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Gender moderates diurnal cortisol in relation to trauma and PTSD symptoms: A study in Sri Lankan adolescents

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ABSTRACT

Trauma exposure and posttraumatic stress disorder (PTSD) have been linked to aspects of diurnal cortisol secretion in adolescents, but little is known about gender differences in these associations. A school-based sample of Sri Lankan adolescents aged 13–16 years took part in this study 4.5 years after the 2004 tsunami had impacted many of their lives to varying degrees. Saliva samples were obtained 4 times a day for 3 days in 84 participants, who also completed measures of lifetime trauma, current stressors, and posttraumatic stress symptoms (PTSS). We used multilevel regression to estimate effects of trauma exposure and symptoms on cortisol level, diurnal slope, and awakening response (CAR). Results indicated higher cortisol in girls and older adolescents. Although trauma, PTSS, and recent PTSD had non-significant main effects, these three variables interacted with gender, with higher cortisol in girls than in similarly traumatized or symptomatic boys. Co-occurrence of internalizing symptoms and PTSS was also associated with higher cortisol. The 28 adolescents with recent PTSD displayed flatter diurnal slopes, reflecting relatively low morning cortisol. Among the 56 trauma-exposed participants, negative trauma appraisals were associated with higher cortisol. Girls were more likely than boys to display elevated cortisol in relation to re-experiencing and hyperarousal symptoms. In contrast to significant findings for cortisol level and diurnal slope, the CAR showed no association with either trauma or PTSS, irrespective of gender. Findings, viewed in light of normative gender differences in HPA activity during adolescence, can contribute to understanding heightened female vulnerability to posttraumatic stress disorder.

1. Introduction

Traumatic experiences have long been linked to dysregulation of the hypothalamic-pituitary-adrenal (HPA) axis, in particular in relation to posttraumatic stress disorder (PTSD) (Yehuda, 2002). Interest in understanding the pathophysiology of PTSD as well as risk factors for developing this or other psychiatric disorders in the aftermath of trauma has driven extensive research. However, studies in adolescents have lagged behind, despite the significance of the transition from childhood to adulthood for the development of psychopathology.

A recent review summarizes current knowledge about childhood adversity, stress hormones, and psychopathology in children and adolescents (Koss and Gunnar, 2018). Although information for adolescents is sparse, there is evidence that traumatic experiences lead to changes in characteristic aspects of salivary cortisol secretion, including cortisol levels over the entire or part of the day, the decrease in secretion from morning to evening (diurnal cortisol slope), and the cortisol awakening response (CAR). Deviant diurnal cortisol patterns have been observed in adolescents who experienced diverse trauma types, including sexual abuse (Keeshin et al., 2014), community violence (Suglia et al., 2010), disasters (Goenjian et al., 1996; Weems, 2015), and various non-intentional traumas (Kuhlman et al., 2015a). Studies also report changes in hair cortisol levels in adolescents traumatized by earthquakes (Gao et al., 2014; Luo et al., 2012) or war (Dajani et al., 2018).

Simply stated, HPA dysregulation can take the form of either hypoactivity and hyperactivity, with low cortisol levels more commonly observed in adults with PTSD, and high levels in children with this disorder (Pervanidou, 2008). The transition from hyper- to hypoactivity may in part reflect longer elapsed time between trauma and cortisol assessment, as found in a meta-analysis of retrospective data (Miller

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et al., 2007). Findings of the few longitudinal studies to date support the notion that post-traumatic HPA hyperactivity can change to hypoactivity over time; in a study of abused girls this transition took several years (Trickett et al., 2010), whereas in Chinese girls exposed to an earthquake, hair cortisol concentrations showed an initial post-trauma increase, which then decreased within months in those who developed PTSD (Luo et al., 2012). Factors such as the specific type(s) of trauma, developmental timing of the exposure, gender, pubertal development, social context, and psychopathology, to name a few, are hypothesized to contribute to divergent findings.

While it has been difficult to untangle effects of trauma and current psychopathology on adult HPA function, both are clearly important (Miller et al., 2007). Although research has focused heavily on PTSD, trauma increases risk of developing a range of psychiatric disorders (Pine and Cohen, 2002). Moreover, comorbidity is common and moderates the effects of trauma on diurnal cortisol patterns in adults. In a community sample, individuals with comorbid lifetime PTSD and major depressive disorder (MDD) had elevated evening cortisol levels, whereas those with trauma exposure alone, or lifetime PTSD without MDD, had normal levels (Young and Breslau, 2004). A similar pattern emerged from a meta-analysis, with comorbid PTSD and MDD being associated with higher afternoon cortisol compared to PTSD alone (Morris et al., 2012). In children and adolescents, symptoms of depression and anxiety are together referred to as internalizing problems; externalizing problems, on the other hand, include aggression, hyperactivity, antisocial behavior, and substance abuse. Extensive research on HPA dysregulation in relation to youth psychopathology has linked internalizing symptoms to increased cortisol levels and heightened stress reactivity, whereas externalizing symptoms are more generally associated with decreased levels and blunted reactivity (Koss and Gunnar, 2018; Marceau et al., 2015). Studies of maltreated children indicate that clinically significant internalizing and externalizing symptoms, occurring together or apart, are associated with distinct diurnal cortisol profiles (Cicchetti and Rogosch, 2001), underscoring the need in trauma research to look beyond PTSD to a broader range of psychopathology. However, to our knowledge, interactions between PTSD symptoms and other emotional and behavioral problems have not yet been investigated with respect to diurnal cortisol profiles in trauma-exposed adolescents.

Whether males and females differ in HPA response to trauma is an important topic, as gender differences could theoretically help explain heightened female vulnerability to PTSD (Olff et al., 2007). Inconsistent gender differences in cortisol secretion have been observed in pre-pubertal maltreated children (Carrion et al., 2002; Doom et al., 2013), and even less appears to be known about adolescents. In one recent study, adolescent refugee girls were more likely than boys to display higher levels of hair cortisol, but this was unrelated to degree of trauma exposure (Dajani et al., 2018). Another study reported that the timing of first trauma exposure was associated with different patterns of diurnal secretion in adolescent girls and boys (Kuhlman et al., 2015b). Such gender differences in diurnal cortisol patterns following trauma must be viewed against the backdrop of normative changes in the HPA axis (Shirtcliff et al., 2012) as well as other biological systems (Garza and Jovanovic, 2017). Recent theories address why gender differences in HPA function and its relationship to stress are likely to emerge during the adolescent transition. Dual-axis investigations, for example, highlight how the HPA axis interacts over the course of pubertal development with the hypothalamic-adrenal-gonadal (HPG) axis, with basal cortisol levels influenced by the androgens dehydroepiandrosterone (DHEA) and testosterone (Marceau et al., 2015). The interplay between HPA and HPG axes, which appears to be moderated by sex and early life stress, may play a role in the development of psychopathology (Koss and Gunnar, 2018; Simmons et al., 2015). The Adaptive Calibration Model (Del Giudice et al., 2011) posits that males and females diverge around puberty in their behavioral and biological responses to environmental stress, with males tending toward hyporesponsivity and females to hyperresponsivity. Although not directly linked to adolescent development, recent reviews have also called attention to possible gender differences in cognitive appraisal, in particular of physically threatening events, in relation to HPA response and heightened risk for PTSD in females (Olff et al., 2007; Pratchett et al., 2010; Zoladz and Diamond, 2013). Negative appraisals are associated with poor post-trauma outcomes and are therefore targeted by effective PTSD interventions (Dalgleish et al., 2005). Appraisals have been linked to cortisol reactivity to experimental stressors (Gaab et al., 2005), but there is little known about effects of real-life trauma appraisals on daily cortisol secretion.

