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Changes and Stability in Daily Cortisol Values and Their Correlation to Attention Measured In a Prolonged Working Task among Finnish Six-Year-Old Day-Care Children

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Abstract

Recently, there has been growing interest in studying stress reactivity in children which is usually accomplished by detecting atypical daily patterns of salivary cortisol. It is assumed that chronic high or low levels of stress hormones are detrimental to attentional performance, a behaviour strongly suggested to be at the core of learning. In the present study, we firstly examined the stability of daily changes in the stress hormone, cortisol. The levels of cortisol were measured five times per day, during one day in autumn and one day in spring. The second aim of our study was to explore whether attention measured in prolonged working tasks was associated with daily cortisol change. Three day care centres in Metropolitan Helsinki, Finland participated in this study and 59 6-year-old preschool children (24 girls, 35 boys) comprised the study sample. The results revealed that the children's daily cortisol variation—in particular, from morning to pre-school and from morning to evening show stability—and this stability was detectable over a six-month period. Children who had lower daily variation in cortisol values in both autumn and spring demonstrated poorer attention. We discuss possible explanations for these findings and implications for early identification.

Keywords: Attention; Cortisol; Preschool children; Stress

Introduction

Attention can be described as the process of concentrating selectively on one aspect of the environment while ignoring other things. Staying focused, or in the other words, being able to concentrate persistently, is crucial for children to process information flow meaningfully, both in informal and formal learning situations. An increasing amount of evidence shows that there are remarkable individual differences among children with regard to how well they can regulate their attention in learning situations before school age [1-5]. From the educational perspective, such differences may contribute to the early onset of both emerging cognitive, emotional, and motivational barriers to learning [6,7].

Regardless of the fact that attention is indeed one of the most rigorously studied topics within educational and neurodevelopmental research, only few studies have focused on the role of the neuroendocrine stress-regulative hypothalamic-pituitary-adrenal (HPA) system in human attention processes. For example, there is no clear evidence on how variations in stress hormones are associated with children's attention in very simple prolonged working tasks before school age, particularly among normative samples.

Neurodevelopmental mechanism of stress-regulation

The human hypothalamic pituary adremedular (HPA) system produces the stress hormone cortisol in response to changes and challenges in environmental stimuli. Activation of the stress system, of which cortisol production is one element, mobilizes energy by putting the metabolism on high alert to meet specific challenges and is fundamental for developing brain functions and learning [8]. This stress response is highly adaptive in the face of short-term stress, but it can turn into a problem in the long-term.

Cortisol levels, key components of the activation of the stress system, are easily and non-invasively assessed from saliva [9,10]. In the absence of acute stress, cortisol follows a circadian rhythm. The highest values are typically seen 30 minutes after awakening (morning peak), followed first by a sharp decline and then by a more gradual decline throughout the day, ending at an evening nadir [11-13]. Cortisol is proven to be a reliable indicator of HPA activity, and cortisol measurements have been widely used in stress studies both in children and adults [14].

Young children have difficulties in shutting down their stress responses independently [15] they need adult support. This means that they are vulnerable to the effects of elevated cortisol levels, which in turn is closely intertwined with their brain maturation. Unregulated stress responses induce chronically elevated cortisol levels, which are known to be toxic to the growing brain and its appropriate functioning [14,16,17]. Elevated or, on the other hand, suppressed amounts of cortisol impact on the integrative and inhibitory areas of the brain, including the hippocampus and prefrontal cortex [17]. These areas are fundamentally important to an individual's attention, learning, memory, and executive functions reference.

Children vary in their capacity for regulating stress responses. There is evidence that elevated or suppressed cortisol levels throughout the day reflect an unbalanced regulation of stress; and atypical diurnal pattern is usually considered a sign of developmental risk [18,21]. Indeed, researchers in the field of early education are becoming increasingly concerned about vulnerable children with hypo- or hyper-reactive stress responses and research related to classroom quality already have shown that higher quality is related to lower stress among both children and teachers [22-26]. Preschool in Finland refers to the year before the onset of school. Children attending to Finnish preschools form a homogenous age group (age 6) to study how attention and cortisol are associated before school-age.

