreation, socializing, and home life. Adolescents are reporting elevated perceived stress (Gotlib, Borchers, Chahal, Gifuni, & Ho, 2021) and are being exposed to objective stressors, including those related to increases in domestic conflict, financial hardship, and health (Guessoum et al., 2020). Youth are also experiencing a loss of protective factors, including educational and extra-curricular activities, social support, and access to supportive adults outside the home (e.g., Guessoum et al., 2020). Moreover, researchers examining pre-COVID-19 ER as a predictor of mental health symptoms during the pandemic have conceptualized the pandemic as a chronic stressor that catalyzed observed changes in symptoms (Breaux et al., 2021). For adolescents living in the Lower Mainland of British Columbia, Canada, impacts of the pandemic on daily life and routines were particularly marked during the first months of the pandemic, when schools and all non-essential businesses and services were closed, physical distancing restrictions were in place, and the region was under lockdown.

As such, the objective of the present study was to assess, for the first time, whether diverse ER strategies prospectively predicted diurnal cortisol production among early adolescents during a significant stressor – the COVID-19 pandemic. Trait ER was assessed immediately before the start of the pandemic (December 2019), and diurnal cortisol was assessed early in the pandemic (June/July 2020), when the strictest physical distancing measures were in place in British Columbia. We hypothesized that reappraisal and reflection, ER strategies that have been associated with effective regulation of affect and physiological responses (e.g., Gross & John, 2003; Jentsch & Wolf, 2020; Kocsel, Köteles, Szemenyei, Szabó, Galambos, & Kökönyei, 2019), would be associated with a healthy diurnal cortisol profile marked by lower cortisol at waking, a reduced CAR, and a steeper decline in daytime slope (Adam et al., 2017; Chida & Steptoe, 2009; Shibuya et al., 2014). In contrast, we hypothesized that brooding and suppression, strategies that have been shown to increase negative affect and physiological responsivity to laboratory stressors (Gross & John, 2003; Jentsch & Wolf, 2020; Woody, Burkhouse, Birk, & Gibb, 2015), would be prospectively associated with an activated diurnal cortisol rhythm characterized by higher cortisol at waking and sustained cortisol elevations throughout the day (evidenced via a flatter daytime slope). In terms of CAR, given the effortfulness of concealing emotions via suppression, we hypothesized that suppression would be associated with an elevated CAR. Conversely, because brooding involves repetitive over-engagement with negative information, we expected that it would overexert the HPA-axis over time and thus, hypothesized that brooding would be prospectively associated with a blunted CAR.

2. Method

2.1. Participants

Participants were recruited for the UBC Study on Adolescents via flyers, local media, and online advertisements. Given the broader goals of the study and the importance of assessing cortisol dysregulation in early adolescence, before it becomes resistant to change, eligibility criteria included being 11–12 years old and fluent in English. Ineligibility criteria included symptoms of a current substance use disorder, lifetime history of mania or psychosis, severe impairment caused by a learning disability, history of severe head trauma, or having an endocrine disorder. Of 79 youth who entered the study, 48 youth completed ER measures in December 2019 and provided saliva samples in June/July of 2020 (when the strictest COVID-19 physical distancing measures were in place in British Columbia). An a priori power analysis indicated that with $\alpha = 0.05$, and power $= 0.80$, this sample size is adequate to detect a moderate effect ($d = 0.35$) based on expected correlations between ER and cortisol, as informed by previous research (e.g., Hilt et al., 2017; Otto et al., 2018). See Table 1 for participant characteristics.

2.2. Context of the COVID-19 pandemic

Cortisol data were collected in June/July of 2020, three months after a state of emergency was declared in British Columbia, Canada. During this time the Lower Mainland of British Columbia was under lockdown – in-person school classes were canceled, non-essential businesses and services were closed, and the strictest physical distancing measures to date in the region were in place. The COVID-19 pandemic is often conceptualized as a markedly stressful period (Breaux et al., 2021). To examine whether this was the case for youth in our sample, participants rated on a Likert scale from 1 (*Not at all*) to 5 (*Very much*) how much their stress increased due to the COVID-19 pandemic and the degree to

Table 1
Participant characteristics.