In summary, the reviewed literature reveals several gaps in understanding of how trauma and mental health outcomes are related to HPA dysregulation in adolescence. First, there is no clear consensus about conditions leading to HPA hyper- versus hypoactivity. Here it appears crucial to distinguish trauma exposure from its psychological sequelae, but studies have often lacked either a non-exposed control group or an exposed group without symptoms. Second, prior research has focused heavily on a subset of traumatic experiences, in particular chronic abuse in clinical samples and single incident traumas such as hurricanes or earthquakes, largely neglecting effects of cumulative exposure (but see Bevans et al., 2008). Moreover, with the exception of research on youth exposed to disasters or war, studies in adolescents have been conducted almost exclusively in Western samples. Given widespread exposure to trauma among youth in developing countries (Fairbank and Fairbank, 2009) and its public health impact, a broader global perspective on biopsychosocial sequelae of trauma is warranted (Worthman and Costello, 2009). Third, although ongoing adversity (e.g., current daily stressors, poverty) exacerbates symptoms and can influence cortisol patterns, trauma studies frequently fail to assess it. Finally, as noted above, gender differences have received relatively little research attention. The same applies to symptoms co-occurring with PTSS and cognitive trauma appraisals, which have been hypothesized to have either a general or gender-specific relationship to cortisol patterns.

The current study was accordingly designed to investigate salivary cortisol secretion in relation to trauma and PTSD symptoms in a sample of Sri Lankan adolescents, many of whom exposed in varying degrees to the 2004 tsunami several years earlier, as well as to other lifetime traumatic experiences. For each cortisol outcome of interest (overall diurnal level, diurnal slope, CAR), we used multilevel regression to test relationships with trauma exposure and PTSS, bearing in mind that HPA abnormalities might be reflected in either hypo- or hypersecretion. In light of documented gender differences in vulnerability to PTSD, all models also tested whether gender moderated the association of trauma exposure or PTSS with cortisol measures. Based on the reviewed literature, we hypothesized that trauma and PTSS would be associated with higher cortisol levels in girls compared to boys. Secondary to these goals, we explored possible moderation of cortisol patterns by internalizing and externalizing symptoms, and by trauma appraisals.

2. Methods

2.1. Participants

Galle District, in southern Sri Lanka, was hard-hit by the 2004 tsunami, with approximately 4000 people killed and 35,000 displaced. In January 2008, to investigate the relationship between various types of traumatic experiences and mental health problems in adolescents, we selected two semi-urban study locations with similar socioeconomic profiles but with varying degrees of tsunami impact (Ponnamperuma and Nicolson, 2016). At that time, 755 secondary school students took part in a large, school-based survey.

In July 2009 (T2), we conducted the current study in a subset of the 2008 (T1) sample, for logistical reasons limited to students drawn from 5 of the original 11 schools: 3 in the high-impact and 2 in the low-

impact tsunami locations. Based on their cumulative trauma exposure and psychological symptoms assessed during the initial survey, 286 adolescents were identified as belonging to one of the following three groups: trauma-exposed meeting criteria for full or partial PTSD, in addition to other clinically significant mental health problems and related daily impairment ($n = 63$), trauma-exposed with no clinically significant PTSS, other mental health symptoms, or impairment ($n = 61$), and trauma-free with no significant mental health symptoms ($n = 162$). From each school, 20 adolescents were invited to participate, such that all three groups would be represented in each school and have similar age and gender distributions in the sample as a whole. Exclusion criteria were serious chronic illness or acute illness during the sampling period. Of the 90 enrolled participants, 6 were later excluded: 4 due to incomplete questionnaires and 2 due to missing collection times on the salivettes. The final sample thus comprised 84 adolescents (49 girls), aged 13 to 16 years. Compared to the non-participants ($n = 664$), the T2 subsample at T1 baseline was slightly younger (13.25 vs. 13.69 yr, t (751) = 3.52, $p < 0.001$); the groups did not differ significantly with respect to gender distribution, cumulative trauma, current daily stressors, or PTSS.

Study approval was obtained from the local ethics committee (Faculty of Medicine, University of Ruhuna), the Sri Lankan Ministry of Education, and the school principals. Informed consent was also obtained from the participants and their parents. Upon study completion, participants received a highly valued schoolbook.

2.2. Procedure

The study was conducted in five schools during five consecutive weeks. At each school, study purpose and procedures were explained to students and consent forms for students and their parents/guardians were distributed one week prior to data collection. On a Friday, participants from a given school met as a group with the research team; after all questionnaires were completed, anthropometric measurements were taken. On the same day, participants received detailed instructions about the saliva sampling procedure (see section 2.3.7). They then collected saliva samples at home, from Saturday through Monday. Completed samples were collected early Tuesday morning. The research team consisted of medical doctors who were Sinhalese native speakers.

2.3. Measures

2.3.1. Traumatic events

We used the adolescent version of the UCLA PTSD Reaction Index for DSM-IV (PTSD-RI; Sinhalese translation provided by Dr. F. Neuner) to assess lifetime exposure to 13 categories of traumatic events, described in this instrument as being "very scary, dangerous, or violent things that sometimes happen to people" (Steinberg et al., 2004). We replaced the original category *being in a big earthquake that badly damaged the building you were in* with *being in the tsunami*. A cumulative trauma score was calculated as the number of different event categories endorsed. Participants also indicated which event they considered the worst, and in what month and year it had taken place.

2.3.2. Posttraumatic stress

Participants who reported one or more traumatic events also completed the section of the PTSD-RI that assesses symptoms (PTSS) and diagnostic criteria. The PTSD-RI showed good psychometric properties in previous studies of Sri Lankan adolescents (Neuner et al., 2006). Participants were asked to rate the frequency of 22 symptoms experienced during the past month, with respect to their worst event, on 4-point scales ranging from 0 *never* to 4 *most of the time*. Following established scoring instructions (Steinberg et al., 2004), we generated a PTSD symptom severity score (PTSS) by summing scores on the 20 relevant items (Cronbach's α = .89), as well as scores for DSM-IV-TR symptom clusters B (re-experiencing), C (avoidance), and D (hyperarousal). Adolescents who reported no traumatic events had, by definition, no posttraumatic stress symptoms; their missing scores were therefore recoded to 0. Individuals meeting DSM-IV criteria for all three symptom clusters were categorized as having full PTSD; those meeting criteria for two of the three clusters were categorized as having partial PTSD. Because the same measures had also been collected 16 months earlier, in 2008, participants who had met diagnostic criteria at that time point could also be identified. In the current analyses, the variable *recent PTSD* (0 *no*, 1 *yes*) indicates which adolescents were assessed as having PTSD (full or partial) in 2008 and/or 2009.

