

Panel Manipulation in Social Stress Testing:  
The Bath Experimental Stress Test for Children (BEST-C)

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## Summary

*Background:* Whilst acute stress paradigms in adults make use of adult panel members, similar paradigms modified for child participants have not manipulated the panel. Most work has utilised an audience of adult confederates, regardless of the age of the population being tested. The aim of this study was to trial a social stress test for children that provided a meaningful environment using age-matched child peers as panel actors.

*Methods:* Thirty-three participants (7-11 years) underwent the Bath Experimental Stress Test for Children (BEST-C). Based on the Trier Social Stress Test (TSST), it comprises a shortened six-minute public speaking task and four-minute maths challenge. It differs from previous stress tests by using age-matched children on the panel, pre-recorded and presented as a live feed, and includes an expanded manipulation check of subjective experience. Salivary cortisol was assessed at four time points, pre–post stress testing; life events, daily hassles and coping strategies were measured through questionnaires. A simple numerical coding scheme was applied to post-test interview data.

*Results:* The BEST-C generated a typical stress and adaptation response in salivary cortisol ( $p=.032$ ). Age and gender differences were observed during recovery. Cortisol responses mapped directly onto three distinct subjective response patterns: i) expected response and recovery; ii) expected response, no recovery; iii) no response.

*Conclusions:* The BEST-C, utilising child confederates of participant target age is a meaningful social stress test for children. This is the first social stress test developed specifically for children that manipulates panel characteristics by using child confederates and a pre-recorded sham panel. Greater cortisol responses to the test were also found to match subjective verbal accounts of the experience. It offers a meaningful acute stress paradigm with potential applications to other child and adolescent age groups. Furthermore, it leads the way in the use of panel manipulation in social stress testing.

*Keywords:* children, cortisol, social stress testing, panel manipulation, social evaluative threat

## 1. Introduction

Psychological stressors (threats to the social self or self-esteem) are triggered by social evaluative threat (SET), activating the hypothalamic-pituitary-adrenal (HPA) axis and resulting in elevated cortisol release (Dickerson et al., 2009; Dickerson and Kemeny, 2004; Gunnar et al., 2009). Coupled with lack of control over the environment, laboratory social stress tests such as the widely used Trier Social Stress Test (TSST), have consistently elicited a stress response-recovery pattern in adult populations (Kirschbaum et al., 1993). Yet laboratory social stress testing in children has yielded inconsistent findings (Dorn et al., 2003; Gordis et al., 2006).

The traditional TSST presentation task involves public speaking and mental arithmetic in front of a live panel (Kirschbaum et al., 1993). Twenty years since its inception, modifications and adaptations have included a group style TSST-G (von Dawans et al., 2011), a placebo version (Het et al., 2009), and a 'friendly' version (f-TSST) (Abelson et al., 2014; Wiemers et al., 2013), the latter two resulting in a lack of HPA axis activation. Similarly, focusing on helping others rather than on self-promotion lessens cortisol responses (Abelson et al., 2014).

The presence of a live panel is a key element in social stress testing. However, for practical reasons it may not always be feasible. Use of a pre-recorded, 'virtual audience' circumnavigates this. Dickerson et al. (2008) demonstrated that the link between performance and cortisol response is due not to mere social presence but to SET. A further adaptation of the TSST addressed this using a virtual reality (VR) TSST in which participants performed tasks using a head tracking system in front of a virtual environment (Jonsson et al., 2010). Use of virtual reality is gathering momentum in adult stress testing and is suggested as an effective alternative (Montero-Lopez et al., 2015; Wallergard et al., 2011). A pre-recorded audience was applied in the Leiden public speaking task with a panel of adolescents in a classroom setting, eliciting a moderate stress response in adolescents aged 12-15 years (Westenberg et al., 2009).

Adaptations to the TSST have also been made to evaluate stress responses in children. The TSST-C adapted the speech (completing an unfinished story) and maths (serial subtraction in 7's) tasks whilst retaining a panel of two adults who gave positive, rather than negative, feedback (Buske-Kirschbaum et al., 1997). The TSST-M modified the speech task component asking children to imagine introducing themselves to a new class, and reduced the duration of the tasks (Yim et al., 2010) but retained an adult panel. Whilst equivalence of the component tasks has been addressed in child stress testing, an age disparity between participant and panel remains.

Evidence for laboratory based social stress testing in children is inconclusive. Most studies of adolescents (aged 13 plus) show an increase in cortisol in response to a stress test but findings for children below this age have been inconsistent: Buske-Kirschbaum et al. (1997), Gordis et al. (2006) and Yim et al. (2010) all found an increase in cortisol; Dorn et al. (2003) found no significant increase; and Westenberg et al. (2009) only moderate responses. In a review of child stress paradigms, Gunnar et al. (2009) report only 12 out of 17 studies using public speaking tasks to show an increase in cortisol production. Explanations for lack of response have been linked to a hypocortisolaemic period in pre-pubescent children (Hankin et al., 2010). Post infancy until early puberty, children show a diminished basal cortisol level and less reactivity to stress (Gunnar and Donzella, 2002). Whilst this may serve an evolutionarily protective function during brain development (Lupien et al., 2009), there is uncertainty surrounding the extent of this hypocortisolaemic state. The reliability of social stress tests to elicit a cortisol response in children under 13 years of age therefore presents an ongoing debate.

In adult stress testing, participants perform in front of an adult panel yet in child stress testing peers have not been age matched, with the exception of work by Westenberg et al. (2009) with adolescents. Performing in front of an adult panel has the potential to create a power dynamic in children that does not exist in adult testing. Removing this dynamic would make the child and adult stress tests more comparable and ecologically valid since as peer

interaction and evaluation is a key aspect of a child's natural environment (e.g. Gunnar et al., 2003).

There is also evidence that underlying chronic stressors or past stressful experiences may influence reactivity in acute stress situations, for example Marin et al. (2009) report a combined effect of exposure to underlying chronic family stress and acute stress events in children with asthma. The complex nature of stressful events, referred to as "compound stressors" (Michaud et al., 2008) is also evidenced in prior life event stress influencing cortisol response to the acute stress of starting school (Turner-Cobb et al., 2008). Long-term memory of prior stressful events appears implicated in acute stress responses (Montirosso et al., 2013) and early life stress (before age 16 years) can negatively diminish acute laboratory stress responses in young adolescents (Lovallo, 2013). Furthermore, coping can act as a moderator in the TSST (Abelson et al., 2014).