Attention

Regardless of the general opinion that attention is a central element involved in human behaviour and learning, there is no general consensus on how attention should be defined and accordingly measured. According to [27], constant paradigm and task sifting in attention research has produced progress only in our theoretical understanding of attention as a measurable phenomenon.

van der Ven's [28,29] work follows the concentration-test tradition [30] and is based on inhibition theory. This theory is specified in various models which are known as inhibition models. One of these models is the so-called Poisson Inhibition Model and it is described in more detail in Smit and van der Ven [31]. The inhibition theory is formulated based on the basic assumption that during the performance of any intentional mental task the subject actually undergoes a series of alternating states of distraction (non-work) and attention (work). It is assumed that during states of attention, inhibition linearly increases with a certain slope, a1, and during states of distraction inhibition linearly decreases with a certain slope, a0. Accordingly, an assumption can be made that when inhibition increases during a state of attention, depending on the amount of increase, the inclination to switch to a distraction state also increases, and when inhibition decreases during a state of distraction, depending on the amount of decrease, the inclination to switch to an attention state increases. The inclination to switch from one state to the other is mathematically described as the transition rate, which makes the entire process of alternating distraction times and attention times a stochastic process [28].

In prolonged over-learned response tasks, a subject takes control over his or her progress through given test items. Before the test, called the Attention Concentration Test (ACT), the subject practices to both over-learn and adjust his working pace for his or her personal upper limit. The ACT consists of long, simple, and boring subtests to exclude the effect of learning and motivation from the measure. In other words, subjects need to balance speed, accuracy, and mental effort, thus their reaction times can be considered to be a pure measure of attention [30,31]. Based on this model we assume that both the over-learning phase and undergoing several test trials serve to diminish situational stress-induced cortisol in the test situation. Thus the measure could be considered to be an indication of the basic underlying mental function which simultaneously affects all intentional cognitive processes such as working memory, and executive processes. Thus, those children who fail or have difficulties in executing the ACT test (that is, those children who have difficulties in balancing speed, accuracy, and mental effort) are likely to be academically at-risk.The study and working habits, and the neuro-developmental history of these children needs to be carefully studied in order for appropriate interventions to be put in place.

Inhibition theory does not adopt a clear position either for or against the developmental factors of attention. Because attention is derived from the observation of reaction times in over-learned continuous response tasks, it can be assumed that faster cognitive processing and accuracy is acquired as children develop [30,32,33].

Objectives of Study

In the present study, we focus on the interplay of stress responses and prolonged attention to obtain a better understanding of the roots of cumulative cognitive disadvantages. Although there has been growing interest in studying stress and its correlates within various age groups, to the best of our knowledge, there is no research examining stability of changes in daily cortisol values (e.g. differences between the waking and evening values) over a six-month period and among pre-school age (aged six) children, and linking this to attention. For example, increasing academic challenges of the pre-schoolers might have effect on cortisol levels and for this reason we are interested in annual variation in cortisol levels.

We are interested in knowing how the measured attention in November correlates with changes in children's cortisol levels measured both in autumn and spring. OurHPA activation and attention in prolonged working tasks in normative child samples. In normative samples, children do not face severe environmental adversity nor present with significant neuro-developmental risks. However, even in normative samples, children can experience loads that can compromise their stress-sensitive systems. For example, learning and facing new things can become a challenging task for children, either positively or negatively, thereby affecting how they are able to regulate their stress system and how attentive they learn to be in such situations [34]. To the best of our knowledge, there is no research that links children's deviant attentional capacity measured in normative samples and variation in their daily cortisol levels. For this reason, we are interested in examining how children's attention levels (determined by identifying those who are at-risk and those who are not based on their ACT test performance) are associated with daily cortisol values and the changes therein.