Variable	Participants $n = 48$
Sex, *n*	
Male	29
Female	19
Gender, *n*	
Boy	29
Girl	19
Age, *M* (*SD*)	13.45 (0.32)
Ethnicity, *n*	
Chinese	10
European	39
Filipino	1
Indigenous	2
Japanese	4
Korean	2
Latinx	2
South Asian (e.g., Indian, Pakistani, Sri Lankan)	2
West Asian (e.g., Iranian, Afghan)	1
Household Income, *n*	
\$20,000 to \$59,999	4
\$60,000 to \$99,999	10
\$100,000 to \$119,999	8
\$120,000 to \$159,999	11
\$160,000 and over	13
Don't know	1
Prefer not to answer or Missing	1
Parents' Education Level, *n*	
Less than high school graduation	1
High school diploma	5
ollege diploma/certificate	18
Bachelor's degree	40
Master's degree	18
Doctorate degree (PhD)	10
Missing	2
Sibling living at home, *n*	
Yes	37
No	11
Increases in stress due to COVID-19, *M* (*SD*)	2.35 (1.36)
Degree life became more negative due to COVID-19, *M* (*SD*)	2.63 (1.27)
Emotion Regulation, *M* (*SD*)	
Brooding	12.69 (3.95)
Reflection	11.23 (3.74)
Suppression	10.85 (3.40)
Reappraisal	20.02 (4.09)
Salivary Cortisol, *M* (*SD*) (nmol/l)	
Wake	8.74 (4.22)
Wake + 30 minutes	10.48 (5.49)
3:00 pm	2.98 (2.06)
Bedtime	1.13 (1.84)

Note. Participants were able to choose more than one option when reporting their ethnicity. $n = 94$ parents.

which their life became more negative as a result of the pandemic. One-sample *t*-tests were conducted to examine whether mean scores differed significantly from 1, which on these scales, indicated that participants' stress did not increase, or their life did not become more negative, due to the pandemic. As shown in Table 1, participants reported a significant increase in stress, $M = 2.35$; $SD = 1.36$, $t(47) = 6.90$, $p < .001$, and indicated that their lives became significantly more negative due to the pandemic, $M = 2.63$; $SD = 1.27$, $t(47) = 8.90$, $p < .001$.

2.3. Measures

2.3.1. Suppression and reappraisal

The Emotion Regulation Questionnaire for Children and Adolescents (ERQ-CA; Gullone & Taffe, 2012) was used to assess suppression and reappraisal. The ERQ-CA is a 10-item self-report measure that consists of a 4-item suppression subscale and a 6-item reappraisal subscale. Participants rate items on a 5-point Likert scale, with higher scores indicating greater tendency to engage in suppression and reappraisal. The ERQ-CA has excellent reliability and validity (Gullone & Taffe, 2012), and Cronbach's alpha was .80 for the suppression subscale and .83 for the reappraisal subscale.

2.3.2. Brooding and reflection

The Ruminative Responses Scale-Adolescent Version (RRS-A; Burwell & Shirk, 2007) was used to measure brooding and reflection. This 10-item self-report questionnaire includes two 5-item subscales that assess brooding, repetitively and passively thinking about problems and feelings, and reflection, purposefully analyzing experiences to gain insight. Items are rated on a 4-point Likert scale, with higher scores indicative of greater brooding or reflection. The RRS-A has strong psychometric properties (Burwell & Shirk, 2007). Cronbach's alpha in the current study was .80 for the reflection subscale and .83 for the brooding subscale.

2.3.3. Diurnal cortisol

Diurnal cortisol data were collected in accordance with consensus guidelines for the assessment of CAR (Stalder et al., 2016). Participants collected 8 saliva samples using Sarstedt Salivettes (Sarstedt, Numbrecht, Germany) at home over the course of two weekdays – immediately upon waking, 30 minutes after waking, at 3:00 pm, and at bedtime. Youth were instructed not to exercise, eat, drink, smoke, vape, chew gum, or brush their teeth two hours before collecting a sample. To promote adherence, participants and their parents received detailed verbal and written instructions on the saliva collection protocol, including the importance of collecting the first sample immediately upon awakening, as part of their collection kits. Participants used a self-report diary to report compliance with instructions and to record their time of awakening and the exact time of saliva collection. Furthermore, participants were informed that accurate completion of the compliance assessments was critical and that their responses would not have implications for their financial compensation or future participation. Using these protocols in the same age range, we found that self-reported timing of collection did not differ from timing recorded using Smart Caps (LeMoult, Chen, Foland-Ross, Burley, & Gotlib, 2015). Participants stored Salivettes in their home freezers until they were transferred on ice to a −30 °C freezer.