The primary aim of the present study was to create a modified social stress test based on the core characteristics of the TSST/TSST-C but with the panel manipulated to address the age disparity with participants. This modified stress test, the Bath Experimental Stress Test for Children (BEST-C), was designed to create a more meaningful environment for children by enlisting children rather than adults on the panel. The BEST-C utilises a pre-recorded child panel delivered via a sham live video link to participants aged 7-11 years. It also includes a post-test interview to assess subjective stress to compare with the objective stress response assessed via salivary cortisol. We hypothesised that children would exhibit an increase in cortisol in response to the BEST-C, followed by post-test recovery. Those reporting more underlying stressful life events, daily hassles, and use of less effective coping strategies were expected to demonstrate greater cortisol reactivity and slower recovery.

## 2. Method

### 2.1 Participants

An opt-in recruitment method with advertisements in local newspapers, schools, and sports clubs was used to recruit 33 healthy children aged seven to 11 years old (17 boys and

16 girls). Exclusion criteria included oral steroid medication, chronic mental or physical illness or special educational needs (SEN). Demographic information such as age, ethnicity, and socioeconomic status (SES) are given in table 1. The three SES factors include parental occupation level, parental education and a two-factor SES score based on the Hollingshead (1975) weighted SES scoring system. The number of participants was derived from a G\*Power calculation which suggested a sample size of 32, based on conducting a MANOVA with a medium effect size of 0.3 (as indicated by Yim et al., 2010, findings), an alpha of 0.05 and power of 0.80.

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Insert table 1 about here

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## *2.2 Measures*

### *2.2.1 Questionnaires: Demographics, life events, daily hassles and coping strategies*

Demographic details and a life events scale were completed by the child's accompanying parent. The demographics questionnaire requested information concerning parental occupation and qualifications (SES factors), and child age, gender, ethnicity, height, weight and BMI. The Social Readjustment Rating Scale (SRRS) is a list of stressful life events that impact health for adults but which has been adapted for children and adolescents (Holmes and Rahe, 1967). This scale is widely used in the literature and was judged to be a good measure of life events in a review assessing its use over 30 years of research (Scully et al., 2000) and the adult version has test-retest reliability of  $r=.71$  over a six week period (Horowitz et al., 1977). It gathers information about stressful life events that have happened in the last year such as death or divorce of parents, changes in acceptance by peers, and hospitalisation of a sibling. Parents recorded a yes or no answer on behalf of their children for each of the 31 items. Space was provided at the end of the questionnaire for participants to include any additional life events not listed that had occurred in the past year.

Assessment of daily hassles and coping styles were completed by the child participant with assistance in reading and writing from their parent as necessary. The Children's Hassles Scale collected information about hassles that had occurred in the last month (Kanner et al., 1987). The scale has been found to have high levels of predictive validity, with more frequent hassles associated with more emotional distress (Blount et al., 2008) and the adult version of the scale had test-retest averages of  $r=.79$  for frequency of hassles and  $r=.48$  for intensity of hassles (Kanner et al., 1981). The 34 item scale included hassles such as falling out with friends, being punished for doing something wrong, and having trouble finishing homework. Children were asked to rate whether they felt 'ok', 'quite bad' or 'very bad' if the event had happened to them in the past month; the options were accompanied by pictorial representations of these emotions.

The Kidcope questionnaire for children aged 5 to 13 was used to measure children's coping responses to a specific stressor (Spirito et al., 1988). This scale was found to have high test-retest reliability using Pearson's correlation for all questionnaire items when tested three days apart (range = .56 to .75) and six of the ten items when tested one week apart (range = .41 to .83) (Spirito et al., 1988). The child was asked to first 'think of a time when you had a problem that bothered you' and briefly describe this problem. They were then asked three distress questions: whether the problem made them feel nervous or anxious, sad or unhappy, and cross or angry. Children rated these distress items on a five point Likert scale (from 0 to 4) with 0 being 'not at all' and 4 being 'very much'. Participants were shown a list of 15 coping strategies and asked to answer yes or no as to whether they used these coping strategies to help with the problem they had described. If they answered yes they were asked how much that strategy helped on a three point Likert scale (from 0 to 2) with 0 being 'not at all' and 2 being 'a lot'.

### 2.2.2 Stress paradigm: BEST-C

The BEST-C was used as the social stress test. Based on the original TSST (Kirschbaum et al., 1993) and the child related modifications (TSST-M) made by Yim et al. (2010) it involves a ten-minute verbal presentation and mathematical challenge in front of a

panel who elicit SET by failing to provide positive feedback. The BEST-C does not require the audience to be physically present in the same room as the participant. Participants were informed that the audience is being shown via a 'live video link' that is projected onto a large screen with a webcam visible above it. In reality, the live link was a pre-recorded video of the panel that ensured that each participant received identical feedback. The BEST-C uses a child panel of the same target age group as the child participants.

The verbal presentation task involved the child giving a speech about themselves as if they had started a new school and been asked to introduce themselves to their new class; they were told they could talk about their likes and dislikes and that they had to keep talking for six minutes. If necessary the researcher prompted with open-ended questions until the time was up. The maths challenge was a serial subtraction task in which were asked to count down from 825 in multiples of three, lasting four minutes. If participants made an error they had to start again at the beginning.

The BEST-C pre-recorded video was ten minutes long and showed two children dressed in white laboratory coats (one girl and one boy, aged 8 and 10 years) in addition to the adult researcher who was present in the room. The researcher and panel members were dressed in laboratory coats, in keeping with the environment and the TSST stress testing paradigm. Panel members performed the standard roles assigned in TSST tests, operationalised in the BEST-C as one member of the panel staring intently and taking notes while the other panel member played with a mobile phone, looked bored and yawned. The video was projected onto a screen so the panel appeared life-sized and the researcher sat next to the screen to give the impression of a panel of three people. The researcher gave neutral feedback, asked open-ended questions if necessary during the speech task and led the maths task.