Methods

Participants

The study sample included 59 (mean age = 6 y. 4 m., SD = 3 m.) children (24 girls). The day care centres participated in this study on a voluntary basis. All procedures concerning children were conducted with the written understanding and consent of the children's parents or guardians. All but two parents consented to having the children's cortisol measured and they were not included to this sample. The following information used to describe the sample was obtained from two background questionnaires completed by parents and preschool personnel in September 2008. Closer examination of the daily cortisol z-scores reduced the sample size to 58 children (24 girls, 34 boys) as outliers were excluded [35]. The number of children in each day care group varied from 18 to 22. There were no other pre-school groups in these day care centres. The day care centres served predominantly middle-class white families; the average annual family income was categorized as medium or high in 83% of the cases. The majority of mothers (67.2%) and fathers (64.7%) were educated either in vocational high school or at university. "The rest of mothers were educated either in vocational schools (16.8%) or in junior high schools (16%). The rest of fathers were educated either in vocational schools (18.2%) or in junior high schools (17.1%)". Most of the children came from families with two parents (80%) and 15 % of the children came from single-parent families. The remaining 5% did not form unequivocal category (children having two or three families involved in their parenting and children with missing parental information). The average number of children in the each family was 2.2 (range 1–6, median 2). The mean age of the children in the beginning of the study was 6 years and 4 months (range = 5.8–7.0). The children had no major developmental disabilities or chronic illnesses, and they were not taking any regular medication. According to the background information the parents provided, the children slept an average of nine hours per night (M = 9 h, SD 30 min). All the children were enrolled in pre-school programs for a minimum of 30 hours per week. The day care centres participated in this study on a voluntary basis. All procedures were conducted with the written understanding and consent of the children's parents. All but two parents consented to having the children's cortisol measured. Information used to describe the sample was obtained from a background questionnaire completed by parents in September 2008. . This study is a sub-project of the LASSO-research project, run by Dr. Sajaniemi, and was covered by the permission of the ethical committee of human sciences.

Saliva collection procedure

All children followed the same protocol during the days that we acquired saliva samples. No other extra activities or deviations from the daily routines were scheduled during the sampling days. The parents did not report anything unusual in the children's sleep during the nights before we collected the cortisol samples. On the sampling days, all children went to day care within one hour after awakening. At that stage, the children were not napping during the day. If a child was ill on the sampling day, the sampling from that child was delayed until he or she was well again. The children did not take any medication in the sampling day nor on the day before.

The children did not eat or drink for at least 30 minutes before the sampling. The saliva sampling procedure was simple and easy for most of the children. The children mouthed two-inch-thick cotton wads until they were wet. These cotton wads were then placed in Salivette tubes according to written instructions and stored in a refrigerator until they could be sent to the laboratory. In the laboratory, the saliva was separated from the wad of cotton by centrifugation (1000G, 5 min.) and stored at -20ºC until measurement.

The autumn saliva samples were collected during a single day in October 2008. The spring saliva samples were collected during a single day in April 2009. Reliability of the day sample in autumn was .67 and that in spring .65. Samples were taken at 5 times across the day as follows: Awakening Value (06:45 a.m. – 7.15 a.m.), Awakening Effect (half an hour after awakening.), Morning Effect (one hour after awakening.), Pre-school Effect (02:30 p.m.), and Home Effect (Nadir) (08:00 p.m.). In sample days, parents were asked to wake up their children around 07:00 a.m. and bring them to preschool at 08:00 a.m.

Measurements of attention

The ACT [36] primarily measures attention or, more specifically, the attention of concentration (variation of series of reaction times). The test is based on the following three assumptions: 1) knowledge should not play a part in the final test score: only simple problems should be used; 2) differences in previous experience with the task should not be allowed; 3) temporal feelings should not play a part: multiple attempts were allowed to acquire the best results. In this study, we used the easiest parameters of the test. In this version, the subject is required to follow a horizontal line of squares each with different colours (yellow, pink, blue, green, orange, and red). Then, the subject is required to find and click on three red squares situated randomly on that line from left to right, and once successful is to press the "next" button situated below the line.

