Reviewer’s report

**Title:** Effect of GSTM2-5 polymorphisms in relation to tobacco smoke exposures on lung function growth: a birth cohort study.

**Version:** 1  **Date:** 8 March 2013

**Reviewer:** Carrie Breton

**Reviewer’s report:**

- **Major Compulsory Revisions**

  1. It remains unclear from the methods section and figure 1, whether all CpG loci in the Illumina 450K array were interrogated with respect to GSTM2-5 genetic variation or only select loci. If only a selection, then how were these chosen? If all were used, what is the rationale? Given the hypotheses of the paper, it would seem the authors would be most interested in evaluating only the CpG methylation within the GSTM2-5 genes. Please provide clarification, as power to detect associations will be affected by which approach was employed.

  2. What quality control measures were applied to the Illumina 450K array data? For instance, did the authors remove CpG loci on X and Y chromosomes, or did they remove CpG loci known to be affected by a SNP at the exact locus, etc? (This filtering is largely irrelevant if they only evaluated methylation withing GSTM2-5 genes). Nevertheless, if they assessed >200 subjects some commentary on evaluation of batch or plate effects is warranted.

  3. Please clarify that none of your observed significant metQTLs are in fact a result of a base change that removes or creates a CpG site directly?

- **Minor Essential Revisions**

  4. Not all references in 2nd paragraph of introduction are formatted properly.

  5. Please specify in the methods section what type of blood sample was used for methylation analyses and at what age i.e. cord blood, whole blood, separated cells, etc?

- **Discretionary Revisions**

  6. One additional reason that the authors only observed a significant interaction between SHS at age 18 and diplotype on methylation of GSTM2 may be the timing of methylation assay. As noted by the authors, methylation was assayed at age 18, and most directly correlates in time. Because methylation is not likely to be static over time, evaluation of methylation only at age 18 may not represent early methylation levels enough to correlate with in utero or early childhood exposures.

  7. It would be interesting if the authors could conduct a mediation analysis to test whether altered DNA methylation of GSTM 2 and 5 is on the causal pathway
between diplotype and lung function.

**Level of interest:** An article of importance in its field

**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 I have no competing interests