**Title:** Hair and Salivary Cortisol and Cumulative Stress Exposure Among Premutation Carrier Mothers of Children with FXS

**Authors:** Jinkuk Hong¹, Christopher Coe², Amita Kapoor³, Leann Smith DaWalt¹, Jan Greenberg¹, David Almeida⁴, and Marsha Mallick¹

**Introduction:** Cortisol has been extensively studied as an indicator of the impact of stress exposure and poor health. Cortisol levels can be measured in various tissue types, including saliva, blood, urine, and hair. In this study, using a longitudinal sample of premutation carrier mothers of adolescents and adults with FXS, we first evaluated the stability of cortisol levels in the mothers across two tissue types (saliva, hair) over a nearly ten-year span, and then examined the associations between cumulative stress exposure, health, and cortisol levels across the study period.

**Method:** Participants (n=86) were drawn from an ongoing longitudinal study, Family Adaptation to Fragile X Syndrome, which has collected data at four time points over 10 years. The salivary cortisol was assessed at Time 1. Study participants, who were mothers of son/daughter with FXS, provided saliva samples four times a day (at awakening, 30 minutes after awakening, before lunch, at bedtime) for four days, which were averaged and used as the measure of salivary cortisol for the present analysis. The hair cortisol was assessed at Time 4. Mothers provided a hair lock from the posterior vortex area, cut close to the scalp, and a 4 cm length finely ground and assayed using liquid chromatography-tandem mass spectrometry.

Correlates of hair cortisol were examined in two domains: stressor exposure and health. The stressor measures include negative life events experienced by the mother and her family members, functional independence of the son/daughter with FXS, and the number of behavior problems of the son/daughter with FXS. The maternal health measures include self-rated health, number of prescription medications, depressive symptoms, and anxiety symptoms. All measures were assessed at all four time points of the study, and in order to assess cumulative effects of stress exposure and health problems, each of these measures was summed over the four waves of data collection.

We hypothesized that higher levels of stress exposure and more physical and mental health problems would be associated with higher levels of hair cortisol at Time 4, controlling for levels of salivary cortisol at Time 1.

**Results:** Regarding our first research question concerning the stability of cortisol measured across nearly ten years using two tissue types, hair cortisol was significantly correlated with salivary cortisol level measured about ten years earlier (r = .258, p=.018). Specifically, hair cortisol was correlated with the daily decline of salivary cortisol (from 30 minutes after awakening to bedtime; r = .316, p = .003), but not with the morning rise (from awakening to 30 minutes after awakening). Regarding the second research question, regression analyses supported the hypothesis that cumulative stress exposure and maternal physical and mental health conditions would be associated with hair cortisol. All regression models controlled for prior levels of salivary cortisol and maternal age. Specifically, negative life events (b = .005, p =.011, see Figure 1) and the functional independence of the son/daughter with FXS (b = .0004, p =.031) had significant curvilinear associations with hair cortisol. A greater number of negative life events was associated with higher levels of hair cortisol. For the functional independence, both lower and higher level of independence were associated with higher levels of hair cortisol. However, behavior problems of the son/daughter with FXS were not clearly associated with hair cortisol levels. Self-rated health was negatively associated with hair cortisol (b = -.107, p = .025): better self-rated health was reflected by lower hair cortisol. The number of prescription medications taken by premutation carrier mothers (b = .045, p = .005), maternal depressive symptoms (b = .007, p = .034), and at a trend level, maternal anxiety symptoms (b = .009, p = .071) were linearly associated with hair cortisol. Cumulative physical and mental health problems were associated with higher levels of hair cortisol.

**Discussion:** This study is one of few that has explored longitudinal associations between hair cortisol and salivary cortisol, and the only analysis to consider a nearly 10 year span and the extent to which cumulative stress exposure and health problems predicted hair cortisol. Consistent with a prior study (Short et al., 2016), hair cortisol was significantly associated with salivary cortisol. One benefit of measuring cortisol levels in hair is that it is not affected by the diurnal rhythm of cortisol secretion or the timing of collection. Furthermore, the associations between stress exposure, physical and mental health problems, and hair cortisol levels were all in hypothesized directions, consistent with prior research (Staufenbiel et al., 2013). These findings suggest that hair cortisol can serve as a summative biomarker of the caregiving stress and health problems experienced by premutation carrier mothers of children with FXS.
References:


3 Waisman Center, University of Wisconsin-Madison.
2 Department of Psychology, University of Wisconsin-Madison.
3 Wisconsin National Primate Research Center, University of Wisconsin-Madison.