The actual test consisted of a series of 25 observed consecutive reaction times. For the actual analysis reaction times were corrected for possible test effects. This is because the length of the reaction times is partly dependent on the number of buttons which had to pressed and partly dependent on the serial number of the last button. To correct for this effect for each reaction time series a multiple regression analysis was performed with the number of buttons to be pressed and the serial number of the last button as independent variables and the reaction time as the dependent variable (N = 25). Next, the original reaction times were corrected by taking the residuals of the multiple regression added by the observed mean of the original reaction times.

The test takes approximately seven minutes to complete by children aged 6. Generally, corrected test scores above one standard deviation from the mean are used to identify concentration deficits (van der Ven, personal communication, July 20, 2009). Here we used the easiest "default" parameters and accordingly we lowered the screening level of possible attention deficits to two standard deviations. The ACT data was collected during November 2008. When collecting the ACT data, pre-school teachers were guided to let children play with the game so that they become familiar with the assessment tool to decrease the possible excitement effect when the test is executed. Children did not play this game when saliva samples were collected. The test-retest reliability (two months difference between measurement points) among first graders (n = 20) and fourth graders (n = 20) in normative samples were (Cronbach's alphas) demonstrated in an independent study to be .76 and .82, respectively (Authors, unpublished manuscript).

Analysis procedures

Statistical analyses were performed using the statistical package PASW statistics 21 for Windows. To detect daily variation in cortisol values, four subtractions against the Awakening Effect value were performed, as recommended by Nicolson et al.[35]. Pearson correlation was used to show the correlation between raw cortisol values and computed subtractions. MANOVA was used to analyze if the background variables gender, day care centre, and age had an effect on the results.

To detect if daily cortisol values and their differences (subtractions) are associated with the attention measured in a prolonged working task, a Pearson correlation was used. Due to relative large number of correlational tests, the likelihood of making a Type I error increased. For this reason, only those correlations having $p<.01$ level are taken into account and discussed. Furthermore, when reading the results of the pair-wise comparisons, reader needs to be cautious about results that were significant between $p<.01$ and $p>.001$ levels. To study how grouping based on the ACT scores predicted differences in daily cortisol changes, an independent sample t-test was used. Effect sizes (ES) of these comparisons were calculated by dividing the difference between means of the comparison groups by the weighted standard deviation to yield a standard score (Cohen 1977). The effect sizes of $r = 0.30$ can be interpreted as a small effect, $r = 0.50$ as a medium effect, and $r = 0.80$ as a large effect.

To determine if daily cortisol variation differed according to children's attention level, two study groups were formed: the first group was named 'at-risk children'. This group consisted of those children who failed to pass (n = 4) the ACT and those (n = 7) whose score was at least two standard deviation higher from the mean scores (this rule of thumb was recommended by van der Ven, personal communication, 2009). The second group (n = 47) was named 'normative' and included the remainder of the children (that is those children whose test scores indicated their attention as normative).

Results

Cortisol reactivity

On average, children exhibited a typical diurnal cortisol pattern across the autumn and spring measurements (Table 1). As expected, there was a significant increase in the mean values of salivary cortisol during the first 30 minutes after awakening (Awakening Effect). Corresponding to earlier findings, the pattern of cortisol production decreased during the day, with the lowest values found in the evening (Home Effect) samples. There was no gender or other background variable effect (Parents SES and day care centre) in any measurement point.

However, MANOVA showed that there was a significant day care centre effect at the p<.01 level [Pillal Trace .695; F(20)=2.51, p=.002]. Post hoc comparisons using the Tukey test indicated that the mean cortisol scores for Day Care Centre 1were significantly (p<.01) lower in autumn for the Awakening Effect (M = 10.78, SD = 4.56) than for Day Care Centres 2 (M=16.50, SD=6.70) and 3 (M=15.33, SD = 6.44). Day Care Centre 1 also showed a lower autumn Morning Effect (M=13.47, SD = 5.92) than Day Care Centre 2 (M=20.24, SD=6.92) and 3 (M=17.85, SD=5.61), Furthermore, in spring, children at Day Care Centre 1 had statistically significantly higher (p< .01) Afternoon Effect values than children in Day Care Centres 2 and 3.

