Reviewer’s report

Title: The rs361525 polymorphism does not increase production of Tumour Necrosis Factor Alpha by monocytes from Alpha-1 Antitrypsin Deficient subjects with Chronic Obstructive Pulmonary Disease

Version: 0 Date: 21 Sep 2015

Reviewer: Manuela Plate

Reviewer's report:

In the present study Gane and colleagues examine the association between the rs361525 SNP and TNF-α expression and secretion in monocytes derived from COPD/alpha-1 antitrypsin deficient subjects. They report no significant association between the SNP and TNF-α mRNA levels or secreted protein at baseline and following LPS stimulation.

Unfortunately there are many limitations to this study, as pointed out by the authors themselves in the discussion.

Major revisions:

1) I appreciate the difficulties in obtaining and culturing primary human bronchial epithelial cells, but given the previous observation that subjects with the rs361525 SNP have significantly increased TNF-α levels in sputum, while systemic levels are not elevated, the manuscript would be stronger if it incorporated also studies on at least sputum-isolated macrophages.

2) The negative result might be due to a statistical type II error, given the small size of the cohort and the high inter- and intra-subject variability. I would therefore suggest that this study be treated as a small pilot study, indicating this in the title and in the text.

3) It might be useful to add an in silico study of the effect of the minor allele of the SNP on transcription binding sites and a brief discussion of this in the context of monocytes and macrophages and other relevant cell types.

4) I recommend using at least 2 validated stably expressed reference genes. This will provide statistically more significant results and help detecting small expression differences.
Minor revisions:

5) The rs361525 SNP is commonly indicated in the literature as being at position -238 while the authors indicate it as at position -237. Please clarify.

Level of interest
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An article of limited interest

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