Concentrations of cortisol were measured using a chemiluminescence immunoassay commercial kit from Immuno-Biological Laboratories (Hamburg, Germany) with high sensitivity set to 0.015 mg/dl. Samples were assayed in large batches to reduce interassay variability and control samples were included to evaluate variability. Each sample from the same child was assayed in duplicate. Intra and interassay coefficients of variation were below 9%. Given the characteristic skew of cortisol data, values were logarithmically transformed.

2.3.4. Potential covariates

Consistent with expert recommendations (Stalder et al., 2016), variables known to influence diurnal cortisol were assessed: psychotropic and non-psychotropic medications, sex, gender, age, parent-reported household income (as an indicator of socioeconomic status), minutes between midnight and the first saliva sample, self-reported deviations from the saliva sampling protocol, days between assessment of ER and cortisol, presence of a pre-COVID psychiatric diagnosis, and pubertal stage (assessed using Tanner Staging; Marshall & Tanner, 1968). Furthermore, pandemic-related variables were collected, including whether youth were living with siblings, self-reported increases in stress due to the COVID-19 pandemic, and subjective evaluation of how much participants' lives became more negative due to the COVID-19 pandemic. Covariates related to sampling day (i.e., season, weekday vs. weekend) were controlled for experimentally by asking all participants to collect saliva on a weekday and by collecting all saliva data during the summer season.

2.5. Procedure

This study was approved by the University of British Columbia Behavioral Research Ethics Board. After phone screening for eligibility, participants attended the laboratory with their primary caregiver. After obtaining consent from caregivers and assent from adolescents, adolescents completed a demographic questionnaire as well as measures as part of the UBC Study on Adolescents. Participants completed online questionnaires assessing ER from home in December 2019, prior to the onset of the COVID-19 pandemic. In May of 2020, caregivers were contacted, and they and their children were invited to participate in an additional wave of data collection related to the pandemic. After obtaining caregiver consent and youth assent, in June and July of 2020, at the height of social-distancing requirements in British Columbia, participants provided saliva samples at home and completed online questionnaires assessing potential cortisol-related covariates.

2.6. Statistical analysis

Hierarchical Linear Modelling (HLM 8.1; Raudenbush & Congdon, 2021) was used given the nested structure of the data, with repeated measurements of cortisol nested within participants. We used full information maximum likelihood to calculate Akaike's Information Criteria (AIC) and restricted maximum likelihood (REML) to estimate model parameters. HLM is robust to sample sizes (Maas & Hox, 2005). Furthermore, rather than adjusting statistical significance for multiple analyses, which creates significant problems for statistical inference (e.g., increasing Type II error rate; Perneger, 1998), we used REML and robust standard errors to reduce bias and provide more conservative parameter estimates (Raudenbush & Bryk, 2002). We conducted a hierarchical model of diurnal trajectories of cortisol within persons as a function of time, which allows for unevenly spaced measurement occasions. The model examined the relative contribution of ER strategies assessed in December 2019 to later trajectories of diurnal cortisol in June/July 2020. Specifically, this hierarchical linear model was conducted examining the association between trajectories of diurnal cortisol at Level 1 and forms of ER (brooding, reflection, reappraisal, and suppression) at Level 2.

3. Results

3.1. Participants

Participants in the current study who completed ER measures in December 2019 and collected diurnal cortisol in June/July 2020 ($n = 48$) were compared to participants from the total study sample who did not participate in one or more of these data collection points ($n = 31$; see Table S1 of the online supplement for characteristics). There were no differences in sex, $\chi^2(1, N = 79) = 1.11$, $p = .293$, gender, $\chi^2(1, N = 79) = 1.11$, $p = .293$, ethnicity, $\chi^2(3, N = 79) = 6.14$, $p = .105$, or age, $t(75) = -0.30$, $p = .765$. Furthermore, there were no differences in ER across sex or gender, $ps > .164$, among participants in the current study.