	Min	Max	Mean	SD		Min	Max	Mean	SD
Autumn 1	2.71	28.47	14.13	6.33	Spring 1	5.74	56.47	15.33	9.90
Autumn 2	5.27	32.00	17.01	6.72	Spring 2	8.86	36.07	18.96	6.54
Autumn 3	2.17	19.00	9.78	4.70	Spring 3	3.87	21.90	10.83	4.09
Autumn 4	1.24	10.50	4.31	2.43	Spring 4	.55	9.21	4.09	2.08
Autumn 5	.32	5.60	1.59	1.36	Spring 5	-2.81	16.80	2.70	4.22
Sum	22.86	87.96	47.67	15.38	Sum	27.97	120.22	51.53	18.74

Table 1: Descriptives of the diurinal cortisol values in autumn and spring

When comparing mean cortisol scores between autumn and spring, it is noteworthy that all effect values were higher in spring than in autumn, except for the fourth measurement (Pre-school Effect). However, this effect was non-significant according to the paired sample t-test.

Our first study question was aimed at ascertaining if the daily changes (both in autumn and spring) were associated with each other. When comparing raw cortisol values between autumn and spring, we found four statistically significant correlations. Autumn Morning Value was negatively correlated to Spring Pre-school Effect (r=-.46, p<. 001) and Spring Home Effect (r =-.33, p<.05). Accordingly, Autumn Morning Effect value was negatively correlated to Spring Pre-school Effect (r = -.37, p < .01) and Spring Home Effect (r=-.33, p<.01).

In order to study the stability of daily variations, we formed new variables by subtracting every other value from the first measurement (Morning Value) for both autumn and spring samples to firstly detect how much the values differ from the starting point and secondly, to determine if the changes are parallel in both the measurement times, namely in autumn and spring (Table 2). Table 2 shows that the values are close to each other; in particular, the last two calculations in both autumn and spring are parallel. Furthermore, the paired samples t-test showed that there were no statistically significant differences, and there was no gender effect related to these analyses.

	Mean	SD		Mean	SD
Autumn 1-2	-4.51	7.42	Spring 1-2	-2.87	8.20
Autumn 1-3	3.63	6.85	Spring 1-3	4.62	7.79
Autumn 1-4	10.00	6.63	Spring 1-4	10.06	6.67
Autumn 1-5	12.08	6.42	Spring 1-5	12.87	7.22

Table 2: Subtractions from the first measurement in autumn and spring

Next, we conducted correlational analyses to detect associations between the subtracted values. Both in autumn and spring, the correlation between the last two measurements (Morning Effect minus Pre-school Effect and Morning Effect minus Home Effect) were high (autumn r=.95, p<.001 and spring r =.91, p<.001) indicating that these both subtractions gave parallel information about daily variation.

When comparing these subtractions pairwise (e.g., Morning value minus Awaking Effect in autumn with the corresponding subtraction in spring), statistically significant correlations could be found between two changes; Morning value minus Pre-school Effect (p<.01) and Morning value minus Home Effect (p < .001) (see Table 3) indicating a level of stability detectable over a six-month period.

			Spring		
		Spring 1-2	Spring 1-3	Spring 1-4	Spring 1-5
	Autumn 1-2	.16	.27*	.32*	.50***
Autumn	Autumn 1-3	.12	.26	.33*	.51***
	Autumn 1-4	.06	.19	.40**	.55***
	Autumn 1-5	.07	.21	.45**	.50***
Note: * = p < .05, ** = p < .01, *** = p < .001					

Table 3: Correlations between subtractions performed in autumn and spring

It is noteworthy that the MANOVA for the day care and subtracted daily values showed that day care centre had a statistically significant effect [Pillal Trace .615; F(20)=2.06, p =.009]. The post hoc Tukey test showed that children belonging to Day Care Centre 1 had statistically significantly (p < .05) lower subtraction values in autumn, both in the Morning Effect minus Pre-school Effect (M = - 6.72, SD = 4.92) in comparison to children in Day Care Centres 2 (M = -10.69, SD = 7.48) and 3 (M = -12.12, SD = 6.40) . There were also differences in the Morning Effect minus Home Effect (M = -9.29, SD = 4.46) in children from Day Care Centre 1 compared to children in Day Care Centres 2 (M = -14.31, SD = 7.15) and 3 (M = -14.15, SD = 6.56) . In other words, children belonging to this particular day care centre did not show a parallel decline in their cortisol values compared to children attending to the other day care centres.