2.3.3. Additional questionnaire measures

Negative trauma appraisals were assessed as the sum of ratings on 8 items derived from a 10-item trauma appraisal questionnaire developed for children (Stallard and Smith, 2007). Participants indicated, on 5-point scales ranging from 0 *not at all* to 4 *all the time*, how often during the last 4 weeks they had experienced negative thoughts in relation to their worst event (α = .79). For details, see Ponnamperuma and Nicolson (2016). The eight statements are conceptually similar to "permanent and distressing change" (PC) and "fragile person in scary world" (SW) items on the Child Post-Traumatic Cognitions Inventory (CPTCI) (Meiser-Stedman et al., 2009), published after our data collection and validated in non-Western as well as Western samples (e.g., de Haan et al., 2016; Liu and Chen, 2015; Palosaari et al., 2016).

Internalizing and externalizing symptoms were assessed with the Sinhalese self-report version of the Strengths and Difficulties Questionnaire (SDQ) for adolescents aged 11 to 17 years (Goodman, 2001; YouthinMind). The SDQ has five subscales, each containing five items rated from 0 *not true* to 2 *certainly true*. As recommended for community samples (Goodman et al., 2010), ratings for the ten items in subscales Emotional Problems and Peer Problems were summed as a measure of internalizing symptoms (α = .59), and ratings for the ten items in subscales Conduct Problems and Hyperactivity as a measure of externalizing symptoms (α = .71).

Participants rated perceived family *socioeconomic status (SES)*, relative to other families, on the youth version of the MacArthur Subjective Social Status Scale, validated in adolescents 12 years or older (Goodman et al., 2001). They were asked to imagine a 10-rung ladder as a depiction of how Sri Lankan society is set up: at the top are the people with the most money, highest education, and most respected jobs, and at the bottom are the people who are the worst off, with the least money, little or no schooling, and either no job or one that no else wants or respects.

To assess *current daily stressors*, we used the Long-term Difficulties Questionnaire for Youth (LDQ-Y) (Ponnamperuma and Nicolson, 2018). Adolescents indicated, on 4-point scales (0 *no difficulties* to 3 *serious difficulties*), the extent to which they had experienced difficulties during the last 4 weeks in each of the following areas: school, financial, living situation in the neighborhood, leisure activities, peer relations, family relations, contacts with other people, health, worries about family members, and worries about the future (α = .79).

2.3.4. Physical development

A research assistant weighed and measured each child to determine body mass index (BMI). Girls indicated whether or not they had achieved menarche.

2.3.5. Salivary cortisol

Participants were asked to collect 12 saliva samples: on awakening, 30 min later, in the late afternoon, and before going to bed, on 3 consecutive days. They received polyester swabs (salivettes, Sarstedt, Germany) in a bottle with an electronic cap (MEMS6, Aardex, Switzerland) that registered opening times, and detailed oral and written instructions. They were specifically told to take the first daily sample immediately upon awakening and to avoid tooth brushing and

eating until after both morning samples. Children placed each saturated swab in a capped salivette tube, recorded the actual collection time on both the tube and a separate form, and then stored the tubes at home in a freezer or a cool place. Immediately after the third day, samples were taken to the university and frozen until shipment to the laboratory at TU Dresden, Germany, where cortisol concentrations were determined with a luminescence immunoassay (IBL, Hamburg, Germany). All samples were analyzed in a single assay. Mean intra-assay and inter-assay coefficients of variation at this laboratory were < 12%.

From the 84 participants with sufficient data for inclusion in the analysis, 27 saliva samples were missing or unlabeled, and 28 samples were excluded due to non-compliant collection times (defined as first daily sample outside the time block 4:00 h to 8:00 h ($n = 2$), second sample not within 20–50 min after the first sample ($n = 28$), third sample outside the time block 14:00 h to18:30 h ($n = 2$), or fourth sample outside the time block 19:30 h to 24:00 h ($n = 24$)). Seven samples (0.67% of the total) were excluded due to physiologically implausible cortisol values (> 50 nmol/l) (see R. Miller et al., 2013). A total of 900 valid samples were obtained, with an average of 10.71 samples per person (range 2 to 12; overall 89.3% of the planned samples). The correlation between participant-reported and electronically monitored collection times was highly significant ($r = .97$, $p < .001$). The final dataset for analyses of cortisol level and diurnal slope (first, third, and fourth daily cortisol samples) comprised 685 cortisol measures from 84 participants. CAR analyses were based on data from the 79 participants with valid samples both at awakening and 20–50 min later on one ($n = 7$), two ($n = 10$), or three ($n = 62$) days.

2.4. Statistical analyses

We used SPSS v24 (IBM Corp., Armonk, NY) for descriptive statistics and Stata/MB v13 (Stata Corp., College Station, TX), procedure MIXED with maximum likelihood estimation, for multilevel regression analyses with cortisol as dependent variable. In these models, fixed effects are expressed as unstandardized beta coefficients; β divided by its standard error (*SE*) is approximately *Z*-distributed. Statistical tests are two-tailed, with $p < .05$ considered significant.

The mixed regression models estimating effects on overall cortisol secretion and diurnal slopes had three levels: (1) data were collected three times a day, (2) on three different days, (3) clustered within participants. Prior to analyses, cortisol values were natural-log transformed to help normalize their distribution; positive skew was reduced from 1.235 to -0.414 and kurtosis from 1.460 to -0.887. All continuous independent variables were centered around grand means of the sample or subsample. To control for cortisol's diurnal rhythm, we included the variable time of day (hours since midnight) as a predictor in all analyses, also estimating a random slope. Preliminary analyses also tested, in separate multilevel regression models, fixed effects on cortisol of age, gender, Age x Gender interaction, BMI, menarcheal status in girls, perceived family SES, current daily stressors, internalizing and externalizing symptoms. Of these, only age and gender had significant main effects. In addition to these two variables, final models controlled for Age x Gender interaction, due to its relevance to pubertal development. Models testing effects of PTSS also controlled for trauma exposure and explored possible interactions of PTSS with internalizing or externalizing symptoms.

Separate models estimated the effects of cumulative trauma and PTSD symptom severity (PTSS) on cortisol, over the sample as a whole. Interaction terms were then added to test for gender differences in the relationship between these variables and cortisol levels. Analyses were repeated in the subset of 56 adolescents who reported at least one traumatic event, including additional models to test whether cortisol levels were associated with symptom scores for the three DSM-IV clusters or with negative trauma appraisals, and whether gender moderated such effects.

The association of a given variable with cortisol diurnal slope is reflected in its interaction with time of day. To determine whether trauma exposure and/or PTSS influenced the cortisol diurnal slope, we extended the previous models with interactions between key variables (trauma, PTSS, recent PTSD) and time of day, also estimating 3-way interactions to assess possible moderation of slope predictors by gender.

Finally, we investigated predictors of the cortisol awakening response (CAR). CAR was first calculated as the change in cortisol from the first to the second valid morning sample taken on the same day. The mixed regression model thus had two levels, with one to three CAR measures nested within each of 79 participants (for details, see Section 2.3.5). Because CAR displayed a normal distribution, it was not transformed. In separate models, preliminary analyses showed no significant effects of participants' age, gender, BMI, perceived SES, or current daily stressors; only gender was retained in subsequent models. To test hypothesized gender differences in the CAR, final analyses tested, in addition to the main effects, interactions between cumulative trauma and PTSS with gender. Models with PTSS as predictor again controlled for trauma exposure.

