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Determinants of hair cortisol and hair cortisone concentrations in adults

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Summary
Background: The analysis of hair cortisol concentrations (HairF) is a promising new tool for the assessment of long-term cortisol. With the development of multiple steroid analyses by means of liquid chromatography tandem-mass spectrometry (LC—MS/MS), the analysis of cortisone in hair (HairE) has also been facilitated. However, the influence of various types of determinants on HairF and HairE is still largely unknown. This study systematically assesses the influence of sociodemographic, health, lifestyle, and hair (treatment) characteristics on HairF and HairE.
Method: Data of 760 psychiatrically healthy participants (71.8% female, mean age 45.89 years) of the Netherlands Study of Depression and Anxiety (NESDA) were used. HairF and HairE were measured in the proximal 3 cm of scalp hair, using LC—MS/MS.
Findings: HairF and HairE strongly correlated. In simple linear regressions, HairF and HairE were higher in older age, in presence of diabetes mellitus, and in men compared to women. More frequent washing of the hair was associated with lower HairF and HairE. Darker hair colours were associated with higher HairF and HairE. An effect of season and of use of oral contraceptives was found for HairF. After full mutual adjustment, only age, presence of diabetes mellitus, hair washing frequency, and season remained significant determinants of HairF.

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Interpretation: This large-scale study shows that HairF and HairE are upregulated in older age and in the presence of diabetes mellitus. This suggests that these levels are important for somatic health and should be taken into account when using hair corticosteroid analysis in future studies.
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1. Introduction

The hypothalamic—pituitary—adrenal (HPA) axis has been subjected to a wide array of research questions, with one of the main interest areas being one of its end products, the hormone cortisol.

Until a decade ago, cortisol had predominantly been measured in saliva, serum or urine, which allows analyzing the dynamics of cortisol production for up to 24 h. Then, a new technique was developed to measure long-term cortisol levels in scalp hair, allowing quantifying the average cortisol production for a period ranging from 1 month to several months or even years (Manenschijn et al., 2011; Raul et al., 2004). The number of studies on hair cortisol concentrations (HairF) substantially increased throughout the past years (Wester et al., 2014a). While HairF tend to remain stable in health, increased as well as decreased HairF have been associated with a range of pathological and psychological conditions such as cardiovascular disease, obesity, Cushing's disease, and mood and anxiety disorders (Dettenborn et al., 2012a; Manenschijn et al., 2012, 2013; Steudte et al., 2011; Wester et al., 2014b).

Determinants of HairF have been described before (Dettenborn et al., 2012b; Feller et al., 2014; Stalder et al., 2012). Two of these studies (Dettenborn et al., 2012b; Stalder et al., 2012) investigated simple associations of different socioeconomic and health variables with HairF, whereas one study (Feller et al., 2014) reported socioeconomic and health-related determinants of HairF in older adults using partial and full mutual adjustment. The first two studies showed associations of HairF with age (higher HairF in young and old age), sex (men show higher HairF than females), metabolic syndrome (MetS) (higher HairF in presence of MetS), and weight-related anthropometric measures (increasing HairF with increasing body mass index (BMI)). The third study confirmed these simple associations and reported positive associations with smoking, diabetes mellitus, mental health, daytime sleeping, working status, and negative associations with diastolic blood pressure. However, in full mutual adjustment, only age and smoking remained independent predictors of HairF. These studies have produced important results, however, a systematic study with a large sample size and partial as well as mutual adjustment for predictors of HairF in a younger age group is still lacking. Furthermore, in different publications, the effects of sociodemographic and health and lifestyle variables on HairF show a noticeable discrepancy. Existing literature aiming at summarizing the effects of influencing factors have reported inconsistent and conflicting results for age, sex, alcohol and nicotine use, physical activity, and for hair (treatment) characteristics. Factors that could account for these incongruent findings are the relatively small sample sizes, therefore possibly rendering power problems. Furthermore, most studies varied in the extent to which they took other covariates into account. Disentangling these effects is of paramount importance to enable future studies to adjust their analyses for potential confounding factors. This seems especially important for research on stress and mental illness, as the effects of these conditions on the HPA-axis have been shown to be small (Staufenbiel et al., 2013), rendering them vulnerable to being overshadowed by stronger effects when these are not controlled for. This could be one of the reasons that for example self-reported stress, has shown divergent relations with HairF, with positive, inverse, and no correlations (see Staufenbiel et al., 2013 for review).

Until now, most studies on HairF have been conducted using immunoassays (Gow et al., 2010). With the recent development and validation of HairF measurements with liquid chromatography tandem-mass spectrometry (LC—MS/MS) (Gao et al., 2013; Noppe et al., 2015), the quantification of other steroids in scalp hair has become possible. As cortisol can be converted into inactive cortisone, the assessment of hair cortisone (HairE) in parallel to HairF may give even more insight into the cumulative amount of active and inactive corticosteroids in the body. Following the argumentation of Stalder et al. (2013), as recent research suggested that under specific circumstances, salivary cortisone may provide a better reflection of systemic cortisol levels than salivary F (Perogamvros et al., 2010). The authors showed that salivary cortisone closely reflected free serum cortisol after adrenal stimulation and hydrocortisone administration, and that it was unaffected by changes in corticosteroid-binding globulin (CBG). Furthermore, because HairE has been shown to be approximately 3—4-fold higher than HairF (Stalder et al., 2013), it might also be more readily measureable than HairF. To further investigate the relevance of HairE, we also calculated the F/E-ratio, and an additive $F \pm E$ measure as an indication of total glucocorticoid supply.

Results on both HairF and HairE have been reported in first publications (Kuehl et al., 2014; Raul et al., 2004; Stalder et al., 2013; Zhang et al., 2013). In all studies, HairF and HairE strongly correlated. However, all reports on HairE have been exploratory, and as of yet no study has set out to systematically assess determinants of HairE. The relevance of these determinants follows the same argumentation as for the determinants of HairF described above (Kuehl et al., 2014).

We therefore set out to examine the wide range of potential determinants of both HairF and HairE levels within one well-studied cohort. This seems necessary for future research as it aids in deciding which factors need to be assessed apart from the variable of interest in order to control for their influence on HairF and HairE. The present study uses detailed information of 760 subjects participating in the Netherlands Study of Depression and Anxiety (NESDA) to examine which (1) sociodemographic, (2) health & lifestyle,

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