Attention and daily cortisol changes

To respond to the second study theme, attention measures were examined. The mean value of the ACT test was 0.72, minimum -0.68 and maximum 3.33, and standard deviation was 0.71. The lower values imply higher attention and vice versa. There were four children who did not manage to pass the ACT regardless of multiple trials. There were no gender, age, and parents' SES effects on attention scores. However, ANOVA showed that there was a significant effect of day care centre at the p < .01 level for attention [F(2, 52) = 6.38, p = 0.037]. Post hoc comparisons using the Tukey HSD test indicated that children's mean attention scores from Day Care Centre 1 (M = 0.98, SD = 0.59) were significantly different than scores of children from Day Care Centre 2 (M = 0.33, SD = 0.58). However, attention scores of children from Day Care Centre 3 (M = 0.59, SD = 0.58) did not significantly differ from scores of children from the other two Day Care Centres.

Next, attention values were compared with raw cortisol values. There were two statistically significant (p < .01) correlations. In the autumn samples attention correlated with Pre-school Effect (p < .01, r = .40) and in the spring with Home Effect (p < .01, r = .33). These correlations identify parallel findings: higher attention was associated with the lower cortisol values in both measurements.

Attention and daily changes

When examining if there is a correlation between the differences of daily cortisol values and measured attention, we obtained the following findings: the difference between Morning value and Pre-school Effect is significantly associated with attention scores in both autumn (r = . 35, p < .01) and spring (r = .33, p < .01) the difference between Morning value and Home Effect Effect is significantly associated with attention scores in autumn (r = .37, p < .01) other daily changes (i.e. differences) did not have statistically significant correlations.

An independent sample t-test was used to examine whether the decline in children's cortisol levels from the morning value to the Pre-school Effect and from the morning value to the Evening Effect is different for those children identified as at-risk for attention problems compared to those in the normative group. There was no interaction effect between grouping and day care centre. Table 4 shows that there are statistically significant between-group differences. Those children belonging to the at-risk attention group had a smaller cortisol decline both from the morning to the pre-school measurement and home measurement times. It is noteworthy that standard deviations are large, particularly within the at-risk group, which implies that there is considerable within group differences and that the group includes children whose cortisol decline is parallel to that of the children in the normative group.

	At-risk children (n = 12)		Control group (n = 47)				
	M	SD	M	SD	t-value	p	Cohen's d
Autumn							
Morning – Pre-school Effect	-3.68	9.48	-10.13	7.41	2.47	.017*	0.82
Morning –Home Effect	-7.95	9.35	-12.96	6.62	2.75	.008**	0.69
Spring							
Morning –Pre-school Effect	- 5.16	4.84	-10.49	7.07	2.46	.017*	0.79
Morning –Home Effect	- 7.96	9.35	-12.96	6.62	2.14	.037*	0.69

Note: * = p < .05, ** = p < .01, *** = p < .001

Table 4: Means and standard deviations of the cortisol subtractions from the morning value to the pre-school and home effects for the attention groups

Discussion

In this study the first aim was to examine if the daily cortisol values and their variations show stability over a six-month period among Finnish six-year-old pre-schoolers. The results showed that individual variations in cortisol values did remain stable over this period. Children who displayed minimal daily cortisol fluctuation in autumn showed a similar pattern in spring. This study enhanced earlier findings which have shown that the daily sensitivity of the HPA to acute stress may not be reflected in overall daily variation in cortisol levels in samples who are assumed to experience normative levels of stress [37].

