Increased salivary and hair cortisol and decreased salivary alpha-amylase concentrations in obese prepubertal girls

# Chrysanthe Papafotiou<sup>1</sup>, Eirini Christaki<sup>2</sup>, Erica L.T. van den Akker<sup>3</sup>, Vincent L. Wester<sup>4</sup>, Filia Apostolakou<sup>5</sup>, Ioannis Papassotiriou<sup>5</sup>, **George P. Chrousos<sup>2</sup> and Panagiota Pervanidou<sup>1</sup>**

<sup>1</sup>University of Athens Medical School, Athens, Greece, <sup>2</sup>University of Athens, Greece, <sup>3</sup>Erasmus MC, <sup>4</sup>Erasmus MC, Rotterdam, Netherlands, <sup>5</sup>Aghia Sophia Children's Hospital, Athens, Greece

#### Introduction

Obesity has been associated with perturbations of both the hypothalamic-pituitary-adrenal (HPA) axis and sympathetic nervous system (SNS). Scalp hair cortisol has been recognized as a reliable index of long-term cortisol concentrations.

Table 1. Basic	population of Obesity Group	omparisons b Normal Weight Group	y group P value
Age (years)	7.4 (1.3)	7.8 (1.1)	0.206
BMI (kg/m²)	24.6 (3.3)	16.9 (1.6)	<0.001**
BMI z-score	2.9 (1.3)	-0.1 (0.5)	<0.001**
Hair Cortisol (pg/mg hair)	4.1 (5)	1.2 (0.6)	0.008*
Student's t-te	st		
Mann-Whitne	y for Hair Cor	tisol	

Figure 2. Salivary cortisol at each time point in obese and normal weight girls (p<0.05 for both groups (Anova-repeated measures)



### Objective

The aim of this study was to compare indices of HPA axis and SNS activities, such as, respectively, the salivary cortisol and alpha-amylase diurnal patterns, and their relations in obese and normal weight prepubertal girls. Furthermore, we compared cortisol concentrations in scalp hair between groups and investigated whether they are correlated with salivary cortisol concentrations

### Results

•Hair cortisol levels were higher in the obese than the normal-weight girls (p<0.05)

•A positive linear correlation between hair cortisol and BMI z-score was found (rho=0.319, p<0.05)



Figure 3. Salivary alpha-amylase at each time point in obese and normal weight girls p<0.05 for sAA at 12:00 and 15:00



## **Participants**

**50 prepubertal girls completed the study** 

- Obesity Group (OG): 26 obese girls (BMI 24.6 ±  $3.3 \text{ kg/m}^2$ )
- Normal Weight Group (NG): 24 normal weight

•Hair cortisol correlated positively with salivary cortisol at 12:00 (rho=0.417, p<0.05), salivary cortisol at 18:00 (rho= 0.338, p<0.05) and AUCg (rho=0.311, p<0.05)

•Significantly higher salivary cortisol concentrations at all time points and Area Under the Curve (AUCg) were observed in the OG (p < 0.05)

girls (BMI 16.9  $\pm$  1.6 kg/m<sup>2</sup>)

Matched for age

**Exclusion criteria:** 

•Tanner Stage > 1 •Syndromic obesity •Use of medication or recent infection

### Methods

- Five saliva samples were collected serially over a weekend day (at 9:00, 12:00, 15:00, 18:00, 21:00 hours)
- Cortisol and alpha-amylase concentrations were electrochemiluminescence by measured kinetic-reaction immunoassay and assay, respectively
- Areas Under the Curve with respect to ground

•Inversely, salivary alpha-amylase at 12:00 & 15:00, as well as Area Under the Curve (AUCg) were significantly lower in the OG

•Negative linear correlations between cortisol and alpha-amylase were found (p<0.05) only in the obesity group

Figure 1. Hair cortisol concentrations between groups (p=0.008\_





# Conclusions

- Obese prepubertal girls demonstrated changes in both salivary cortisol and alpha-amylase diurnal secretions compared to normal weight controls
- These results suggest altered stress system function in the obese group
- Increased hair cortisol concentrations and positive correlations with salivary measurements suggest chronic stressrelated activation of the HPA axis in obese girls
- Hair cortisol appears to be a sensitive measure of hypercortisolism in obesity.

# References

- (AUCg) for cortisol and alpha-amylase were calculated
- Cortisol and alpha-amylase linear correlations were investigated in each group
- Hair samples from the posterior vertex of the scalp were cut as close to the scalp as possible and analyzed for cortisol with LC-MS/MS-based method
- Cortisol was extracted overnight in methanol, solid by followed phase extraction. Quantification of cortisol was performed using a Waters Xevo TQ-S LC-MS/MS system
- BMI z-scores were calculated based on the contemporary Greek growth charts

Hair Cortisol (pg/mg dry weight)

#### Table 2. Hair Cortisol correlations

BMI z-score	r=0.319	p=0.037*
Salivary Cortisol 12:00	r=0.417	p=0.008*
Salivary Cortisol 18:00	r=0.338	p=0.033*
Salivary cortisol AUCg	r=0.311	p=0.048*
Spearman's rho *p<0.05		

Chrousos GP. Stress and disorders of the stress system. Nature reviews Endocrinology. 2009;5(7):374-81

Veldhorst MA, Noppe G, Jongejan MH, Kok CB, Mekic S, Koper JW,

et al. Increased scalp hair cortisol concentrations in obese children. The Journal of clinical endocrinology and metabolism. 2014;99(1):285-90.

Pervanidou P, Bastaki D, Chouliaras G, Papanikolaou K, Laios E, Kanaka-Gantenbein C, et al. Circadian cortisol profiles, anxiety and depressive symptomatology, and body mass index in a clinical population of obese children. Stress. 2013;16(1):34-43

Pervanidou P, Chrousos GP. Metabolic consequences of stress during childhood and adolescence. Metabolism: clinical and experimental. 2012;61(5):611-9

# Sources of research support

The authors declare that there are no conflicts of interest. This study received no private or corporate financial support. This work was supported by University of Athens and the "Aghia Sophia" Children's Hospital in Athens, Greece