### *2.2.3 Salivary cortisol sampling and assays*

Four saliva samples were collected from participants throughout the stress test using a Salivette® (Sarstedt): i) a pre-stressor sample to capture a baseline cortisol level; ii) a sample 20 minutes after the start of the stress test to assess peak response (Dieleman et al., 2010); and



two samples 30 and 45 minutes after the start of the stress test to capture return to baseline (Buske-Kirschbaum et al., 1997). Participants were instructed to refrain from eating or drinking for an hour prior to providing the first saliva sample. Following testing, samples were refrigerated for up to three days before being centrifuged (3000 rpm for three minutes). Samples were then frozen until data collection was complete wherein they were transported on dry ice for commercial testing (Salimetrics, UK). Assays were conducted in duplicate using salivary cortisol enzyme immunoassay (EIA) kits following an established and well-validated protocol with a lower limit of the assay sensitivity of 0.007  $\mu\text{g}/\text{dL}$  and the mean interassay coefficient of variability of 6.3% (determined using duplicate assays of a standard solution of 100  $\mu\text{g}/\text{dL}$  cortisol performed using 10 separate plates). Cortisol levels were converted from  $\mu\text{g}/\text{dL}$  to  $\text{nmol}/\text{l}$  in line with standard reporting values (Jessop and Turner-Cobb, 2008).

#### *2.2.4 Post-test manipulation check: Brief interview*

Following the relaxation period, children were interviewed using a structured interview protocol including questions about how they felt before, during and after the task, what it was like doing the tasks, what they were thinking during the tasks, how they had coped and how they felt about the live feed deception. Questions are given in table 2. The interview acted as an elaborated manipulation check to assess the subjective stress experience of the participant to the social stress test and their use of coping strategies. The interviews were audio recorded, transcribed, and self-reported response type coded quantitatively using a simple numerical scheme. Most participants reported feeling nervous before and during the task and relief once the task was over, indicating the normal pattern of response expected to a social stressor. These participants were coded as group 1 (normative response). Some participants claimed to feel stressed after the task had ended, during the recovery period; these participants were coded as group 2 (continued stress). A small sub-sample of participants did not find the task stressful at all; these were coded as group 0 (no stress response).

Further coding of interview responses using Nvivo and analysis using thematic analysis will be reported elsewhere and only the quantitative analysis for these interviews are presented here.

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Insert table 2 about here

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### *2.3 Procedure*

Testing occurred in the late afternoon to account for diurnal variability in cortisol. Verbal assent from the participant and written consent from the parent were obtained. Parent and child participants completed their respective questionnaires and height, weight, and body mass index data were gathered by the researcher. Approximately 20 minutes after arriving at the laboratory, each participant provided a baseline saliva sample. On completion of the questionnaires, the researcher verbally explained the stress test, described as a ten minute ‘speech and maths task’ in front of an audience and allowed the participant five minutes to prepare some notes and ideas for the speech. Parents were able to assist participants in their preparation for a couple of minutes before being escorted to the waiting room to allow the child to spend the last three minutes preparing by themselves.

During the stress test the child was asked to stand in front of the researcher and the on-screen child panel. When the six minute verbal presentation and four minute maths task were complete participants were escorted to the debrief room to re-join their parent. Twenty minutes after the commencement of the stress test (ten minutes from the end of the stress test) a second saliva sample was taken. Participants and their parent were left to relax for a further ten minutes before a third sample was taken. A final saliva sample was taken fifteen minutes later (45 minutes after the beginning of the stress test).

On completion of all samples, participants and their parent were debriefed and made aware of the deception used during the stress test. Participants were then interviewed.

### *2.4 Statistical analysis*

Cortisol and demographic data were analysed using a Multivariate Analysis of Covariance (MANCOVA) in which age, gender and self-reported stress group were entered as IVs, cortisol during and post-test (time 2 and time 3) entered as DVs and baseline cortisol controlled for as a covariate. Follow-up ANOVAs and t-tests examined group effects. Life events were calculated using the Holmes and Rahe (1967) weighting system, hassles were added up to a total score, and coping was split into two factors assessed by the questionnaires (frequency and efficacy) for the three types of coping (problem-focussed, emotion-focussed, avoidant) (Turner-Cobb and Steptoe, 1998). Relationships between the psychosocial questionnaire data were analysed using bivariate correlations.

Data screening identified one outlier across all four cortisol samples, with this one participant displaying consistently higher cortisol levels than the rest of the sample presenting sufficient justification for exclusion from analysis. The final sample size consisted of 32 participants.

### 3. Results

#### *3.1 Baseline cortisol levels*

It was expected that the first sample taken 20 minutes after the participants arrived at the laboratory would reflect their baseline cortisol level. However, mean cortisol levels were found to be significantly elevated at time 1 compared to post-test levels for each of the five age groups, indicating an anticipation effect of the task. This issue of high cortisol baseline measures was recently listed as one of the main challenges in laboratory-based tasks assessing salivary cortisol reactivity in children (Tolep and Dougherty, 2014). To address this, the fourth sample (taken 45 minutes after the beginning of the stress test) was used as a 'proxy' baseline measure (Nicolson, 2008). Baseline substitution has been carried out in other studies, most recently by Abelson et al. (2014) who found that pre-stressor levels of ACTH and cortisol reflected an anticipation effect and so used the mean of two recovery samples (45 and 60 minutes post-stressor) as their proxy baseline measure. The remaining analyses were

conducted with these three saliva time points rather than four: a proxy baseline measure; stress reactivity; and recovery (figure 1 displays cortisol means across the stress test).

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Insert figure 1 about here

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### 3.2 *Self-reported stress*

As described in section 2.2.4, participant subjective reports of their perceived stress experience before and during the task and in the recovery period were coded into three groups. When these three groupings were mapped onto the cortisol data over the stress testing period the following patterns emerged: i) participants who self-reported in the interview that they did not find the test stressful (group 0) did not show the expected increase in cortisol response to the stress test at time 2 (16% of the sample); ii) participants who reported that they found the task stressful but felt better straight afterwards (group 1) showed the expected stress response and recovery pattern of increase post stress followed by a decrease in cortisol (56% of the sample), and iii) participants who reported feeling stressed in response to the task and that they continued to feel stressed afterwards (group 2) showed an increase in cortisol post-test at time 2 and failed to recover as expected at time 3 (28% of the sample). Figure 2 displays the pattern of responses based on self-reported stress and cortisol data. Differences between these three groups were not significantly different across the three time points when data was analysed using a split-plot ANOVA. There was no main effect of time ( $p = .184$ ) or stress group ( $p = .628$ ) and no interaction between time and group ( $p = .697$ ).

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Insert figure 2 about here

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### 3.3 *Cortisol responses across the BEST-C*

Paired samples t-tests showed a significant increase in cortisol from baseline to time 2,  $t(31) = -2.29$ ,  $p = .029$ , but no significant difference between baseline and time 3 ( $p = .103$ )

or between time 2 and time 3 ( $p = .497$ ). This suggests that the BEST-C effectively elicits an increase in cortisol in this population. There was little difference between cortisol levels at baseline and during recovery, as expected. There was also no significant difference between the reactivity and recovery time points, explained by the age and gender differences in the recovery period (outlined below, section 3.4).