The second aim of the study was to identify if attention measured in a prolonged working task correlates with daily cortisol values and their variation. Our findings show that attention scores correlated statistically significantly to the lower Pre-school effect in autumn, lower Home effect in spring and daily cortisol variation in autumn and spring. The first effect findings indicate that those children whose cortisol level is the lowest either on Pre-school effect time or alternatively on Home effect time have higher attention. It seems that in autumn when pre-school in the Finnish day-care system addresses for the first time in children's life for school-like behaviour the quality of preschool has more effect on causing lower cortisol level [38]. Such association was not present in the spring time, but in turn between attention and Home effect. Nevertheless, in both cases lower cortisol values can interpreted as a better recovery from daily stress which in turn is associated with higher attention. The variation findings provides strong evidence for the association between children's cortisol variation and attentional capacity. It shows that the larger the differences are between both morning and afternoon and morning and evening cortisol values (that is the greater the decline in cortisol across the day), the better are children's attention scores. Thus the skills associated with improved concentration (balancing speed, accuracy and mental effort regardless of the simplicity of the working task) are associated with larger changes in the daily HPA values. This suggests that those children with sufficiently flexible HPA functioning can adjust their inner state and behaviour towards environmental stimuli, remain focused, and accomplish tasks with minimal variation in reaction time. This is supported by our finding of cortisol differences between children belonging to the at-risk attention group and those belonging to the normative attention group [39].

In addition, children belonging to the at-risk attention group had significantly lower autumn and spring Awakening Effect and less daily cortisol variation than the children in the normative group. We propose the children in the at-risk group may experience cumulative difficulties in the learning processes particularly given the connection between limited cortisol variation and learning difficulties revealed in previous studies [40].

Interestingly, the actual centre children attend impacts on their cortisol reactivity and attention. We found the existence of the Pre-school Effect on three cortisol measures in autumn and in one cortisol measure in spring; moreover, there was a day care centre effect on attention. We did not measure the quality of the learning environment in this study, but earlier findings suggest that the pre-school environment is challenging for some children and may have a negative effect on their stress regulation [38,42,43]. In our previous studies, we found that high quality day care was correlated with lower levels of measured cortisol on afternoon, comparable to Pre-school measure in this study. That might indicate that challenges to participate in preschool activities are related to pre-school quality issues [25]. In turn, dysfunction of stress regulation negatively affects attentive abilities, as has been shown in other studies [44]. This relationship extends beyond the early years. For example, in early adolescence, a flat diurnal rhythm is associated with mental health symptoms, which are themselves affected by social context [45]. Given that pre-school emotional support level appears to be associated with a greater decline in cortisol from morning to afternoon, and thus improved attention and the better learning associated with this, interventions targeted to improve day care quality becomes even more essential [23,24].

In Finland, the preschool year is the year before formal schooling begins at the age of seven. The goal of the preschool year is to enhance learning abilities including attention regulation. To prevent the cumulative effects of possible stress-related attention difficulties, the identification of at risk children is of utmost importance. For example, low variation in daily cortisol levels can indicate compromised stress reactivity [46]. The ability to sustain attention depends on activation in the inhibitory prefrontal brain circuits [15,47]. The function of these circuits is sensitized to cortisol variation [17,48]. Atypically high or low cortisol values have harmful effects on brain activation [49]. It might be that those children who were categorized as at-risk children in our study have a tendency towards HPA hypo-reactivity; therefore, they do not have sufficient energy for prolonged, attentive work. Our findings are in line with previous findings which indicate that attention deficit and hypo-arousal of the HPA system are intertwined [50].