3. Results

3.1. Descriptive statistics

The top half of Table 1 summarizes participant characteristics for the total sample. Boys and girls did not differ significantly in age, BMI, current stressors, trauma exposure, or symptoms, with the exception of more frequent externalizing symptoms in boys. Of the 49 girls, 41 had reached menarche (1 case missing); compared to premenarcheal girls, postmenarcheal girls were not significantly older (means of 14.6y vs. 13.9y, Mann-Whitney U-test, $p = .109$), but had higher BMIs (means of 17.7 vs. 14.1, Mann-Whitney U-test, $p = .001$). Age and BMI were correlated in both sexes: in girls, $r = .49$, $p = .001$; in boys, $r = .44$, $p = .009$. The majority (56.0%, $n = 47$) of participants reported experiencing at least one category of current daily stress, most frequently financial problems (31.0%), followed by health (22.6%) and peer problems (20.2%).

With respect to trauma exposure, 56 of the 84 participants (66.7%) reported having experienced at least one category of traumatic event; 28 participants reported multiple event types, with a maximum of 10 of the 13 categories. As shown in Fig. 1, the December 2004 tsunami was the most frequent trauma type, reported by one third of all participants and representing 22% of total events. Experiencing interpersonal violence (physical or sexual abuse, witnessing domestic violence) was less common than tsunami exposure or the possibly tsunami-related experiences of seeing a dead body or learning of the violent death or injury of a loved one; nevertheless, 16.7% reported its occurrence and 7.1% referred to such violence as their worst event. There were no significant gender differences in exposure to interpersonal violence (Fisher exact tests; $p = .56$ for any exposure, $p = .39$ for worst event). Among the 49 participants who indicated when their worst event had occurred, the interval between event and study enrollment ranged from 1 month to 11 years, with 21 (42.9%) listing an interval of 4 or 5 years, corresponding to the period of maximum tsunami impact. For 4 participants, the worst event preceded the tsunami; for the remaining 24 participants, their worst event had occurred more recently, in the 3 years prior to the study. At the initial screening in 2008, 25 of the current participants met criteria for full or partial PTSD. In 2009, only 7 participants (4 girls and 3 boys), of whom 3 new cases, met criteria for current full or partial disorder. A total of 28 participants (16 girls, 12 boys) thus had clinically significant PTSD symptoms either at present or 16 months earlier.

The bottom half of Table 1 summarizes characteristics of the trauma-exposed subgroup that are relevant for further analyses. As in the total sample, the trend towards greater cumulative trauma in boys was non-significant. Negative trauma appraisal scores were higher in boys than in girls. With the exception of a marginally significant trend

Table 1
Participant characteristics and gender differences.

	Total sample (N = 84)			Boys (n = 35)		Girls (n = 49)		Gender difference	
	Mean	(*SD*)	Range	Mean	(*SD*)	Mean	(*SD*)	*t*	*p*
Age	14.42	0.97	13-16	14.40	1.01	14.43	0.96	0.13	.90
Body mass index	17.07	2.78	12.1-26.4	16.77	2.55	17.29	2.95	0.82	.41
Perceived family SES	5.65	1.25	3-10	5.77	1.45	5.57	1.08	−0.72	.47
Daily stressors	2.26	4.15	0-30	2.20	3.09	2.31	4.80	0.12	.91
Cumulative trauma	1.51	1.84	0-10	1.89	2.34	1.24	1.35	−1.46	.15
PTSS total score[a]	6.42	8.60	0-34	7.66	9.49	5.53	7.89	−1.12	.27
Internalizing	3.89	2.32	1-11	3.43	2.12	4.22	2.42	1.57	.12
Externalizing	4.69	2.58	0-17	5.74	2.83	3.94	2.11	−3.35	.001
	Trauma-exposed (n = 56)			Boys (n = 24)		Girls (n = 32)		Gender difference	
	Mean	(*SD*)	Range	Mean	(*SD*)	Mean	(*SD*)	*t*	*p*
Cumulative trauma	2.27	1.83	1-10	2.75	2.36	1.91	1.23	−1.74	.09
Negative appraisals	3.70	4.05	0-13	5.13	3.94	2.63	3.85	−2.38	.02
PTSS total score	9.63	8.96	0-34	11.17	9.60	8.47	8.41	−1.12	.27
Re-experiencing[b]	3.45	3.50	0-13	3.33	3.38	3.53	3.64	0.21	.84
Avoidance[b]	3.40	3.83	0-14	4.50	4.41	2.56	3.14	−1.92	.06
Hyperarousal[b]	2.79	3.20	0-13	3.33	3.40	2.38	3.03	−1.11	.27

Note.
[a] If no traumatic events, PTSS score = 0.
[b] PTSD symptom cluster scores (DSM-IV).

towards higher avoidance symptoms in boys than in girls, there was no evidence of gender differences in PTSD symptom severity. A linear regression model adjusted for age, gender, and their interaction showed that cumulative trauma exposure was positively associated with PTSS (standardized β = .564, p < .001), with girls displaying more severe symptoms for a given level of reported trauma than boys (Trauma x Gender interaction effect: standardized β = .518, p < .001).

3.2. Cortisol levels

Table 2 summarizes multilevel regression estimates for predictors of overall cortisol levels. The significant time of day effect confirmed the expected diurnal decrease in logcortisol levels. In an initial model, both age (β = 0.182, p = .002) and female gender (β = 0.236, p = .042) were independently associated with higher overall cortisol; after inclusion of an Age x Gender interaction term, only gender remained a significant predictor. Controlling for these variables, separate models tested the effects of cumulative trauma (model *a*), current PTSS (model *b*), and recent PTSD (model *c*). Results indicated no significant main effects of trauma, PTSS, or recent PTSD on overall cortisol levels. However, the addition of interaction terms to the models revealed significant moderation by gender in all three cases. Thus, girls with greater trauma exposure, more severe PTSS, or recent PTSD displayed elevated cortisol levels relative to similarly exposed or symptomatic boys. These elevations were above and beyond the main effect of gender, which remained significant in all models, with higher cortisol in girls. To illustrate how gender moderated trauma effects, we used Stata command marginsplot to graph the association between trauma exposure (categorized into three levels) and untransformed cortisol values over the day, separately for girls and boys. Fig. 2 reveals a pattern of elevated cortisol in more heavily trauma-exposed girls, whereas greater trauma exposure was associated with lower cortisol in boys. A similar pattern (not depicted in the figure) was found for PTSS: in contrast to significantly higher cortisol in girls with many versus few