### *3.4 Interaction effects between age and gender*

Using Roy's largest root, there was a significant age \* gender interaction at times 2 and 3,  $\Theta = .1.83$ ,  $F(3, 12) = 7.33$ ,  $p = .005$ , (with a strong effect size of  $\eta^2_p = .647$ ). Separate univariate ANOVAs on the outcome variables revealed a significant age \* gender interaction at time 3,  $F(3, 12) = 7.07$ ,  $p = .005$ , (with a strong effect size of  $\eta^2_p = .639$ ) but not at time 2. A follow-up independent t-test for gender (with the file split by age) found that the only age group that had a significant effect at time 3 was 11 year olds,  $t(5) = 4.73$ ,  $p = .005$ , and that in this age group it was the boys with the higher level of cortisol.

### *3.5 Main effects of age and gender on cortisol reactivity (time 2) and recovery (time 3)*

The MANCOVA and follow-up ANOVAs demonstrated significant main effects for age and gender but not self-reported stress group. Using Pillai's trace, there was a significant effect of baseline at times 2 and 3,  $V = .598$ ,  $F(2, 11) = 8.18$ ,  $p = .007$ , (with a strong effect size of  $\eta^2_p = .598$ ). Separate univariate ANOVAs on the outcome variables revealed a significant effect of baseline at time 2,  $F(1, 12) = 5.86$ ,  $p = .032$ , (with a moderate effect size of  $\eta^2_p = .329$ ) and at time 3,  $F(1, 12) = 15.96$ ,  $p = .002$ , (with a strong effect size of  $\eta^2_p = .571$ ).

Using Roy's largest root, there was a significant effect of age at times 2 and 3,  $\Theta = 1.76$ ,  $F(4, 12) = 5.29$ ,  $p = .011$ , (with a strong effect size of  $\eta^2_p = .638$ ). Separate univariate ANOVAs on the outcome variables revealed a significant effect of age at time 3,  $F(4, 12) = 4.72$ ,  $p = .016$ , (with a strong effect size of  $\eta^2_p = .611$ ) but not at time 2. A series of follow-up independent t-tests were carried out to compare age groups at time 3. The only age-related difference in cortisol levels at time 3 was between 7 and 9 year olds however when a

Bonferroni correction was applied to take into account the multiple t-tests carried out there were no significant differences between age groups.

Using Pillai's trace, there was a significant effect of gender at times 2 and 3,  $V = .719$ ,  $F(2, 11) = 14.06$ ,  $p = .001$ , (with a strong effect size of  $\eta^2_p = .719$ ). Separate univariate ANOVAs on the outcome variables revealed a significant effect of gender at time 3,  $F(1, 12) = 28.73$ ,  $p = .001$ , (with a strong effect size of  $\eta^2_p = .705$ ) but not at time 2. A follow-up independent samples t-test for gender found that boys had higher levels of cortisol than girls at time 3,  $t(30) = 3.08$ ,  $p = .004$ . Gender differences in the stress responses pattern can be seen clearly in figure 3.

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Insert figure 3 about here

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### *3.6 Analysis of questionnaire data: Life events, daily hassles and coping strategies*

Means and standard deviations for the questionnaire data are included in table 1. Bivariate correlations were conducted to examine relationships between life events, daily hassles and frequency and efficacy of the three types of coping strategies at each of the three time points. Life events (coded using the Holmes and Rahe (1967) weightings) were found to be significantly negatively correlated with cortisol levels at time 2 ( $r = -.376$ ,  $p = .034$ ) and time 3 ( $r = -.419$ ,  $p = .017$ ) suggesting that participants with more major life events had lower cortisol levels. Daily hassles were not found to be correlated with cortisol levels at any of the three time points. A one way ANOVA comparing the number of life events and daily hassles in each of the three self-reported stress groups was non-significant for life events ( $p = .345$ ) and daily hassles ( $p = .711$ ).

Correlations examining the frequency of the three coping strategies (problem-focussed, emotion-focussed, and avoidant) and cortisol levels showed a significant negative relationship between frequency of emotion-focussed coping and baseline cortisol ( $r = -.367$ ,  $p$

= .039) and between frequency of emotion-focussed coping and cortisol levels at time 2 ( $r = -.381, p = .031$ ) suggesting that participants who more frequently used emotion-focussed coping strategies had lower cortisol levels at baseline and time 2. There were no significant correlations between the efficacy of the three coping strategies and cortisol levels at any of the three time points. A split-plot ANOVA comparing the frequency of the three coping strategies (within subjects factor) in each of the self-reported stress groups (between subjects factor) showed no significant effects between the three groups ( $p = .399$ ). Similarly, there were no significant differences between the three self-report groups in relation to efficacy of the three coping strategies ( $p = .596$ ).

#### 4. Discussion

##### *4.1 The BEST-C as a meaningful task for inducing a stress response in children*

Findings from the present study show support for the BEST-C as a meaningful social stress test appropriate for use in children aged 7-11 years. Based on traditional stress paradigms, this adapted child stress test includes a panel manipulated to directly address the participant characteristic of age. It uses a pre-recorded panel and also includes a post-test interview of subjective experience. Overall, a significant increase in cortisol was observed across the whole sample twenty minutes after the commencement of the stress test. A decline in cortisol was observed post-testing, however due to age and gender differences during this recovery period the reduction in cortisol was not significant. Intragroup examination revealed three distinct response groupings that mapped directly onto subjective reports surrounding stress testing and post-task adaptation. The BEST-C was shown to elicit both a physiological (cortisol) and psychological (self-reported experience) stress response supporting its application and efficacy for use with child participants.

Results provide convincing evidence in the on-going debate surrounding the ability of social stress tests to reliably elicit cortisol reactivity in children. Support is given to studies that report an increase in cortisol in response to a meaningful stressor in children under the age of 13 years (Buske-Kirschbaum et al., 1997; Gordis et al., 2006; Yim et al., 2010).

Despite childhood being a period of relatively low cortisol compared to the post pubertal period, inability to elicit a cortisol response in previous research may have been due to the appropriateness of the test design rather than solely to the presence of this hypocortisolaemic period. This result suggests that the combined public speaking and cognitive task were successful at inducing a cortisol response due to inclusion of stressor characteristics, uncontrollability and SET (Dickerson and Kemeny, 2004). Features of SET identified in interview were a definite fear of the child panel and reports of not wanting to “perform badly” or “look silly” in front of an audience.