Our findings have some practical implications. Firstly, they show that the ACT, used as a screening tool to identify those children who have impaired attention, may identify by proxy, those children who might have atypical cortisol patterns. This, in turn, creates a basis for intervention. When there are clear differences between the pre-school centres in the attention measure, accompanied by atypical cortisol values, it might indicate that concerns with service quality which should be addressed with targeted intervention for both teachers and children. Recently, there has been growing interest in interventions which have attempted to enhance children's skills to balance their behaviour [5]. Indeed, our ongoing intervention project targeted for first (n = 20) and fourth (n = 20) graders, along with equally balanced control groups, examines if the intervention targeted to improve children's awareness of both physical and mental tension and relaxation by passive and active physical exercises (20 × 20 min) can improve their ACT performance (Authors, unpublished manuscript). However, in the project, we do not measure activity of the HPA axis. This current project indicates the need to combine both measurements in our future intervention studies. Secondly, there is a value in using screening tools, such as the ACT, because these provide a more scientifically sound argument for determining whether lack of concentration, maladjustment, and poor school achievement are really due to basic deficiencies in concentration ability or just caused by a poor student-environment-fit. For example, if the measure shows that there are no deficits in students' attention and concentration, the problem is either environmentally or socially constructed and accordingly requires modifications in teaching practices instead of medical treatment, which has become increasingly common. Because attention processing is fundamental for learning, it is essential to precisely measure such ability in children to shape teaching appropriately. Thus far, there is no practical objective measure available for assessing attention capacity [51].

When studies such as ours provide more evidence on the correlation between stress responses and attention, a computer-based method such as the ACT could be used as a screening tool to identify children with deviant attention. However, when planning interventions, assessment of the quality of environment and needed changes should be accomplished along with the small group or individual exercises targeted to both balance children's stress responses and enhance attention. Difficulties in adjusting to learning possibilities involve, besides emotional dysregulation, a mismatch between attention, perception and action. This mismatch is evidenced by a number of studies revealing visuospatial difficulties in children with language impairments, attention deficits, and dysfunctions in motor coordination [52,53].

Although the present study has yielded some preliminary findings, its design is not without flaws. Firstly the study is limited by the small sample size. We followed the recommended sample size (n > 50) [54]. But in selecting a normative sample, we were limited in the number of children who could be identified as at-risk for attentional deficiencies. We believe it is rather unusual that we found 23% children who could be considered to belong to the at-risk group on the basis of their performance in the attention test. We were limited by the fact that there were no clear age-normative rules, thus we used rule of thumb which has worked with school-age children in the past (van der Ven, personal communication, July 20, 2009) to create clear cut-off scores for at-risk children.

The second caveat lies with our cortisol measure. It is recommended that daily cortisol be measured on consecutive days to acquire a more trustworthy indicator for the daily profile [35]. Here, we used only one day in the autumn and one day in the spring, which we acknowledge is far from the ideal assessment. By the time of data gathering our knowledge and resources were limited with this regard. . For this reason, it is recommendable that reader considers this study as a preliminary one and in the future there is need to measure the cortisol on consecutive days rather than only a daily profile to gain more reliable indicators for the cortisol stability measures. In addition, there has been growing interest in studying both daily salivary alpha amylase and cortisol ratio in order to obtain a more comprehensive picture of daily stress systems [55]. We were not able to do so in this study.

The third caveat is that regardless of the fact that there were between-group differences (attentional at-risk and normative groups) in children's daily cortisol decline, there is a large variation in the cortisol scores of each group-particularly within the at-risk group-which suggest that there are children who have a normal cortisol decline in the at-risk group. This suggests that the factors contributing to children's at-risk status may be different for different children. In future, it is of utmost importance to differentiate more precisely those children who have socio-emotional difficulties and those whose difficulties are of neurobiological origin.

Although much remains to be done in the future, our study has generated important findings in the field of early education. We are tentatively able to show the stability of daily cortisol rhythm over a sixth-month period and how variation of that daily cortisol rhythm is linked to attention in normative pre-school aged children. Additional research with larger samples is needed to explore this relationship further. Our research suggests the need for further studies exploring how we can modify environments in a manner that promotes the development of typical cortisol daily rhythm patterns among those children with problems both with attention and with an atypical daily cortisol rhythm indicating unbalanced stress responses. It is becoming more and more clear that quality in early childhood services is not best represented by a one-size-fits-all regulated programme, but by a shared understanding of principles of children's learning and development with the power to be flexible in order to address each individual child's unique neurobiological and contextual profile.

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