The authors thank the editor and the reviewers for their invested time and the important suggestions.
Please find a point-by-point response to every reviewer's comments below. The newly corrected parts are marked with the "Track Change System" within the manuscript.

We hope that these changes are sufficient to result in acceptance of the manuscript.

**Reviewer 3**

This manuscript reports a comprehensive study into the transcriptome of visceral adipose tissue in dogs in which chronic hypercortisolemia was induced by treatment with ACTH. The data presented are extensive and indicate that, effectively, a model of Cushing's Syndrome was developed in the dogs.

I feel that the study has been well done and reported. I have a few comments that the authors may consider in any revision of their manuscript.

R: The authors highly appreciate the overall comments regarding the content and main findings of the project, and all the suggestions to improve the overall quality of the manuscript.

While the discussion of the findings is comprehensive, and the possibility of the dog as a model of Cushing's Syndrome in humans is apparent, I would like to have seen the findings placed into more context physiologically. How are the findings relevant to the onset and physiological regulation of Cushing's Syndrome beyond simply presenting potential targets for further clinical studies? I feel that the development and presentation of a hypothesis in the Introduction would assist in dealing with this point.

R: The authors thank the reviewer for this remark and suggestions. In the present work, hypercortisolism clearly induced multifactorial effects on VAT physiology and function. The factors highlighted in the manuscript include, i.a., regulators of cortisol availability (HSD11B1/2), growth factors (IGF1 and FGF1), ECM regulators (TIMP4), immune modulators (TLR7, IL1R1, CCR2, CXCR4), vascular factors (ECE1, THBS1) and hormone receptors (ESR1 and PRLR). Thus, the discussion section spanned over several mechanisms that might be associated with cortisol-induced changes on VAT activity. There was an attempt to explore all these functions throughout the discussion and highlight possible relations they may present with the causes and consequences of the clinical signs observed in Cushing's Syndrome. All these, while keeping in mind the clear limitations of NGS, i.e. that gene expression does not directly refer to protein availability and/or molecular function. Thus, in these regards, the authors suggest not to go deeper on physiological regulation, due to the risk of making over interpretations of their own data. This appears to be acknowledged by the comments from the reviewer 2. Nevertheless, following the reviewer's suggestion, a general hypothesis has been added to the manuscript, being further highlighted twice in the discussion section.

Was sex taken into consideration in the analyses? Sex differences in the functioning of the hypothalamo-pituitary adrenal axis have been well documented in a range of species.

R: A high homogeneity between all the samples within each group, observed, e.g., in the hierarchical correlation of samples presented in supplemental figure 1A, suggests a low effect of confounding variables, such as sex, age or other individual variations in the current transcriptomic analysis results. Thus, a more refined analysis of possible sex-effects was not performed. Furthermore, by having only 5 animals, statistical significance of such analysis would not be possible. The authors have included this point as limitation of the manuscript.

The number of subjects in this study was small. Was this adequately accounted for in the analyses?

R: We are aware that the number of dogs was low with only 5 animals. The experimental setting with a higher number of dogs would have been difficult also concerning the animal experimental permission. We have included this as a limitation in the manuscript, and the number of replicates entered the statistical analysis and was considered when computing the p-values.

The terminology for glucocorticoid receptor needs to be clarified in the manuscript. Glucocorticoids bind to mineralocorticoid receptors, also termed glucocorticoid receptor I, and glucocorticoid receptors, also termed glucocorticoid receptor II.

R: We thank the reviewer for this critical remark. We were not aware of this terminology.

What we found within the literature was the following in “Glucocorticoid Receptor” Nicolas C. Nicolaides, MD, PhD, George Chrousos, MD, MACP, MACE, FRCP, and Tomoshige Kino, MD, PhD in Endotext, National Library of Medicine, Feingold KR, Anawalt B, Boyce A, et al., editors.South Dartmouth (MA): MDText.com, Inc.; 2000. …”. GR is a member of the steroid hormone receptor (SR) subfamily (subfamily 3) of NRs (nuclear receptors). This receptor family of vertebrates consists of six evolutionarily related SRs: two for estrogens (estrogen receptor (ER) a and ERβ) and one each for androgens (androgen receptor: AR), progestins (progesterone receptor: PR), glucocorticoids (GR), and mineralocorticoids (mineralocorticoid receptor: MR). These steroid receptors are also categorized as type I receptors. However, in the manuscript, we used the term “glucocorticoid receptor” always in combination with its gene name “NR3C1”, which per definition is the glucocorticoid receptor (nuclear receptor subfamily 3, group C, member 1), while the name of the mineralocorticoid receptor is “NR3C2”.

Therefore we think that there shouldn't be any confusion for the reader. If this is not in agreement with the reviewer, we would appreciate a suggestion on how to adapt the text, to avoid misunderstanding for the reader of the manuscript.

Please note that the word data is plural, so it should be data “were” uploaded and date “were” trimmed.

R: We thank the reviewer for this remark and have changed it accordingly in the text.