#### *4.2 The impact of life events, daily hassles and coping strategies*

Participants reporting more stressful life events, daily hassles and less effective coping strategies were expected to be slower to recover post task. However, the experience of more stressful life events in the past year showed lower levels of cortisol at post-test (times 2 and 3) and no significant effect of daily hassles on cortisol levels. This suggests that prior life stress may be protective of sustained reactivity to a subsequent acute social stressor. This finding is in line with work reported by Lovallo (2013) in adolescents with prior life event history. Unlike this previous work, we would interpret our finding more positively, as those faced with past stressful acute life events having learnt effective ways of coping, which they were able to draw on during acute social stress.

As predicted, there was a significant relationship between frequency of emotion-focussed coping strategies and cortisol levels at baseline and post stress (time 2). Lower levels of cortisol at these time points were associated with more frequent use of emotion-focussed coping, indicating that its use was protective under acute stress. The theoretical implications of the present study extend to the well accepted transactional model of stress and coping (Lazarus and Folkman, 1984). As nothing can be done to change the stressor during this experimentally confined context the only option available is for the child to regulate their feelings towards the



Whilst emotion-focussed coping is often seen as having less positive outcomes (Compas et al., 2001) these results show the importance of context in defining the adaptability of the response.

#### *4.3 Age and gender differences in the recovery period*

Significant main effects and interactions occurred at time three, the second post-test assessment, 30 minutes after the commencement of the stress test. The sample were relatively homogenous in their reactivity to stress as indicated by cortisol levels at time two (20 minutes after test commencement) but showed differences during the post-test recovery period. Boys revealed higher levels of cortisol than girls during recovery, suggesting that girls adapted more readily and boys continued to experience stress after the task had ended. This pattern could be explained by less frequent use of emotion-focussed strategies in boys during the stress test, as seen in other studies (Connor-Smith et al., 2000).

#### *4.4 Self-reported stress levels mapped onto cortisol data demonstrating three distinct patterns of stress response*

An unexpected, secondary finding in this study was that children's subjective reports of the BEST-C stress experience, as described in post-test interview, matched their cortisol response patterns. Children were able to identify how they felt before, during and after the stress test and this very accurately corresponded to the objective assessment of their stress as assessed by salivary cortisol. It is unusual for self-report to match fluctuations in biological data such as cortisol, particularly in adult samples. Other work with children has found that although self-report of stress levels during the TSST was very accurate, it was much less so pre- and post-stressor (Hellhammer and Schubert (2012). We would suggest that children may have a more intuitive awareness of their feelings of stress, be more honest about their negative feelings and more willing to share those feelings with the researcher than adults may be.

That not all children had the same pattern of response and adaptation is in accord with work by Smyth et al. (1997). Of most concern are not those who responded to stress but failed to adapt post-task or failed to respond to the stressor. Such patterns indicate early

development towards allostatic profiles that may potentially be detrimental to health if subsequently continued and reinforced (McEwen and Stellar, 1993).

#### *4.5 Strengths and limitations*

There are many positive aspects of the present study, such as its novel development of an adapted stress test in which the panel rather than the participant or task was manipulated. The use of a pre-recorded audience proved to be a successful adaptation for children, as in research with adolescents (Westenberg et al., 2009). The current study was conducted in an experimental setting and a mixed methods approach taken that provides a more complete picture of the impact of social stress testing. However, we acknowledge a number of limitations. Firstly, the small sample size. Although a power calculation determined 33 participants to be appropriate, this was minimal and a larger sample would have enabled more confidence in the generalizability of the findings. Issues of protocol relating to the baseline measure provided another limitation. Whilst stress testing protocol recommendations were followed, taking the initial baseline sample 20 minutes after the participant arrived at the laboratory to enable time to adapt to the novel environment and researcher (Gunnar et al., 2009), this was not sufficient to obtain a baseline assessment. Children showed a higher than expected response at 20 minutes after arrival, despite efforts to minimise this novelty effect, and we captured an anticipation period rather than a baseline assessment. Stress anticipation could have been due to a number of factors, including uncertainty or worry about the research nervousness about going to a new place and meeting a stranger, or to events outside of the laboratory including a stressful journey and difficulty finding the laboratory. We addressed this by using the fourth sample taken at 45 minutes after the onset of the stressor as this represented the recovered cortisol state post-testing and hence provided a useful proxy measure of baseline cortisol. We would draw attention to the need for sufficient laboratory time prior to baseline assessment to obtain a true baseline and the particular relevance of this in child stress testing. Some researchers have obtained a baseline away from the laboratory in the child's naturalistic environment a day or more prior to the laboratory stress test (Hostinar et al., 2015; Lovallo et al., 2010). In future work we would

endorse this approach to minimise time required in the laboratory and to obtain a truer baseline uncontaminated by anticipatory stress effects.

#### *4.6 Future research*

The present study has found initial evidence for the BEST-C as an effective tool for inducing a cortisol response in 7-11 year old children. Replication and further validation is needed with a larger sample size. Correlation between lower cortisol levels and higher numbers of stressful life events was unexpected and requires further investigation. Gender differences found in cortisol levels during the reactivity and recovery periods, as well as their relationship with coping strategies, particularly emotion-focussed coping, warrant further attention to tease out the psychosocial factors that could improve children's stress responses. Since not all children responded to the stress test in the same way, future work is called, focusing on individual differences, to explain differential coping with acute social stressors. Further methodological issues also require further testing, such as the impact of having an adult as the panel lead compared to using an older child trained in this role, to enable differentiation of panel versus experimenter effects. Findings could help with the design and implementation of stress-reduction or coping strategy enhancement interventions.

#### *4.7 Conclusions*

This is the first stress test to use child confederates on the panel and the first to use a pre-recorded video as the panel audience in a pre-adolescent child stress test. The present study has confirmed the effectiveness of the BEST-C as a social stressor for children aged 7-11 years. Age and gender differences found in the stress recovery period are findings relating to coping strategies highlight emotion-focussed coping as a useful strategy under acute social stress. The BEST-C also used an innovative mixed-method approach including interview of subjective experience that was coded numerically and analysed in conjunction with the cortisol data, demonstrating that children were accurate in their assessments of their feelings towards stress as their responses matched their biological response to stress. This secondary finding provides convincing support for some of the key stress and coping theories. The BEST-C is the first stress test to use children of the target age group 7-11 years on the stress

panel in conjunction with a pre-recorded video presented to participants as a live feed to ensure consistency of panel response. It offers a meaningful acute stress paradigm with potential applications to other child and adolescent age groups for investigating relationships between stress, coping and health outcomes.