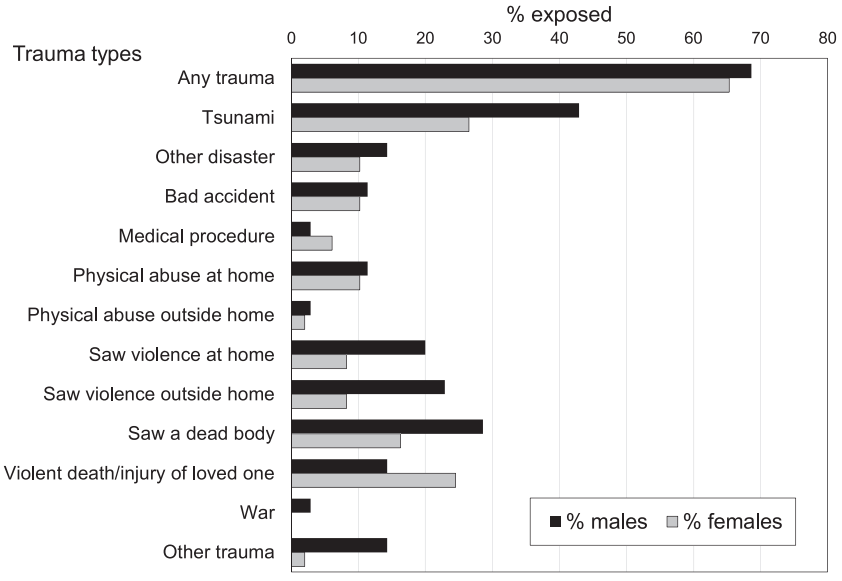

Fig. 1. Percentage of participants (35 males, 49 females) reporting traumatic events, by type of event and gender.

Table 2
Multilevel regression estimates for effects of trauma exposure, PTSS, and recent PTSD on salivary cortisol levels, as moderated by gender.

	ß	*SE*	*Z*	*p*	[95% CI]
Intercept	0.750	.088	8.48	< .001	[0.577, 0.923]
Control variables					
Time of day (hr)	**−0.152**	**.006**	**24.02**	**< .001**	**[-0.164, -0.139]**
Age (yr)	0.092	.090	1.03	.305	[-0.084, 0.269]
Gender	**0.236**	**.115**	**2.04**	**.041**	**[0.009, 0.462]**
Age x Gender	0.159	.120	1.33	.184	[-0.076, 0.394]
(a) Cumulative trauma exposure					
Trauma (main effect)	0.003	.032	0.10	.918	[-0.060, 0.067]
Gender (main effect)	**0.238**	**.120**	**2.02**	**.044**	**[0.007, 0.469]**
Trauma x Gender	**0.161**	**.070**	**2.31**	**.021**	**[0.024, 0.298]**
(b) PTSS					
PTSS (main effect)	−0.002	.013	0.15	.879	[-0.026, 0.023]
Gender (main effect)	**0.238**	**.118**	**2.02**	**.043**	**[0.007, 0.469]**
PTSS x Gender	**0.038**	**.014**	**2.72**	**.007**	**[0.010, 0.065]**
(c) Recent PTSD					
PTSD (main effect)	−0.153	.143	1.07	.285	[-0.434, 0.127]
Gender (main effect)	**0.245**	**.118**	**2.09**	**.037**	**[0.015, 0.476]**
PTSD x Gender	**0.537**	**.255**	**2.10**	**.035**	**[0.036, 1.037]**

Note. Estimates for fixed effects are based on 685 logcortisol measurements nested within 84 participants. Regression coefficients are unstandardized. The model intercept represents logcortisol at the midpoint (14:29 h) of the sampling times, which ranged from 4:00 h to 24:00 h. Because results of preliminary multilevel models separately estimating effects on cortisol of BMI, menarcheal status, perceived SES, and current daily stressors did not approach significance (p values > 0.10), these variables were excluded from final models. Models *a*, *b*, and *c* were estimated separately, controlling for time, age, gender (0 *male*, 1 *female)*, and Age x Gender interaction. To rule out the possibility that estimated effects of PTSS/PTSD simply reflect greater exposure, models *b* and *c* also controlled for cumulative trauma (ß = 0.011, p = .855, and ß = 0.023, p = .538, respectively). % change in raw cortisol per unit change in predictor = [exp(ß) – 1]. Significant results are shown in boldface.

symptoms, boys with more severe PTSS tended to display lower cortisol levels than those with fewer symptoms.

We had hypothesized that internalizing symptoms would exacerbate the effects of trauma exposure and PTSS on cortisol levels. Results were in line with expectations, with no main effect of internalizing (β = 0.010, p = .705) but, in separate models, significant interactions between internalizing and cumulative trauma (β = 0.030, p = .018) and between internalizing and PTSS (β = 0.006, p = .022): cortisol levels were elevated in adolescents with both internalizing and PTSD symptoms. The interactions with gender presented in Table 2 remained significant in these models (Trauma x Gender: β = 0.168, p = .016; PTSS x Gender: β = 0.030, p = .034), indicating that concurrent internalizing symptomatology did not explain higher cortisol observed in girls with greater trauma exposure or more severe PTSS. The Internalizing x PTSS effect remained significant (β = 0.006, p = .031) after controlling for externalizing symptoms. Moreover, the moderating effect of emotional and behavioral problems co-occurring with PTSS appeared to be specific to internalizing symptoms, as models estimating Externalizing x Trauma and Externalizing x PTSS interactions yielded non-significant results (β = -0.014, p = .112, and β = -0.002, p = .245, respectively).

In the trauma-exposed subset of 56 adolescents (447 cortisol measures), results were similar, with no main effect of either cumulative trauma (β = 0.052, p = .216) or PTSS (β = 0.001, p = .934) on cortisol levels, but significant moderation by gender (for trauma: β = 0.277, p = .003; for PTSS: β = 0.044, p = .011): girls with more traumatic events or more severe symptoms had higher cortisol levels than similarly traumatized or symptomatic boys. When both Trauma x Gender and PTSS x Gender terms were included in the same model, only trauma remained significantly moderated by gender (β = 0.383, p = .045), with more heavily exposed girls having higher cortisol levels than boys.

We next investigated, again in the trauma-exposed subgroup, whether certain clusters of PTSD symptoms were specifically associated with cortisol. As shown in Table 3, the general pattern of results was similar to that described in Table 2 and above for total PTSS, with non-significant main effects of PTSD symptom clusters on cortisol levels, but moderation of results by gender. Specifically, more severe re-experiencing symptoms (cluster B) and hyperarousal symptoms (Cluster D) were associated with elevated cortisol levels in girls, compared to boys.

Finally, we tested the hypothesis that negative trauma appraisals contribute to cortisol hypersecretion in exposed adolescents and might possibly help explain how gender moderated effects of PTSS in our previous analyses. Results confirmed that negative cognitions were related to higher cortisol (β = 0.042, p = .038), with an even stronger effect (β = 0.062, p = .020) after controlling for cumulative trauma and PTSS. The predicted Appraisals x Gender interaction effect, however, was non-significant (β = 0.062, p = .122), suggesting that negative trauma appraisals impacted cortisol similarly in boys and girls.

