Reviewer's report

Title: Polymorphisms in the glutathione pathway modulate cystic fibrosis severity: a cross-sectional study

Version: 5 Date: 11 June 2013

Reviewer: Andreas Hector

This study by Fernando AL Marson and colleagues describes associations of SNPs in the GSH pathway with clinical parameters of CF patients. Various SNPs were associated with e.g. PSA infection, the Bhalla score, oxygen saturation or osteoporosis. Yet, in this reviewer's view, some aspects are lacking in this manuscript.

Major Compulsory Revisions:

• Although the authors use the Shwachman-Kulczycki score and other clinical scores, in this reviewer's opinion, lung function tests would be preferable for this kind of studies. Large gene modifier studies in the USA use preferentially longitudinal lung function data as described by Schluchter et al. (AJRCCM 2006). This reviewer would also rather choose this clinical parameter (for example estimated FEV1 at age of 20 years) for genetic association studies instead of cross-sectional analyses. Surely, longitudinal lung function data are not available in all CF centers, but the authors should at least discuss, why they have not taken the lung function tests into account in this study. And if longitudinal lung function is not available, association with cross-sectional lung function should be included.

• The authors conducted statistical analyses by subdividing the CF population by the numbers of CFTR mutations they have found in each patient (0, 1 or 2 mutations identified). Unfortunately, to this reviewer it is not clear, why the authors chose this method. Why would the numbers of found mutations be important for the effect of gene modifier? Please explain in the manuscript.

• Furthermore, are there any associations between the CFTR mutations and the SNPs? Is the OR of the various SNPs higher (or lower) for example in dF508 carrying individuals (homozygous/heterozygous) compared to non-dF508 subjects?

• One limitation of this study is the low numbers of CF patients included. This should be discussed by the authors.

Minor Essential Revisions:

• There was a formatting problem with the legend of Table 1 - it was hidden behind the table.

Discretionary Revisions:
• In gene modifier studies, it is also very important to investigate functional effects of the SNPs. Because all the SNPs assessed in this study are involved in the GSH pathway, it would be very helpful to measure GSH/GSSG levels in blood or even airway samples of the CF patients. Alternatively or additionally activity of the enzymes could be measured. This would most likely further increase the quality of this study.

• The authors describe the association between PSA (non-mucoid and mucoid) with different SNPs. However, was there a difference for different infection stage (for example according to the consensus paper by Doering et al. (JCF 2012) - chronic infection > 50 % positive microbiological analyses in the last 12 month, intermittent infection > 0 and # 50 %, negative or never)?

• In this reviewer's opinion, the study by Gu et al. (Nature 2009), which is a important recent gene modifier studies in CF, should be discussed in this manuscript, too. The latter study was a whole genome study and, to this reviewer's knowledge, none of the investigated genes in this manuscript was found to be relevant for the CF lung disease.

Level of interest: An article whose findings are important to those with closely related research interests

Quality of written English: Acceptable

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.