Reviewer #1: This is an interesting observational study of a rare category of patients. The findings give insight into the effects of prolonged GH-exposure in humans and have implications beyond acromegaly. However, the design raises serious concerns which should be addressed further.  
The use of non-functioning pituitary adenoma patients as control group is problematic. This could, at least partly, be addressed by a more thorough description of the subjects. What medications did the patients use? Where any of these treatments paused during examinations, and if so for how long? Many drugs like beta adrenergic receptor blockers, insulin and oral antidiabetic drugs can influence the gene-expression profile in adipose tissue and this should be addressed and discussed. Similarly all co-morbidities of the patients should be presented and discussed.

**We have full information of patient co-morbidies and medications. As mentioned, one of the Acromegly patients was treated (with an insufficient clinical response) with a somatostatin analog . The rest were untreated. One patient in each group was treated with Metformin. Two of the non-functioning adenoma patients were treated with beta blockers. None of the patients was treated with insulin. This information has been added to the methods section.**

The use of 3T3-F442A exposed to GH for 48 hours as a comparator to acromegaly is oversimplified (page 8 lines 183-193) and it conflicts with the premises for the study where both direct and indirect effects of GH are investigated. The number of genes regulated does not match (418 vs. 560 genes lines 187-188). The paragraph should either be revised or omitted from the manuscript.

**We agree this this experiment is a sub-optimal mimic for our study, and as requested it has been removed from the revised manuscript**

The lipolytic enzyme ATGL is regulated by expression of several co-factors (CIDEA, CIDEC and G0S2) (PMID: 24577718). Where mRNA for any of these co-factors altered in acromegaly?

**CIDEA and CIDEC expression were not different between the groups. G0S2 trended toward being induced (1.4 fold, p=0.03 q=0.2). These data are now included in the revised manuscript:**

**We also examined *CIDEA/B/C* and *G0S2*, which have also been proposed to be positive regulators of lipolysis [1]. While there were no changes in the former, we did observe a non-significant elevation in *GOS2* (1.53 fold, q=0.246; Supplementary Figure 2C).**

Reviewer #2: This manuscript by Hochberg et al. analyzed gene expression signature in adipose tissue of acromegaly patients vs. non-functioning pituitary adenomas patients by high throughput mRNA sequencing. The authors found that expression of many genes are altered in the adipocytes of acromegaly patients and suggested that alterations of these genes likely responsible for the observation of insulin resistance, increased lipolysis, and metabolic changes in the patients. While the study is interesting and may provide explanations to the observed phenotypes in acromegaly patients, this study faces difficulties in sample size and technical limitations.  
Major points:  
1. The major caveat of the study is small sample size. In addition, the subjects used are not age-matched. While the authors have tried to take the age effects on this study into consideration and separated the subjects to two groups, under 60 or over years old, without knowing the age distribution of the subjects it is not clear why 60 years old was chosen. Along this line, the acromegaly subjects over 60 are 2 and the subjects under 60 years old is 5 (according to Fig. 3C). The sample size for acromegaly patients under and over 60 years old is even smaller. Thus, it not clear that the gene signature identified in this study would make meaningful implications regarding acromegaly phenotypes.

**We chose 60 as the cutoff based on previous studies on acromegaly, and this age being median within our data set [2]. This PCA plot has been added as Supplementary Figure 1 and has been described in the revised methods section as such:**

**To account for potential age-dependent changes in the subjects, we separated the patients into two groups, based on the median value, under 60 years of age versus 60 and above as has been previously reported for acromegaly studies [2]. As shown in Supplementary Figure 1, an age cutoff of 60 separated our samples along the first principal component efficiently into two distinct phenotypes, with 7/8 of the acromegalic subjects in one group and 10/11 of the controls in the other group.**

2. The authors’ conclusion is based on the analysis of mRNA-seq. The difference in gene expression between acromegaly patients and control needs to be validated with another gene expression analysis, such as qPCR, to further support the results.

**We considered performing qPCR studies to ‘re-validate’ some of our gene-expression findings but are unable to as there is insufficient remaining RNA and tissue. There is scant evidence that qPCR analyses from the same samples would add any extra validity to our data as previous studies have shown extremely close correlations between qPCR and RNAseq data** [3–6]**, with differences largely being attributable to probe bias or non-linearity of qPCR not RNAseq. Ideally, we would re-validate our findings in a separate cohort of acromegalic patients, but due to the difficulty in obtaining new samples, those experiments are not possible at this time.**

3. What are serum GH levels of all the subjects?

**Serum GH for the acromegaly patients was in the range of 0.7-28.7 ng/ml, with a mean 8.4 ng/ml and median of 7.1 ng/ml. Serum GH levels were not measured for our non-secreting adenoma subjects but the average normal range of GH is < 5ng/mL.**

Minor points:  
  
1. The statement on p9, lines 207-213 should be removed. While the serum IGF1 levels correlates with the mRNA levels in adipose tissue, it is not clear if this is due to the increase in IGF1 mRNA in adipose tissue. Since liver IGF1 mRNA levels are not analyzed in acromegaly patients, it is not clear that the increase in adipose tissue IGF1 mRNA contributes the increase in serum IGF1 levels.

**We agree with this comment and will omit this speculation.**

2. Should “larger” in line 169 be changed to “smaller”? From Fig. 3, the difference in gene expression in subjects over 60 years old vs. control is smaller.

**We have made this change, and thank the reviewer noticing this error.**

**References**

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3. Griffith M, Griffith OL, Mwenifumbo J, Goya R, Morrissy a S, et al. (2010) Alternative expression analysis by RNA sequencing. Nat Methods 7: 843–847. doi:10.1038/nmeth.1503.

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6. Shi Y, He M (2014) Differential gene expression identified by RNA-Seq and qPCR in two sizes of pearl oyster (Pinctada fucata). Gene 538: 313–322. doi:10.1016/j.gene.2014.01.031.