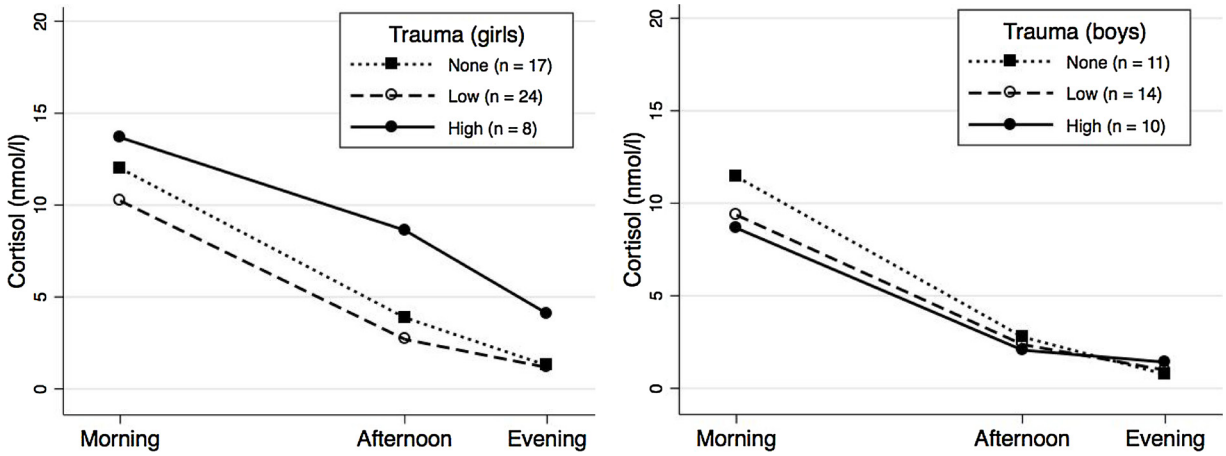

Fig. 2. Diurnal cortisol levels in adolescents with different levels of cumulative trauma exposure, as modelled separately for boys and girls. The figure depicts modelled untransformed cortisol values (nmol/l), as calculated over the three time blocks: morning (4:00 h - 8:00 h), afternoon (14:00 h - 18:30 h), and evening (19:30 h - 24:00 h), in groups defined by level of cumulative trauma exposure: None = no reported traumatic events (n = 28); Low = 1–2 reported event categories (n = 38); High = 3–10 event categories (n = 18). The multilevel regression model controlled for age. See Section 3.2 for statistical results of the analysis testing effects of trauma (continuous variable) and Trauma x Gender interaction on logcortisol levels, controlling for time (continuous covariate), age, gender, and Age x Gender interaction.

Table 3
Multilevel regression estimates of associations between PTSD symptom cluster scores with cortisol level, as moderated by gender, in trauma-exposed adolescents.

	ß	*SE*	*Z*	*p*	[95% CI]
Intercept	**0.606**	**.124**	**4.88**	**< .001**	**[0.363, 0.850]**
Control variables					
Time of day (hr)	**−0.151**	**.009**	**17.55**	**< .001**	**[-0.168, -0.134]**
Age (yr)	0.201	.117	1.72	.086	[-0.028, 0.430]
Gender	**0.323**	**.151**	**2.14**	**.032**	**[0.027, 0.619]**
Age x Gender	0.098	.151	0.65	.516	[-0.198, 0.395]
Cumulative trauma	0.052	.042	1.24	.216	[-0.030, 0.135]
Model Cluster B: Re-experiencing					
Cluster B (main effect)	0.021	.029	0.73	.463	[-0.036, 0.078]
Gender (main effect)	0.300	.154	1.94	.052	[-0.003, 0.602]
Cluster B x Gender	**0.125**	**.044**	**2.87**	**.004**	**[0.040, 0.211]**
Model Cluster C: Avoidance					
Cluster C (main effect)	0.002	.051	0.07	.942	[-0.048, 0.051]
Gender (main effect)	**0.325**	**.153**	**2.12**	**.034**	**[0.025, 0.625]**
Cluster C x Gender	0.067	.042	1.60	.109	[-0.015, 0.149]
Model Cluster D: Hyperarousal					
Cluster D (main effect)	−0.023	.032	0.71	.480	[-0.086, 0.040]
Gender (main effect)	**0.321**	**.150**	**2.14**	**.032**	**[0.027, 0.615]**
Cluster D x Gender	**0.097**	**.048**	**2.04**	**.041**	**[0.004, 0.191]**

Note. Estimates of fixed effects are based on 447 logcortisol measurements nested within 56 participants (24 boys, 32 girls). Regression coefficients are unstandardized. All models controlled for time, age, gender (0 *male*, 1 *female)*, Age x Gender interaction, and cumulative trauma; effects of control variables (top of the table) were estimated before running separate models to test effects of each of the three symptom cluster scores. (% change in raw cortisol per unit change in predictor = [exp(ß) – 1]. Significant results are shown in boldface.

3.3. Cortisol diurnal slope

Over the full sample, results revealed no significant moderation of diurnal slope by either trauma or PTSS (Trauma x Time: β = 0.002, p = .660; PTSS x Time: β = .001, p = .633). A significant PTSD x Time effect (β = 0.029, SE = .013, p = .028) indicated flatter slopes in adolescents with a recent history of clinically significant PTSD; as shown in Fig. 3, this finding reflects lower morning cortisol levels in those with recent PTSD, with no pronounced differences between the two groups later in the day. Diurnal slopes were similar in boys and girls (Gender x Time: β = 0.009, p = .494). Analyses of 3-way interactions showed no moderating effect of gender on slopes in relation to trauma, PTSS, or PTSD.

3.4. Cortisol awakening response (CAR)

The mean collection time for the first saliva sample, taken at awakening, was 5:57 h, with a mean interval of 30.2 min (*SD* 4.34) between the first and the second sample. As expected, cortisol levels increased sharply after awakening, from a mean of 10.5 nmol/l to 13.7 nmol/l (mean CAR = 3.2 nmol/l, *SD* = 6.9); paired t-test, t = 6.74, df = 212, p < .001. A minority (27.7%) of the CARs were flat or negative. Results of preliminary models indicated that blunted CARs were not attributable to either time of awakening (first sample) or interval between the two morning samples: earlier awakening and longer intervals were only marginally associated with a smaller CAR (β = -0.847, p = .215, and β = -0.187, p = .082, respectively). However, as commonly observed (Stalder et al., 2016), CAR was significantly smaller when cortisol at awakening (S1) was higher (β = -.611, p < .001). Subsequent models controlled for these three potential confounders. With respect to demographic variables, CAR was not associated with either age (β = .590, p = .230) or gender (β = 1.838, p = .099), but older girls had larger CARs (Gender x Age: β = 2.671, p = .011).

In separate models, we tested whether either cumulative trauma exposure or PTSS were associated with a deviant cortisol awakening response. By adding interaction terms, we also investigated possible gender differences in these associations. We found no evidence for effects of lifetime trauma (β = -0.110, p = .734) or PTSS (β = 0.009, p = .945) on the CAR, and, in contrast to results for overall cortisol levels, gender was not a significant moderator of either variable (Trauma x Gender: β = 1.227, p = .108; PTSS x Gender: β = 0.221, p = .145).

