Reviewer's report

**Title:** High end of normal ACTH and cortisol ranges are associated with specific cardiovascular risk factors in pediatric obesity: cross-sectional study

**Version:** 1  **Date:** 13 September 2012

**Reviewer:** Jack A Yanovski

**Reviewer's report:**

Prodam et al present cross-sectional data from a large cohort of children and adolescents regarding associations between ACTH/cortisol and metabolic parameters. These data may be of interest, but there are concerns about the analysis that must be answered. In particular, we have insufficient information about the timing for ACTH/cortisol collection to reach a conclusion regarding the validity of these data. In addition the use of BMI, rather than age- and sex-specific BMI-Z score, as a covariate may have affected results.

Specific suggestions:

1. The manuscript could benefit from a careful reading by a native English speaker – there are instances of awkward phrases and incorrect grammar throughout. For example, “strictly associated” isn’t really well defined in English – presumably the authors mean associated even after accounting for covariates, but that isn’t clear.

2. Last sentence in abstract: As these findings are cross-sectional and far from definitive on the causal direction, revise sentence to read: “These specific associations suggest complex mechanisms through which the HPA axis may contribute to metabolic impairments in obesity.”

3. Specify the IFG and IGT cut points used in Methods

4. Page 6: At what time, exactly, were cortisol and ACTH measured? As the authors certainly know, these variables are very time-sensitive. “Morning” is not sufficiently precise. Did the time of day for phlebotomy vary significantly among the three tertiles of ACTH/cortisol? This variable should have been taken into account in the analysis.

5. Page 8: It is unclear in Methods how you will use the blood pressure cut-offs of 95th and 90th percentiles. Please indicate here that 95th was used to define hypertension and 90th was used for your definition of one criterion of metabolic syndrome.

6. Page 7: Does “diet-naïve” mean “weight-stable”? If so for how long?

7. Page 8: As part of the metabolic syndrome evaluation, was waist circumference measured? If measured, please include in the manuscript.
8. Page 8: “Triglycerides and HDL-cholesterol percentiles were referred to Lipid Research Clinic Pediatric Prevalence Study [21]” – does this mean you used the cut-points for Total and LDL-cholesterol specified in Table 1 of this document (total >200, LDL >130)? Please be more exact.

9. Page 9: Minor point: “Analysis of covariance was used to determine differences in those with an without....” should be “Analysis of covariance was used to determine differences in those with and without...”

10. Was a power calculation performed for this study? Regardless, please indicate in Results the power of the study for detecting differences in the studied variables.

11. Page 9: If the authors truly meant what I believe the phrase “near tertiles” means, then they wouldn’t have cut points like 16.10 and 26.93 for ACTH – these would be 16 and 27. It seems likely that exact tertiles were used.


13. Minor point – the authors likely mean “sex” when they use “gender” – please correct throughout the manuscript.

14. Hypertension was diagnosed in a remarkably large percentage of these subjects – these data are far from what is reported generally. Is there some referral bias that could account for this strange finding? Please also specify what the American Academy of Pediatrics criteria are, either here or in Methods. The sentence “Only 1 subject fulfilled all the cardiovascular criteria, while 63 (15.5%) failed to meet the criteria” is very unclear at present.

15. Page 10 – It appears, from the legend in Table 2, that the paragraph starting with “ACTH and cortisol levels were positively associated with...” should indicate that the first results are unadjusted for covariates – please revise to: “In unadjusted analyses, ACTH and cortisol levels were positively associated with...” Probably preferable is not to present the unadjusted results at all – simply remove the unadjusted discussion and remove Table 2, which appears to show unadjusted data, from the manuscript.

16. The phrase “positivity for cardiovascular risk factors” is not proper English – please revise throughout the manuscript to read “presence of cardiovascular risk factors”

17. For the analysis of ACTH and cortisol among those with metabolic abnormalities, again, only the adjusted results should be discussed.

18. Discussion – for this paper, an explication of prior findings in adults would actually be quite useful to place these results in perspective.

19. Please carefully rephrase the sentence, “We showed that both hormones
were continuously and directly associated with glucose, triglycerides, and blood pressure, while cortisol also with LDL-cholesterol independently by confounders including gender, age, puberty, BMI and insulin resistance.”

20. Page 14: “but not HDL-cholesterol” should be “but not lower HDL-cholesterol”

21. Discussion seems a bit too directed towards the notion that high ACTH and cortisol are causing the metabolic abnormalities observed – the resolution of high ACTH and cortisol after weight reduction suggests HPA axis dysregulation is more frequently a consequence, rather than a cause, of obesity-associated abnormalities. Most of the references to Cushing syndrome could thus profitably be removed.

22. Another significant limitation not mentioned is the absence of true body fat measurements from DEXA. Socioeconomic status, which has been found to affect stress/cortisol levels, was not accounted for. Also absent is any normal weight group. There is also no statistical control for the many comparisons being made in the paper.

23. Table 1 would be improved by dividing the children into the two available groups (overweight, and obese) - and indicating how many children in each group had abnormal results for the metabolic parameters. Give pubertal stage information separately for boys and girls.

**Quality of written English:** Not suitable for publication unless extensively edited

**Statistical review:** No, the manuscript does not need to be seen by a statistician.

**Declaration of competing interests:**

I declare that I have no competing interests (Dr. Condarco similarly has no competing interests).