3.2. Diurnal cortisol pattern

We first examined the average trajectory of diurnal cortisol production across the day. Participants' level of cortisol was significantly different than zero at waking, $t(47) = 29.24$, $p < .001$, significantly increased from waking to 30-minutes post-waking, $t(47) = -2.17$, $p = .035$, and significantly decreased from 30-minutes post-waking to the late evening, $t(47) = 22.51$, $p < .001$. Based on significant diurnal changes, visual inspection of the data (see Table 1), and study-specific hypotheses, we used a piecewise hierarchical model to capture levels of cortisol at waking, the slope of change in cortisol from waking to 30-minutes post-waking (i.e., CAR), and the slope of change in cortisol from 30-minutes post-waking to the late evening (i.e., the cortisol daytime slope) at Level 1. Piecewise models have been used in numerous studies of diurnal cortisol (e.g., Charles, Mogle, Piazza, Karlamangla, & Almeida, 2020; Jopling, Rnic, Tracy, & LeMoult, 2021; King et al., 2017; LeMoult et al., 2015), and are recommended when there is evidence of a nonlinear trajectory of change and researchers wish to examine correlates of different periods of the growth trajectory (Raudenbush & Bryk, 2002).

3.3. Potential covariates

Before testing the effects of ER strategies on each of the Level 1 parameters, we tested a series of variables as possible covariates in relation to participants' diurnal cortisol profiles: psychotropic and non-psychotropic medications, sex, gender, age, household income, minutes between midnight and the first saliva sample, days between assessment of ER and cortisol, whether the participant deviated from the saliva collection protocol, presence of a pre-COVID psychiatric diagnosis, whether youth were living with siblings, subjective evaluation of how much participants' lives became more negative due to COVID-19, and self-reported increases in stress due to COVID-19.[1] Deviations from the collection protocol was associated with higher cortisol levels at waking, $B = 0.44$, $t(40) = 3.08$, $p = .004$, and a steeper daytime slope, $B = -0.001$, $t(40) = -2.11$, $p = .042$. Stress was associated with a steeper daytime slope, $B = -0.0003$, $t(42) = -2.61$ $p = .012$. No other potential covariates were associated with cortisol production at waking, CAR, or daytime slope, $ps > .135$. Thus, in order to retain the most parsimonious model, as recommended by Raudenbush and Bryk (2002), only deviations from the collection protocol and stress were included as covariates in the relevant Level 2 equation(s).

3.4. ER and the diurnal cortisol pattern

We next tested the effects of ER strategies on each of the Level 1 parameters; significant covariates were included in the relevant Level 2 equation(s). Greater use of reflection was associated with lower cortisol at waking, $B = -0.05$, $t(40) = -3.24$, $p = .002$, while greater brooding was associated with higher cortisol at waking, $B = 0.08$, $t(40) = 5.70$, $p < .001$. Greater brooding was also associated with a blunted CAR, $B = -0.002$, $t(41) = -2.61$, $p = .013$, whereas greater use of suppression was associated with a steeper CAR, $B = 0.002$, $t(41) = 2.39$, $p = .021$. ER strategies were not associated with the daytime slope of cortisol, $ps > .074$. All results are presented in Table 2, and significant associations are depicted in Fig. 1 (see Fig. S1 in the online supplement for figures of nonsignificant results).

4. Discussion

The present study examined whether diverse ER strategies predicted diurnal cortisol among early adolescents during a significant stressor – the first months of the COVID-19 pandemic. ER was assessed in late 2019 before the start of the pandemic, establishing a robust baseline. Findings indicated that, as hypothesized, ER prospectively predicted diurnal cortisol production during stress; however, the direction of results differed based on the specific ER strategy.

Reflection and brooding demonstrated opposing effects on cortisol concentrations at awakening. As hypothesized, reflection was associated

[1] We did not include pubertal stage as a potential covariate because 10.4% of our sample chose not to provide data on pubertal timing. Therefore, inclusion of this variable in the main analyses would substantially reduce power and alter the composition of our sample. When the association between pubertal stage and cortisol patterns was tested in the subset of participants who reported this data, pubertal stage was associated with cortisol at waking, $B = 0.20$, $t(37) = 2.72$, $p = .010$. However, including pubertal stage in the main analyses did not change the pattern of findings.