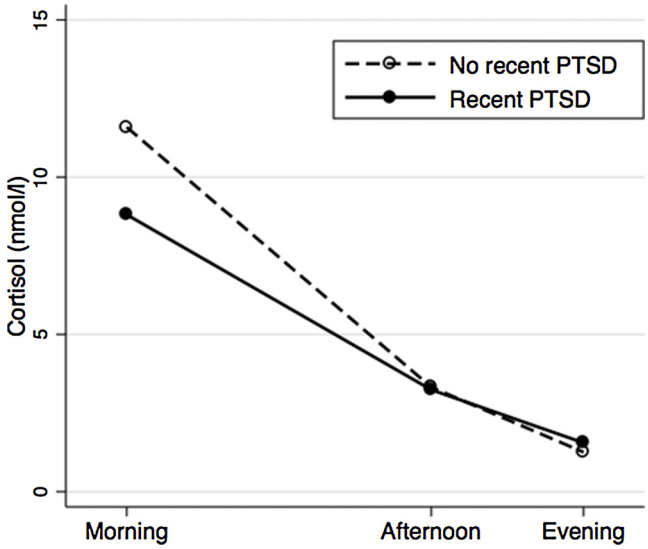

Fig. 3. Diurnal decline in cortisol levels in adolescents with recent PTSD (n = 28) compared to those without recent PTSD (n = 56). As described in Section 2.3.2, participants were categorized as having recent PTSD if they met DSM-IV criteria for full or partial PTSD at present (T2) or at screening 16 months earlier (T1). The figure depicts modelled untransformed cortisol values (nmol/l), by group, as calculated over three time blocks: morning (4:00 h - 8:00 h), afternoon (14:00 h - 18:30 h), and evening (19:30 h - 24:00 h). The multilevel regression model controlled for cumulative trauma, age, gender, and Age x Gender interaction. See section 3.3 for statistical results of the analysis testing effects of PTSD (dummy-coded), time (continuous variable), and PTSD x Time interaction on logcortisol measures.

4. Discussion

The current study's primary goal was to determine whether cumulative trauma exposure or PTSD symptoms in adolescents were associated with either heightened or blunted activity of the HPA axis, as reflected in three indices of daytime salivary cortisol secretion. In addition, we examined gender differences in these associations and - with respect to cortisol levels - possible influences of co-occurring internalizing and externalizing symptoms, PTSD symptom clusters, and trauma appraisals. Contrary to expectation, multilevel regression analyses yielded no evidence for main effects of either trauma or PTSS on cortisol levels, diurnal slope, or CAR. As a partial explanation, we note that trauma exposure in our sample - although widespread and clearly associated with mental health problems and impairment - was not extreme compared, for example, to that of adolescents exposed to war or sexual abuse. Moreover, current symptomatology was generally in the non-clinical range, whereas HPA dysregulation has more often been reported in patient populations or in individuals with diagnosed PTSD. Our finding of flatter diurnal slopes in adolescents who met PTSD criteria in the past 16 months is suggestive in this regard, but without confirmatory clinical diagnosis should be interpreted with caution.

Exploratory analyses provided some new insights into the role of internalizing symptoms. These moderated trauma and PTSS associations with cortisol, whereas externalizing symptoms did not. The observed higher cortisol levels in adolescents with both greater cumulative trauma and more internalizing symptoms is consistent with

findings of a study in maltreated children (Cicchetti and Rogosch, 2001). It is important to note that our school-based sample included few adolescents with clinically significant internalizing or externalizing symptoms, which suggests that findings may be underestimates. As traumatic stress has been linked to the development of a wide range of disorders, studies of cortisol patterns in exposed adolescents should more often consider whether internalizing, externalizing, or other psychological symptoms may be involved. Our findings also underscore the importance of investigating comorbidity. In our case, adolescents with internalizing symptoms in addition to PTSS had higher cortisol levels, as we had predicted based on findings in depressed adults with PTSD (Young and Breslau, 2004). How comorbid symptomatology is related to HPA function appears to be understudied in traumatized adolescents, warranting replication in clinical as well as community settings. Longitudinal studies are essential to determine whether cortisol abnormalities reflect existing symptoms or mediate the development of psychopathology over time.

A striking finding of the current study was the robust influence of gender on daily cortisol output, including more specifically moderation by gender of how trauma exposure, PTSS, and recent PTSD diagnosis were associated with cortisol. Cortisol levels were elevated in more severely traumatized or symptomatic girls, over and above the main effect of gender, whereas boys tended toward HPA hypoactivity. As outlined in the introduction, despite predictions on theoretical grounds, empirical support for gender differences in basal HPA activity in relation to trauma or PTSD is surprisingly limited, for adolescents as well as children. That gender moderated HPA activity in relation to trauma as well as PTSD symptomatology in a community sample of adolescents is thus a novel finding, which can also help explain why cortisol levels were not significantly associated with trauma or psychopathology over the group as a whole.

What might have contributed to the observed gender differences? As girls and boys were similar with respect to overall trauma exposure, PTSS, and recent PTSD, we considered three additional explanations. First, it seemed plausible that relative elevation or blunting of cortisol levels might reflect different patterns of internalizing versus externalizing symptoms: adolescent girls generally display more internalizing and boys more externalizing symptoms - differences that have been posited to reflect divergent life history strategies in dangerous or unpredictable environments (Del Giudice et al., 2011). As discussed above, internalizing symptoms amplified positive associations of trauma and PTSS with cortisol levels. However, girls in the current sample did not display significantly more internalizing symptoms than boys. Moreover, regression results showed that internalizing moderated associations of trauma and PTSS with cortisol levels to a similar extent in boys and girls. Boys did report more externalizing symptoms than girls, but we found no evidence that externalizing was associated with lower cortisol, either in general or in the context of trauma or comorbid PTSS.

Second, we considered possible gender differences in the association between PTSD symptom clusters and cortisol in the trauma-exposed subgroup. Here we observed the same pattern (no significant main effects but relatively greater cortisol elevation in girls) for total PTSS and the individual cluster scores, in particular re-experiencing and hyperarousal. Interestingly, boys and girls had similar scores on these two clusters, whereas gender did not moderate the association between cortisol and avoidance symptoms, the only cluster on which boys tended to score higher than girls.

Third, we explored the hypothesis that gender differences in cognitive interpretations of trauma might influence HPA response. Results confirmed the expected link between negative appraisals and higher cortisol levels, but adjusting for appraisals increased rather than reduced the size and significance of the Trauma x Gender interaction effect. This may reflect the fact that boys in this sample had higher negative appraisal scores than girls.

