Reviewer’s report

Title: Aberrant GSTP1 promoter methylation is associated with increased risk and clinical stages of breast cancer: a meta-analysis of 19 case-control studies

Version: 3
Date: 10 June 2015
Reviewer: Saverio Alberti

Reviewer’s report:

Major Compulsory Revisions

Redox regulation has been amply studied in human tumors. GSTP1 in particular has been the subject of considerable investigation. Although a comprehensive study on the role of GSTP1 in cancer is of potential interest, this reviewer finds several debatable issues in the proposed meta-analysis.

1. A first key issue is: only one of the studies (Brooks 2010, ref 25) did not find more aberrant methylation in cancer patients. Hence, what is the fundamental reason of performing the meta-analysis, as the result was expected, as essentially concordant in most studies?

2. An additional issue would, on the other hand, have required much more debate: why is blood DNA methylation expected to be a faithful indicator of promoter methylation in cancer?

Several studies have indicated heterogeneity in methylation patterns among different tissues for different genes.

If GSTP1 is being selected by the process of tumor progression, the more so if higher association is with late stage disease, it should be experimentally defined why methylation differences are expected in DNA from cells that had not been subjected to such a selection.

Not surprisingly, the negative study cited above was only conducted on blood cell DNA.

Consistent, the meta-analysis itself finds much lower hazard ratios in PBL than in cancer (OR=4.02, 95%CI=1.12-14.38), and CI border the value of 1.

3. An additional reason of worry is the detection method. Best-quality results are expected from quantitative methylation analyses. These provided by far the smallest HR (Quantitative: OR=4.73, 95%CI=1.84-12.12). Highest associations were found for Semi-quantitative (OR=10.33, 95%CI=3.32-32.10) and Non-quantitative: OR=12.55, 95%CI=5.72-27.55) analyses, but seemed to have no impact on the conclusions of the study.

4. When only quantitative analyses of GSTP1 promoter methylation in blood DNA are pooled, is association of promoter methylation and cancer cases still confirmed?
5. Publication bias was found by this analysis, but was not discussed, and had no impact on the conclusions of the study.

Minor Essential Revisions

1. HR for late-stage disease appear ‘reversed’. As increased association is with late stage disease a better presentation of these findings would be late stage/early stage ratios, for increased HR as associated with late stage.

2. When removing the study by Brooks et al, 2010 heterogeneity measures decrease. This is almost tautologic (this was the only negative study included) and should be avoided.

Level of interest: An article of limited interest

Quality of written English: Acceptable

Statistical review: Yes, and I have assessed the statistics in my report.

Declaration of competing interests:

I declare that I have no competing interests