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Splitting hair for cortisol? Associations of socio-economic status, ethnicity, hair color, gender and other child characteristics with hair cortisol and cortisone

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The aim of this study was to examine associations of SES and ethnicity with hair cortisol and cortisone and to identify potential child and family characteristics that can assist in choosing covariates and potential confounders for analyses involving hair cortisol and cortisone concentrations. Hair samples were collected in 2484 6-year-old children from the Generation R Study, a prospective cohort in Rotterdam, the Netherlands. Measurements for cortisol and cortisone were used as the outcome in regression analyses. Predictors were SES, ethnicity, hair color and child characteristics such as birthweight, gestational age at birth, BMI, disease, allergy, and medication use. Lower family income, more children to be supported by this income, higher BMI and darker hair color were associated with higher hair cortisol and cortisone levels. Boys also showed higher levels. Ethnicity (Dutch and North European descent) was related to lower levels. High amounts of sun in the month of hair collection was related to higher levels of cortisone only. More recent hair washing was related to lower levels of cortisol and cortisone. Gestational age at birth, birth weight, age, medication use, hair washing frequency, educational level of the mother, marital status of the mother, disease and allergy were not associated with cortisol or cortisone levels. Our results serve as a starting point for choosing covariates and confounders in studies of substantive predictors or outcomes. Gender, BMI, income, the number of persons in a household, ethnicity, hair color and recency of hair washing are strongly suggested to take into account.

1. Introduction

Hair cortisol is increasingly used as a biological marker of chronic stress and is regarded a promising and nonintrusive method of detecting individual differences in long-term reactions to stressful life experiences. Scalp hair is easy to collect, cheap to store and it is relatively straightforward to obtain cortisol and cortisone concentrations. In the current study we examined the influence of SES and ethnicity on hair cortisol levels in 2484 6-year-old children, and we tested hair characteristics as additional covariates or potential confounders. We expect that key socio-demographic risk indicators such as lower SES and ethnic minority status are associated with higher hair cortisol levels, even after taking into account hair color, sun hours in the month of hair sampling, exposure and hair treatment. We refer to these variables as potential confounders, although any of these variables may function as

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either a covariate, confounder or mediator in itself, depending on context.

The corticosteroid hormone cortisol is an important biomarker in studies on stressful life experiences (Fridman et al., 2012; Yehuda et al., 1995). In addition, cortisol plays a pivotal role in the brain with respect to cognition and mental health (De Kloet et al., 2005). The formation of glucocorticoids in the adrenal glands occurs via the enzyme 11-beta-hydroxysteroid dehydrogenase (11B-HSD) type 1 (Best and Walker, 1997; Ferrari and Funder, 2007). Cortisone is in turn metabolized from cortisol in the peripheral tissues by the enzyme 11-beta-hydroxysteroid dehydrogenase type 2. Cortisol has much higher glucocorticoid activity than cortisone, thus, cortisone can be considered an inactive metabolite of cortisol. Disturbances in 11B-HSD levels have been shown to have potentially adverse effects related to the metabolic syndrome (Wake and Walker, 2004), even prenatally (Benediktsson et al., 1997; Seckl, 2004). Reverse metabolism from cortisone to cortisol occurs continuously, but at a slower pace, resulting from lower cortisone activity. Cortisol levels in the human body follow a diurnal pattern (Weitzman et al., 1971), with peak levels in the early morning and the lowest levels three to five hours after sleep onset (Gunnar and Vazquez, 2001; Gunnar and Donzella, 2002; Lupien et al., 2009).

In response to both short and long term stressors (either psychological or physical), cortisol levels become elevated (Miller et al., 2007; Sauvé et al., 2007), even in neonates (Yamada et al., 2007). Cortisol levels seem to be gender-specific (Halbreich et al., 1984; Kirschbaum et al., 1992, 1995), with males having higher cortisol reactivity to daily stressors. The awakening response seems however to be more prolonged in females than in males (Van Cauter et al., 1996; Therrien et al., 2007; Fries et al., 2009). This holds for non-human primates as well (Laudenslager et al., 2012). Stewart et al. (1999) showed a relation between elevated cortisol levels and increased body fat as measured by the Body Mass Index (BMI), which is tissue-specific (Rask et al., 2001) and holds in the general population (Fraser et al., 1999). Several other factors have been shown to be correlates of cortisol levels: biological factors, psychosocial factors (including maternal depression, childhood adversity, novelty seeking behavior (Laudenslager et al., 2011)), and demographic factors such as socio-economic status (SES) (Mackenbach, 1992) and ethnicity (Cohen et al., 2006). Among the biological factors, higher cortisol levels have been related to sleep deprivation (Leproult et al., 1997) and augmented caffeine use (Lovallo et al., 2006). Exercise (both aerobic and anaerobic) has been found to be related to increased levels of cortisol (Kindermann et al., 1982; Skoluda et al., 2012). Long-term effects of (mild) adversities more specifically related to SES on cortisol secretion have been observed as well (e.g., Bosch et al., 2012; Lupien et al., 2009; Halldorsson et al., 2000; Van Hooijdonk et al., 2008; Jansen et al., 2009). Ethnic minority status is often related to lower SES and thus to higher cortisol levels but may also imply difficulties to capture commensurable corticosteroid concentration due to differences in pigmentation that may affect hair cortisol levels (Bennett and Hayssen, 2010).