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## References

- Abelson, J.L., Erickson, T.M., Mayer, S.E., Crocker, J., Briggs, H., Lopez-Duran, N.L., Liberzon, I., 2014. Brief cognitive intervention can modulate neuroendocrine stress responses to the Trier Social Stress Test: Buffering effects of a compassionate goal orientation. *Psychoneuroendocrinology* 44, 60-70.
- Blount, R.L., Simons, L.E., Devine, K.A., Jaaniste, T., Cohen, L.L., Chambers, C.T., Hayutin, L.G., 2008. Evidence-based assessment of coping and stress in pediatric psychology. *Journal of pediatric psychology* 33, 1021-1045.
- Buske-Kirschbaum, A., Jobst, S., Wustmans, A., Kirschbaum, C., Rauh, W., Hellhammer, D., 1997. Attenuated free cortisol response to psychosocial stress in children with atopic dermatitis. *Psychosomatic Medicine* 59, 419-426.
- Compas, B.E., Connor-Smith, J.K., Saltzman, H., Thomsen, A.H., Wadsworth, M.E., 2001. Coping with stress during childhood and adolescence: Problems, progress, and potential in theory and research. *Psychological Bulletin* 127, 87-127.
- Connor-Smith, J.K., Compas, B.E., Wadsworth, M.E., Thomsen, A.H., Saltzman, H., 2000. Responses to stress in adolescence: Measurement of coping and involuntary stress responses. *Journal of Consulting and Clinical Psychology* 68, 976-992.
- Dickerson, S.S., Gruenewald, T.L., Kemeny, M.E., 2009. Psychobiological responses to social self threat: Functional or detrimental? *Self and Identity* 8, 270-285.
- Dickerson, S.S., Kemeny, M.E., 2004. Acute stressors and cortisol responses: A theoretical integration and synthesis of laboratory research. *Psychological Bulletin* 130, 355-391.
- Dickerson, S.S., Mycek, P.J., Zaldivar, F., 2008. Negative social evaluation, but not mere social presence, elicits cortisol responses to a laboratory stressor task. *Health Psychology* 27, 116-121.

- Dieleman, G.C., van der Ende, J., Verhulst, F.C., Huizink, A.C., 2010. Perceived and physiological arousal during a stress task: Can they differentiate between anxiety and depression. *Psychoneuroendocrinology* 35, 1223-1234.
- Dorn, L.D., Campo, J.C., Thato, S., Dahl, R.E., Lewin, D., Chandra, R., Di Lorenzo, C., 2003. Psychological comorbidity and stress reactivity in children and adolescents with recurrent abdominal pain and anxiety disorders. *Journal of the American Academy of Child and Adolescent Psychiatry* 42, 66-75.
- Gordis, E.B., Granger, D.A., Susman, E.J., Trickett, P.K., 2006. Asymmetry between salivary cortisol and amylase reactivity to stress: Relation to aggressive behaviour in adolescents. *Psychoneuroendocrinology* 31, 976-987.
- Gunnar, M.R., Donzella, B., 2002. Social regulation of the cortisol levels in early human development. *Psychoneuroendocrinology* 27, 199-220.
- Gunnar, M.R., Sebanc, A.M., Tout, K., Donzella, B., van Dulmen, M.M.H., 2003. Peer rejection, temperament, and cortisol activity in preschoolers. *Developmental Psychobiology* 43, 346-358.
- Gunnar, M.R., Talge, N.M., Herrera, A., 2009. Stressor paradigms in developmental studies: What does and does not work to produce mean increases in salivary cortisol. *Psychoneuroendocrinology* 34, 953-967.
- Hankin, B.L., Badanes, L.S., Abela, J.R.Z., Watamura, S.E., 2010. Hypothalamic-Pituitary-Adrenal Axis Dysregulation in Dysphoric Children and Adolescents: Cortisol Reactivity to Psychosocial Stress from Preschool Through Middle Adolescence. *Biological Psychiatry* 68, 484-490.
- Hellhammer, J., Schubert, M., 2012. The physiological response to Trier Social Stress Test relates to subjective measures of stress during but not before or after the test. *Psychoneuroendocrinology* 37, 119-124.
- Het, S., Rohleder, N., Schoofs, D., Kirschbaum, C., Wolf, O.T., 2009. Neuroendocrine and psychometric evaluation of a placebo version of the 'Trier Social Stress Test'. *Psychoneuroendocrinology* 34, 1075-1086.

- Hollingshead, A.A., 1975. Four-factor index of social status. Yale University, New Haven, CT.
- Holmes, T.H., Rahe, R.H., 1967. The social readjustment rating scale. *Journal of Psychosomatic Research* 11, 213-218.
- Horowitz, M., Schaefer, C., Hiroto, D., Wilner, N., Levin, B., 1977. Life Event Questionnaires for Measuring Presumptive Stress. *Psychosomatic Medicine* 39, 413-431.
- Hostinar, C.E., Johnson, A.E., Gunnar, M.R., 2015. Parent support is less effective in buffering cortisol stress reactivity for adolescents compared to children. *Developmental Sci* 18, 281-297.
- Jonsson, P., Wallergard, M., Osterberg, K., Hansen, A.M., Johansson, G., Karlson, B., 2010. Cardiovascular and cortisol reactivity and habituation to a virtual reality version of the Trier Social Stress Test: A pilot study. *Psychoneuroendocrinology* 35, 1397-1403.
- Kanner, A.D., Coyne, J.C., Schaefer, C., Lazarus, R.S., 1981. Comparison of two modes of stress measurement: daily hassles and uplifts versus major life events. *Journal of behavioral medicine* 4, 1-39.
- Kanner, A.D., Feldman, S.S., Weinberger, D.A., Ford, M.E., 1987. Uplifts, hassles, and adaptational outcomes in early adolescents. *The Journal of Early Adolescence* 7, 371-394.
- Kirschbaum, C., Pirke, K., Hellhammer, D.H., 1993. The 'trier social stress test': A tool for investigating psychobiological stress responses in a laboratory setting. *Neuropsychobiology* 28, 76-81.
- Lazarus, R.S., Folkman, S., 1984. *Stress, appraisal and coping*. Springer, New York.
- Lovallo, W.R., 2013. Early life adversity reduces stress reactivity and enhances impulsive behavior: Implications for health behaviors. *International Journal of Psychophysiology* 90, 8-16.