Although our analyses ruled out a few explanations for gender moderation of cortisol levels in relation to trauma or symptoms, the underlying causes are undoubtedly much more complex. The observation that PTSD symptoms were much more closely associated with cumulative trauma in girls than in boys supports the notion of greater sensitivity to traumatic stress in females. Ideally, we would have also liked to assess cortisol reactivity to acute stressors, to further clarify how hypothesized gender differences in stress reactivity patterns are involved in the pathway from childhood stress to adolescent and adult psychopathology (Chaplin et al., 2018). Findings should also be viewed in the context of normative developmental changes in HPA function. Studies have documented effects of both age and pubertal development on basal salivary cortisol, with increasing levels more consistently reported in girls than in boys; with respect to pubertal stage, there is some evidence for an inflection point in girls, with cortisol levels increasing sharply at Tanner stage III (e.g., Gunnar et al., 2009; Netherton et al., 2004; Shirtcliff et al., 2012). Consistent with this literature, we found evidence of age- and gender-related increases in daily cortisol output, and larger CARs in older girls. Gender interacted with age in predicting cortisol levels, but as we did not assess Tanner stages we could not differentiate effects of age versus pubertal development in either sex. Given the 13–16 y age range, adolescents in our sample were very likely in mid- to late puberty, when sex differences related to pubertal development would already be evident. In a large Sri Lankan sample, median age at menarche was 11.2y, with all girls ≥ 13y in advanced stages of puberty (Tanner stage IV or V) (Wickramasinghe et al., 2009). These data, and the fact that 84% of the girls in our sample had reached menarche, strongly suggest that all were in Tanner stages ≥ III. First remembered ejaculation is considered to be a marker of pubertal onset in boys; in a large European sample mean age at ejacularche was 13.27 y +/- SD 1.08 (Tomova et al., 2011). As noted above, evidence that cortisol changes in relation to pubertal stage in boys is inconsistent, and to our knowledge there is no information available concerning HPA activity in relation to timing of ejacularche. Future studies could, with the addition of a single question, investigate this possibility, although Tanner stages would be more informative. Studies designed to tease apart effects of age and pubertal development, over a wider age range and including earlier pubertal stages in both sexes, remain valuable, as these interrelated variables are likely to have different psychosocial as well as biological correlates that may influence vulnerability to trauma and its mental health sequelae. As an example, effects of early life stress on daily cortisol, in particular the CAR, have been found to vary according to pubertal stage (early versus late) (King et al., 2017). Although beyond the scope of our own study, determination of DHEA and testosterone, in addition to cortisol, in saliva would allow investigation of how HPA and HPG axes interact during adolescence; such information would be especially relevant in studies that focus on the role of gender and childhood stress in the developmental neurobiology of PTSD, depression, antisocial behavior, and other disorders.

Cortisol has been measured in adolescents in diverse cultural, ethnic, and socioeconomic contexts, but the dearth of cross-cultural comparative studies makes it difficult to determine whether specific characteristics of the population we studied might limit generalizability. Our sampling framework (schools in high and low tsunami-impacted areas) meant that many of the participants had been directly affected by the 2004 tsunami; one-third retrospectively reported this natural disaster as a traumatic experience. Moreover, participants in the current study were selectively drawn from a much larger sample surveyed 16 months earlier, based on trauma exposure (yes/no) and PTSD symptoms (high/low) reported at that time. No conclusions can thus be drawn about the frequency of traumatic experiences or the severity of PTSD symptoms in the general population of adolescents living in this area. However, with respect to our main findings, it is important to note that cortisol patterns reflected gender differences in response to cumulative trauma exposure, and not only to the tsunami or any other single category of event. This increases generalizability. More research, particularly in cross-cultural contexts, nevertheless remains essential to

understand how culture - in interaction with gender - affects trauma exposure, appraisal, coping, and psychological outcomes (Norris et al., 2001). Cultural differences may play an important role in shaping responses of the HPA axis and other stress-sensitive systems (Taylor et al., 2007).

The current results should be interpreted in light of some methodological limitations. First, lifetime trauma and psychological symptoms were assessed by self-report questionnaires, whereas interview methods are considered the gold standard for both trauma assessment and psychiatric diagnosis. Some measures we used were not fully validated in our population (LTD-Y; negative appraisals) or were drawn from a screening instrument with too few items to ensure good reliability (SDQ internalizing and externalizing scales). Results of exploratory analyses with these variables should be interpreted with caution and replicated in studies tailored to test specific hypotheses, using multi-informant symptom measures when feasible. Second, timing of exposure was assessed for the worst event only, which limits confidence in the null finding concerning time since trauma and cortisol level. Comparison of results with those of other studies of HPA dysregulation in trauma-exposed youth is difficult, as most concern disasters or accidents that occurred months rather than years ago. Given evidence that recent events may be associated with higher cortisol and more distal events with lower cortisol (Weems and Carrion, 2007), and that the effects of trauma depend on their developmental timing (Schalinski et al., 2016), future studies should use instruments that allow more fine-grained temporal analyses (the MACE is one example; see Teicher and Parigger, 2015). Parental reports can be useful in assessing long-term effects of trauma in the first years of life, especially considering previous findings that developmental timing of trauma was associated with different diurnal cortisol patterns in adolescent boys and girls (Kuhlman et al., 2015b). Third, many participants reported multiple categories of trauma, which made it impossible to tease apart effects of, for example, natural disaster versus interpersonal violence. Whether trauma subtypes have different effects on HPA functioning remains an important question, which can probably only be addressed in a much larger sample or in participants selected on the basis of specific trauma types (e.g., Kuhlman et al., 2015a). Fourth, the study adhered to most but not all current guidelines for measuring the cortisol awakening response (CAR) (Stalder et al., 2016). Thus, samples were obtained on three consecutive days but, to reduce participant burden, at waking (w) and w + 30 min only, omitting the recommended w + 45 min measure. This third sampling point may be important, for example in investigating gender differences or pubertal development, as post-menarcheal girls are reported to have a later CAR peak than pre-menarcheal girls (Oskis et al., 2009). Sample collection times were monitored electronically, but we were unable to verify reported waking times, for example with actigraphy or a forced waking protocol. CAR analyses adjusted for the initial post-waking cortisol value, which reduces but does not eliminate possible inaccuracy due to faulty compliance. Finally, we did not obtain measures of sympathetic activation (for example, salivary alpha-amylase), which would provide a more complete picture of stress system responses to trauma and their dysregulation in PTSD (Gordis et al., 2008; Keeshin et al., 2015).

The current study also had several notable strengths. The school-based sample included trauma-exposed adolescents with negligible to severe PTSS as well as adolescents reporting no traumatic experiences. The three most salient indices of diurnal cortisol secretion (overall levels, diurnal slope, CAR) were obtained, based on multiple saliva samples over three consecutive days. Participants' compliance with the protocol was excellent, as confirmed by electronic monitoring of saliva collection times. Multilevel regression analyses, designed to take missing values and day-to-day variability into account, controlled for likely confounders (e.g., current stress) and several theoretically important covariates.

In conclusion, the observed robust gender differences in associations of trauma, symptoms, and recent PTSD with patterns of daily cortisol secretion support the notion of gender-specific increases in the sensitivity of the HPA system to trauma during mid-adolescence. Apart from gender effects, negative trauma appraisals heightened trauma impact on cortisol levels, whereas internalizing symptoms amplified effects of both trauma exposure and PTSS. These findings point to some directions for future research. Longitudinal studies following traumatized adolescents of both sexes into early adulthood will be necessary to resolve questions about the timing and correlates of the shift from trauma-related HPA hyperactivity to hypoactivity, and whether this process differs in males and females. Prospective studies are also needed to determine whether prior HPA abnormalities mediate the development of post-trauma psychopathology; there is already evidence that this is the case, but the causal pathways are highly complex and very likely transactional (Koss and Gunnar, 2018). Future research must therefore continue efforts not only to clarify how trauma-related patterns of cortisol secretion in adolescence are linked to current symptoms but also, more importantly, to understand how changes in HPA activity heighten risk for mental and physical health outcomes over the lifespan. This knowledge is key to designing effective prevention and treatment strategies.

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Author statement

Both authors made substantial contributions to the work:

NN: study conception and design; funding acquisition; data analysis and interpretation; writing, revising, and approving the final version. TP: study design; data acquisition and management; original draft; reviewing, editing, and approving the final version.

Both authors take responsibility for the integrity and accuracy of all parts of the work.

Conflict of interest

None.

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