Table 2
Emotion regulation strategies predicting cortisol response to stress.

	Coeff	*SE*	*t*	*p*
Waking				
Intercept	**1.95**	**.051**	**38.64**	**.000**
Deviations from saliva collection protocol	**.40**	**.118**	**3.37**	**.002**
Brooding	**.08**	**.013**	**5.70**	**.000**
Reflection	-.05	**.015**	**-3.24**	**.002**
Reappraisal	.01	.012	0.87	.388
Suppression	-.02	.020	-0.92	.365
Cortisol Awakening Response				
Intercept	.002	.002	1.17	.248
Brooding	**-.002**	**.001**	**-2.61**	**.013**
Reflection	-.0001	.001	-0.20	.846
Reappraisal	-.0001	.001	-0.17	.864
Suppression	**.002**	**.001**	**2.39**	**.021**
Daytime Cortisol Slope				
Intercept	**-.003**	**.0001**	**-25.35**	**.000**
Deviations from saliva collection protocol	**-.001**	**.0003**	**-2.88**	**.007**
Stress	**-.0003**	**.0001**	**-3.61**	**.000**
Brooding	.0001	.00004	1.84	.074
Reflection	.0001	.00004	1.65	.107
Reappraisal	.00003	.00004	0.70	.490
Suppression	-.00002	.00004	-0.50	.619

Note: Coeff = regression coefficient; *SE* = standard error.
Significant *p*-values are presented in bold.

with lower cortisol at waking, consistent with low HPA-axis activity. This suggests that, consistent with a transactional stress-coping framework (e.g., Lazarus & Folkman, 1984), reflection may have helped to prevent the experience of the pandemic from exceeding coping resources. Conversely, brooding was associated with higher cortisol levels at waking, which has been associated with risk for depression (Dienes, Hazel, & Hammen, 2013), and is indicative of a hyperactivated HPA-axis. Although past research has examined the association of rumination with cortisol at waking, findings have been inconsistent, with some reporting that rumination is associated with lower waking cortisol (Hilt et al., 2017), and others finding no effects (Zoccola et al., 2017). Importantly, past studies have examined total rumination scores, without differentiating brooding versus reflection. The current findings show that each facet of rumination has opposing effects, highlighting the importance of examining reflection and brooding separately. Furthermore, these findings add to growing evidence that low waking cortisol is a component of a healthy diurnal profile, as indicated by research documenting that elevated morning cortisol predicts the onset of depression (Harris et al., 2000), and that higher waking cortisol is associated with greater perceived stress, greater loneliness, and with being at-risk for depression (Dienes et al., 2013; Jopling et al., 2021; Sladek et al., 2020).

Consistent with hypotheses, brooding and suppression were both associated with an aberrant CAR during the pandemic – brooding predicted a blunted CAR, whereas suppression was associated with a steeper CAR. The current findings extend past cross-sectional research in adults that reported that suppression was associated with a steeper CAR (Otto et al., 2018). They are also the first to demonstrate that brooding was associated with a blunted CAR, although this should be interpreted in the context of the finding that brooding predicted higher waking cortisol, which may have contributed to a smaller change in the first 30 minutes after waking. While it may appear contradictory that suppression and brooding – which both have been conceptualized as maladaptive emotion regulation strategies (Gross & John, 2003; Treynor, Gonzalez, & Nolen-Hoeksema, 2003) – predicted opposing effects on CAR, divergent outcomes likely resulted from marked differences in the phenomenology of these ER strategies. Brooding involves prolonged, passive over-engagement with affective content, which, over time, may overexert the HPA-axis, ultimately leading to a hypoactive CAR. Conversely, suppression encompasses active concealment of emotional responses. Effortful attempts to suppress emotions may intermittently increase HPA-axis activity, evidenced here by a steeper CAR. Moreover, when individuals behaviorally disengage from emotional experiences, these may instead be expressed physiologically (Richards & Gross, 1999), resulting in greater cortisol concentrations. Altogether, findings suggest that brooding and suppression interfered with effective coping, thereby exceeding resources to meet the demands of the pandemic. Consistent with transactional stress-coping models (Lazarus & Folkman, 1984), this likely resulted in stress, as reflected by diurnal cortisol profiles.