- Lovallo, W.R., Farag, N.H., Vincent, A.S., 2010. Use of a resting control day in measuring the cortisol response to mental stress: Diurnal patterns, time of day, and gender effects. *Psychoneuroendocrinology* 35, 1253-1258.
- Lupien, S.J., McEwen, B.S., Gunnar, M.R., Heim, C., 2009. Effects of stress throughout the lifespan on the brain, behaviour and cognition. *Nat Rev Neurosci* 10, 434-445.
- Marin, T.J., Chen, E., Munch, J.A., Miller, G.E., 2009. Double exposure to acute stress and chronic family stress is associated with immune changes in children with asthma. *Psychosomatic Medicine* 71, 378-384.
- McEwen, B.S., Stellar, E., 1993. Stress and the individual: Mechanisms leading to disease. *Archives of Internal Medicine* 153, 2093-2101.
- Michaud, K., Matheson, K., Kelly, O., Anisman, H., 2008. Impact of stressors in a natural context on release of cortisol in healthy adult humans: A meta-analysis. *Stress-the International Journal on the Biology of Stress* 11, 177-197.
- Montero-Lopez, E., Santos-Ruiz, A., Garcia-Rios, M.C., Rodriguez-Blazquez, R., Perez-Garcia, M., Peralta-Ramirez, M.I., 2015. A virtual reality approach to the Trier Social Stress Test: Contrasting two distinct protocols. *Behavior research methods*.
- Montiroso, R., Tronick, E., Morandi, F., Ciceri, F., Borgatti, R., 2013. Four-Month-Old Infants' Long-Term Memory for a Stressful Social Event. *Plos One* 8.
- Nicolson, N.A., 2008. Measurement of cortisol, in: Luecken, L.J., Gallo, L.C. (Eds.), *Handbook of physiological research methods in health psychology*. Sage, Los Angeles.
- Scully, J.A., Tosi, H., Banning, K., 2000. Life event checklists: Revisiting the social readjustment rating scale after 30 years. *Educ Psychol Meas* 60, 864-876.
- Smyth, J.M., Ockenfels, M.C., Gorin, A.A., Catley, D., Porter, L.S., Kirschbaum, C., Hellhammer, D.H., Stone, A.A., 1997. Individual differences in the diurnal cycle of cortisol. *Psychoneuroendocrinology* 22, 89-105.
- Spirito, A., Stark, L.J., Williams, C., 1988. Development of a brief coping checklist for use with pediatric populations. *Journal of pediatric psychology* 13, 555-574.

- Tolep, M.R., Dougherty, L.R., 2014. The Conundrum of the Laboratory: Challenges of Assessing Preschool-Age Children's Salivary Cortisol Reactivity. *J Psychopathol Behav* 36, 350-357.
- Turner-Cobb, J.M., Rixon, L., Jessop, D.S., 2008. A prospective study of diurnal cortisol responses to the social experience of school transition in four-year-old children: Anticipation, exposure, and adaptation. *Developmental Psychobiology* 50, 377-389.
- Turner-Cobb, J.M., Steptoe, A., 1998. Psychosocial influences on upper respiratory infectious illness in children. *Journal of Psychosomatic Research* 45, 319-330.
- von Dawans, B., Kirschbaum, C., Heinrichs, M., 2011. The Trier Social Stress Test for Groups (TSST-G): A new research tool for controlled simultaneous social stress exposure in a group format. *Psychoneuroendocrinology* 36, 514-522.
- Wallergard, M., Jonsson, P., Osterberg, K., Johansson, G., Karlson, B., 2011. A Virtual Reality Version of the Trier Social Stress Test: A Pilot Study. *Presence-Teleop Virt* 20, 325-336.
- Westenberg, P.M., Bokhorst, C.L., Miers, A.C., Sumter, S.R., Kallen, V.L., van pelt, J., Blote, A.W., 2009. A prepared speech in front of a pre-recorded audience: Subjective, physiological, and neuroendocrine responses to the Leiden public speaking task. *Biological Psychology* 82, 116-124.
- Wiemers, U.S., Schoofs, D., Wolf, O.T., 2013. A friendly version of the Trier Social Stress Test does not activate the HPA axis in healthy men and women. *Stress-the International Journal on the Biology of Stress* 16, 254-260.
- Yim, I.S., Quas, J.A., Cahill, L., Hayakawa, C.M., 2010. Children's and adults' salivary cortisol responses to an identical psychosocial laboratory stressor. *Psychoneuroendocrinology* 35, 241-248.

Table 1. Means and standard deviations (SD) for demographic information, number of life events, daily hassles and coping strategies (n = 32)

	<b>Boys (n=17)</b>	<b>Girls (n=15)</b>
Age (years)	8.94 (1.48)	9.33 (1.40)
Ethnicity (percent)		
White British	82.0	80.0
White European	18.0	-
White British/American	-	7.0
Prefer not to state	-	13.0
Parent SES		
Occupation level	35.59 (6.82)	38.33 (4.88)
Education level	19.06 (4.49)	18.80 (4.60)
Body Mass Index (BMI)	10.73 (2.84)	12.12 (3.02)
Life events score	124.53 (166.61)	156.47 (111.42)
Everyday hassles score	34.06 (20.64)	32.93 (23.05)
Use of coping style		
Problem-focussed	2.29 (1.36)	2.67 (0.90)
Emotion-focussed	3.53 (1.38)	3.67 (1.63)
Avoidant	1.59 (0.87)	2.07 (1.16)
Efficacy of coping type		
Problem-focussed	7.88 (2.80)	7.53 (1.73)
Emotion-focussed	10.47 (3.45)	9.47 (4.14)
Avoidant	11.88 (2.09)	10.87 (2.53)

Table 2. Questions used in the post-stress test interview

- 
1. What was it like doing the talk?
  2. What was it like when you had to count backwards?
  3. Was there a difference in how you felt about the talk and how you felt about the maths?
  4. How did the talk/counting make you feel?
  5. What were you thinking about during the task? Were any thoughts going through your head?
  6. You managed to complete the task even though it was hard. How did you do that?
  7. How do you feel now that task is over?
  8. How do you feel about us pretending that the other children were watching you?
  9. If another child was doing the study and was worried about it, what kind of things would you tell them?
  10. You can say anything you like about the study (good or bad). Do you have any comments?
-

