Reviewer's report

Title: Molecular analysis of non-culture CD133+ GBM cells revealed different signatures among high grade gliomas.

Version: 1 Date: 19 April 2010

Reviewer: Geoffrey John Pilkington

Reviewer's report:

The authors have addressed an important and topical issue of the association with the putative brain tumour stem cell "marker" CD133 with gene expression profiling when compared with CD133 negative tumour cell populations from glioblastoma tissues, rather than from in vitro cell lines.

Major Compulsory Revisions:

a) The technique to produce a single cell suspension for downstream analysis is not fully described. Any enzymatic treatment or mechanical disaggregation prior to filtration of biopsy material must be included.

b) CD133 remains controversial as both a brain tumour cancer stem cell marker and as an indicator of tumour initiating cells within the brain. For these reasons it would be better if the authors concentrated upon the possible functional significance of CD133 in glioblastoma in the light of their gene expression signatures for CD133+ve and CD133-ve populations.

c) No figures are given for the precise percentage of CD133+ve and CD133-ve cells in each case. These should be given as should full clinical details be given (tables are shown but without full legends).

d) the genetic signature information is confusing and sample numbers are too low to derive meaningful patterns emerging. Additional data would be of value.

Minor Essential Revisions

a) The use of "ex-vivo" rather than "non-culture" in both title and manuscript would add clarity.

b) The English language requires attention throughout.

c) Numerous typographical errors, mis-use of English language and lack of introduction of abbreviations (e.g., FISH) require rectification.

d) The CD133/2 (293C3) phycoerythrin-conjugated antibody does not recognise the same CD133 epitope as the commonly used AC133 antibody. Authors should comment on the possible consequences of studying different epitopes on putative cancer stem cell-like populations.
Discretionary Revisions

a) Use of the word "metastatic" in the last line of the abstract requires clarification. Do the authors mean locally invasive or have these glioblastomas demonstrated the rare propensity for dissemination elsewhere in the body?

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

**Quality of written English:** Not suitable for publication unless extensively edited

**Statistical review:** Yes, but I do not feel adequately qualified to assess the statistics.

**Declaration of competing interests:**

I declare that I have no competing interests