Unexpectedly, reappraisal was not associated with any indices of

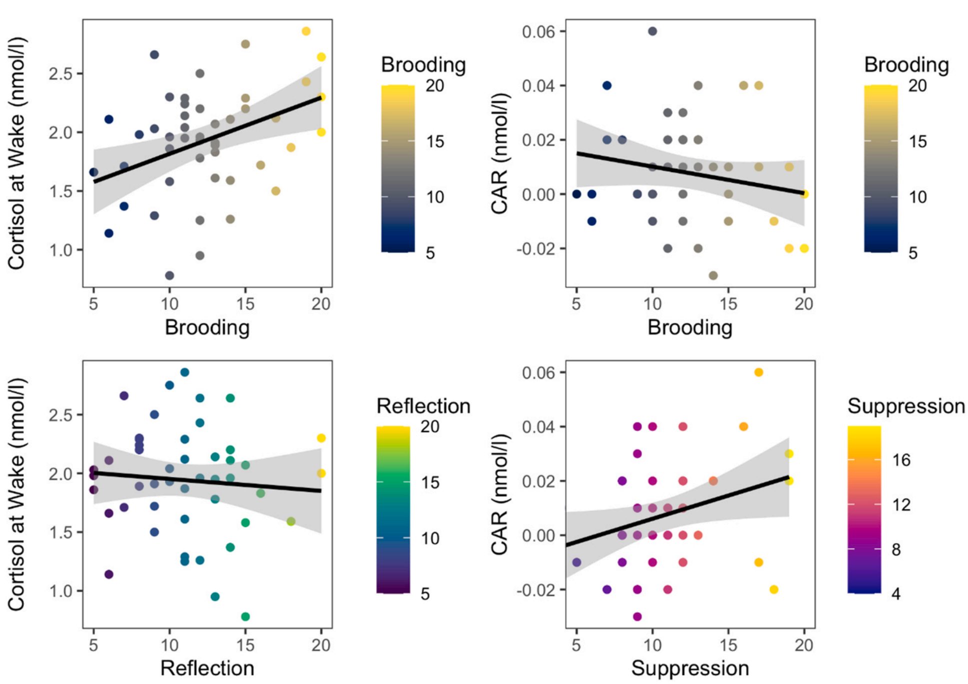

Fig. 1. *Associations between Emotion Regulation in December 2019 and Diurnal Cortisol in June/July 2020. Note.* Cortisol values were logarithmically transformed to account for skew. CAR = cortisol awakening response.

diurnal cortisol. Although reappraisal has been linked to increased momentary cortisol following an acute stressor (Raymond, Marin, Juster, & Lupien, 2019), past research has found that it does not predict diurnal cortisol in adults (Otto et al., 2018). The current study replicates this finding in adolescents. It is possible that during periods of prolonged stress, other, less effortful ER strategies (e.g., acceptance; Troy, Shallcross, Brunner, Friedman, & Jones, 2018) may be more effective in modulating biological stress responses.

In addition to addressing key gaps in the ER and stress literature, this research answers an urgent call to assess mechanisms underlying endocrinological functioning during the pandemic (Dantzer, Heuser, & Lupien, 2020). This study is among the first to examine predictors of biological functioning during COVID-19. Importantly, given that various forms of childhood adversity show broad and nonspecific neurobiological and behavioral effects, and given that stress-response systems are sensitive to many types of adversity (Smith & Pollak, 2021), there is strong empirical reason to expect that the current findings and implications are relevant to other periods of stress outside of the context of the first months of the COVID-19 pandemic in British Columbia. Moreover, our findings indicate that there is empirical cause for concern for the immediate and long-term physical and psychological wellbeing of youth exposed to stress. However, the present findings are also cause for optimism. First, results suggest that youth who use reflection show a diurnal cortisol pattern associated with resilience. Second, findings have implications for identifying youth who are at risk for aberrant biological responses to stress and who may benefit from early intervention. ER strategies are amenable to change, and developing skills in effectively selecting and implementing ER strategies may foster healthier diurnal cortisol rhythms. Indeed, there are evidence-based interventions available that target ER (Watkins, 2018; Greenberg, 2011), some of which have been associated with reductions in cortisol (Urizar & Muñoz, 2011). Given that the current findings documented prospective associations between ER and biological stress responses known to significantly influence long-term mental and physical health, this study provides support for interventions that target ER. By changing trait ER, such interventions have the potential to shift biological responses to stress, including but not limited to the COVID-19 pandemic. Implications of our findings for intervention efforts are particularly relevant for youth, as biological stress responses have not yet become highly embedded at this developmental stage. Moreover, given that early adolescence is a time when ER efficacy and its underlying neurocircuits are rapidly developing (Young et al., 2019), leading to potentially large differences across youth of similar ages, early interventions focused on enhancing ER may reduce and prevent later health discrepancies. Examining the incremental utility of targeting ER strategies in youth is a key direction for future research, as is examining whether potential treatment implications extend to more ethnically diverse samples, older or younger individuals, and individuals with lower socioeconomic status.