Figure 1

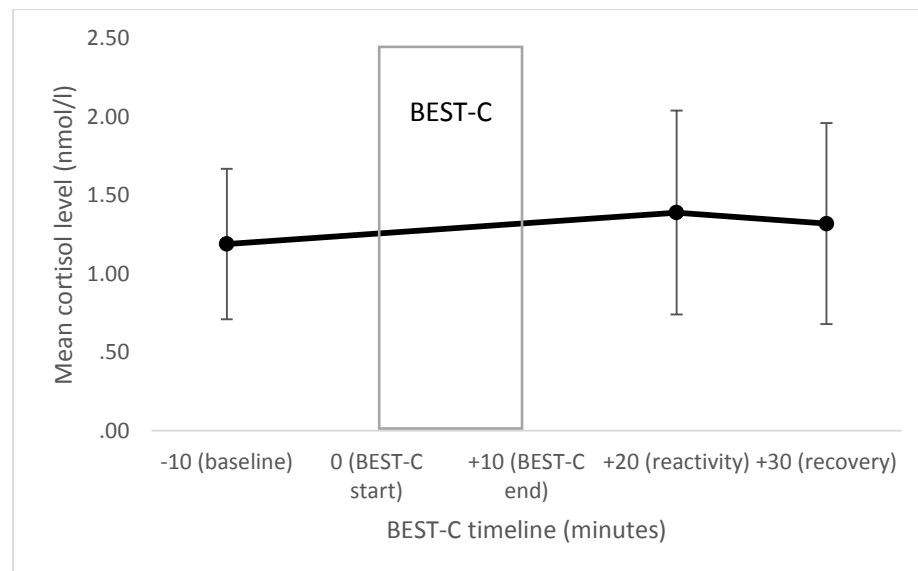
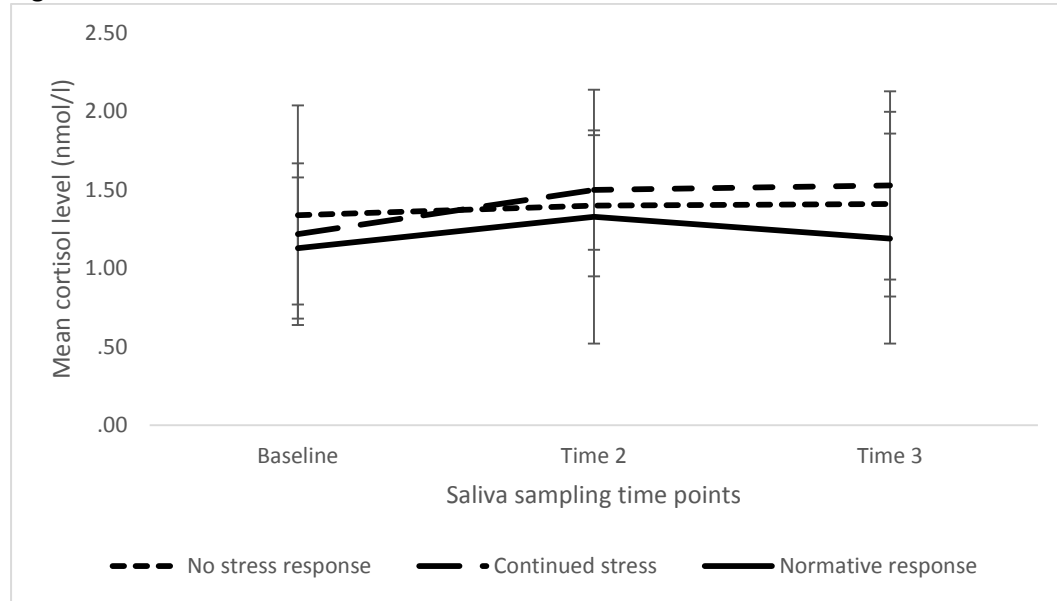


Figure 1. Mean cortisol levels at the three time points (baseline, time 2 and time 3)

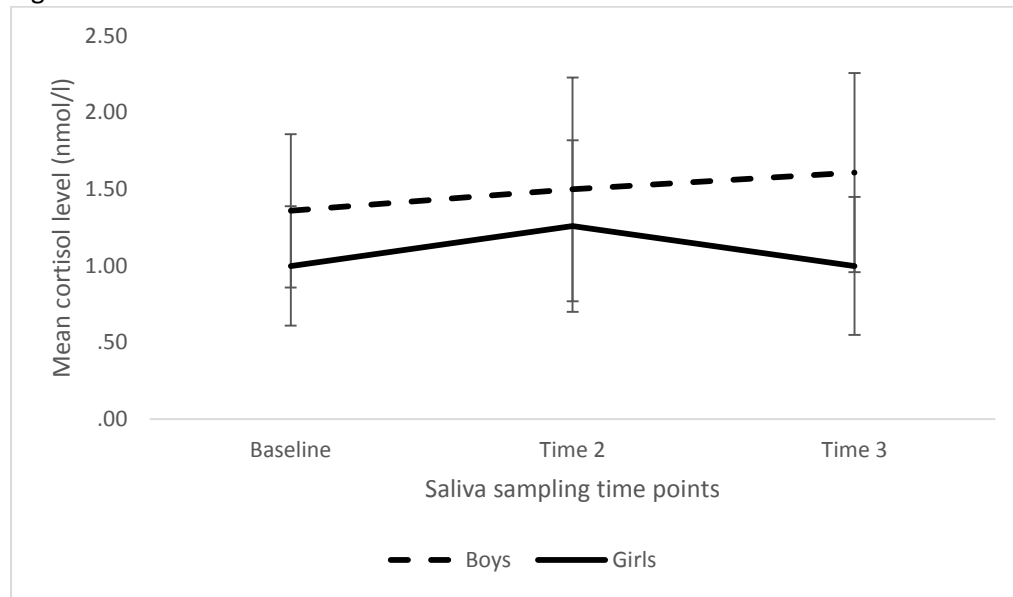
*Note:* The mean cortisol level presented in figure 1 as the baseline sample is in fact the sample taken at time 4 (45 minutes post-stressor) as this better represented the participants' true baseline cortisol levels than the sample taken at time 1.

Figure 2



*Figure 2.* Mean cortisol levels at the three time points for the three different subjective groupings: i) no stress response; ii) continued stress with no recovery; and iii) the normative pattern of stress response followed by recovery.

Figure 3



*Figure 3.* Mean cortisol levels at the three time points for boys and girls. Girls show the normative stress response pattern (reactivity and recovery) and boys show normal reactivity but continued high cortisol in the recovery period.