Hair cortisol and cortisone are increasingly used as a biological marker of heightened physiological activity or chronic stress (Vanaelst et al., 2013; Staufenbiel et al., 2013), and, besides clinical metabolic information, hair sampling is regarded a promising and nonintrusive method of detecting individual differences in children's long-term reactions to stressful life experiences (Groeneveld et al., 2013). The conventional measures of cortisol extraction from saliva or blood (serum) reflect cortisol levels at the time of sampling, potentially influenced by daily fluctuations. These fluctuations can be addressed by repeated sampling, but this is rather invasive, especially in young participants. Urinary measurements of cortisol represent cortisol secretion of the past 24 h–48 h. Storage of such samples is subject to very specific (and expensive) demands. In contrast, concentrations of cortisol and cortisone extracted from hair samples may reflect accumulated concentrations of cortisol levels up to several months (Braig et al., 2015; Manenschijn et al., 2011a,b, 2012; Noppe et al., 2014). Acquisition of hair samples is non-invasive and samples are both easy and cheap to store.

Similar association patterns as in general cortisol have been found with hair cortisol in adults and children (Noppe et al., 2014; Vanaelst et al., 2012). Similar to cortisol in general, hair cortisol levels are significantly higher in men than in women (Russell et al., 2012; Staufenbiel et al., 2015). Seasonal effects have also been shown in Staufenbiel et al. (2015) and Braig et al. (2015), with higher hair cortisol levels in summer and autumn compared to winter. Wosu et al. (2015) showed that blacks and Hispanics have higher hair cortisol concentrations compared to whites and other ethnicities. However, as discussed above, in small samples it might be difficult to differentiate between associations of ethnic variability with cortisol from associations of SES with cortisol (Lupien et al., 2000, 2001, 2009; Vaghri et al., 2013; DeSantis et al., 2007). Several of the factors related to cortisol in blood or saliva were also examined in relation to hair cortisol levels, in different samples and under different conditions. Dettenborn et al. (2012) studied relations to hair cortisol in a relatively small sample ($n = 360$) aged between 1 and 91 years, using an earlier method of cortisol extraction. However, family characteristics were not taken into account. Braig et al. (2015) examined hair cortisol concentrations during delivery and showed that cortisol concentrations reflected more adverse circumstances in the last trimester in pregnancy. Further associations of demographic characteristics with hair cortisol and cortisone concentrations in adults were described by Staufenbiel et al. (2015).

The aim of this paper is to explore the influence of socio-economic and other demographic variables on hair cortisol levels in a large, multi-ethnic sample of homogeneous age. Furthermore, the influence of various characteristics of hair such as hair color, use of hair products, hair washing, and amount of sun light (in hours) in the month of visit on hair cortisol levels will be examined, in order to present a set of potential confounders to be taken into account in substantive studies on hair cortisol and cortisone. In the current study we examined the influence of SES and other family characteristics, as well as individual characteristics and ethnicity on hair cortisol levels in 2484 6-year-old children, and we tested hair characteristics as potential confounders. Our hypothesis is that lower SES and ethnic minority status are associated with higher hair cortisol levels, even after taking into account potential confounders including hair color, sun hours in the month of hair sampling, and hair treatment.

2. Methods

2.1. Study sample

The current investigation was embedded in the Generation R Study, a prospective cohort investigating development from fetal life into young adulthood in Rotterdam, the Netherlands (Jaddoe et al., 2012; Tiemeier et al., 2012; Kruithof et al., 2014). Written informed consent was obtained from parents of all participants. The study has been approved by the Medical Ethical Committee of the Erasmus MC, Rotterdam.

The selected sample ($n = 2484$) was a mixed-ethnicity subsample of the full population-based cohort, for which measures of hair cortisol and cortisone were available. A comparison of characteristics in the full cohort and the selected subsample, and for males and females within the selected sample, is shown in Table 1.

In total 6690 children visited the Generation R research center at age 6. Hair samples for cortisol assessment were collected during