This study has several strengths. It longitudinally examined the relation of multiple ER strategies with diurnal cortisol. Assessing the relative contribution of ER strategies is vital given that they are frequently used in tandem (Szasz et al., 2018). Moreover, establishing temporal precedence is particularly important because higher levels of cortisol may make the use of some ER strategies more difficult (Tsumura, Sensaki, & Shimada, 2015), calling the directionality of cross-sectional effects into question. Importantly, findings emerged over and above the effects of significant covariates in the model (i.e., self-reported increases in stress due to COVID-19 and deviations from the saliva sampling protocol), thereby highlighting the robustness of the current results. In addition, this study is the first, to our knowledge, to examine the relation of risk and resilience factors assessed *before* stress, in this case, the COVID-19 pandemic, to biological stress responses *during* stress. This design characteristic is central for understanding how individuals' baseline risk and resilience factors influence functioning during a time of ongoing stress and uncertainty.

Results of this study should also be interpreted in the context of its limitations. Our sample was relatively limited in terms of ethnic, socioeconomic, and geographic diversity due to collecting saliva from adolescents during the early months of the COVID-19 pandemic. Moreover, there were somewhat more male participants in the sample than there were females. Therefore, findings should be replicated in larger, more generalizable samples with more equivalent sex and gender distributions of boys and girls and males and females. Further, although we collected self-reported compliance with our saliva collection protocol (i.e., time at awakening, time at which participants collected each sample, deviations from sampling instructions), and conducted analyses that controlled for deviations, consensus guidelines recommend the use of objective measures of wake and sampling times (Stalder et al., 2016). Thus, the present findings need to be replicated using objective assessments of awakening time, such as actigraphy, and objective verification of sampling times, such as Smart or MEMS track cap containers. It is also important to note that there are various methods for computing daytime cortisol slope, with some researchers computing a wake-to-bedtime slope that excludes any CAR data points (see Adam et al., 2017). Though the same direction of findings emerged when we tested a model based on a wake-to-bedtime slope in the current data, future research is needed to understand the implications of various analytic approaches for assessing daytime slope.

The current study is the first to assess the relative and longer-term prospective influence of ER strategies – including reappraisal, suppression, brooding, and reflection – on diurnal cortisol. It is also the first to examine associations of pre-stressor ER with cortisol during a significant and ubiquitous stressor. Moreover, the present study was conducted with a sample of early adolescents, among whom these processes can have marked ramifications on development. Reflection, suppression, and brooding predicted subsequent diurnal cortisol production. Whereas findings underscore the negative association that baseline risk factors have with biological responses during stress, we also found evidence of resilience. Given that ER is amenable to change, prevention and intervention efforts aimed at improving the selection and effective use of ER strategies can have important benefits. These interventions have strong potential to mitigate short- and long-term secondary effects of stress on the health and wellbeing of youth.

Acknowledgements

This research was supported by a UBC Institute of Mental Health Marshall Postdoctoral Fellowship to KR, a Women's Health Research Institute Fellowship to KR, a Killam Fellowship to KR, a UBC Four Year Doctoral Fellowship Award to EJ, a Vanier Canada Graduate Scholarship to EJ, a UBC 4YF Award to AT, a SSHRC Grant 430-2017-00408 to JL, a CIHR Grant PJT-156053 to JL, an NSERC Grant RGPIN-2018-04837 to JL, and a Michael Smith Foundation for Health Research Scholar Award 17713 to JL.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.biopsycho.2021.108212.

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