Reviewer’s report

Title: Analysis of folylpoly-gamma-glutamate synthetase gene expression in human B-precursor ALL and T-lineage ALL cells

Version: 2 Date: 15 March 2006

Reviewer: Joseph R Bertino

Reviewer’s report:

General Experiments are well done- interpretations need to be reconsidered

Major Compulsory Revisions (that the author must respond to before a decision on publication can be reached)

Fig 1B. It appears that the B-lineage cells have only the transcript that expresses the cytosolic form of FPGS except the NAlm 6 cell line that has both the the longer and the shorter transcript. CEM cells have the longer transcript that expresses the both the cytosolic and mitochondrial isoforms .

They do not emphasize this point, other than indicating in both lineages the transcription start site is the same. This difference alone may account for the difference in FPGS activities between B and T cells. This difference needs to be further investigated . Also, what does V stand for in this figure?

Page 10, line 13- it is really not correct to say that the pGL3588-2628 has no promoter /enhancer activity as compared to pGL2256. This construct has almost a 2 fold difference and NALM6 has two times more activity than CEM. The fold difference should be normalized to an empty vector.

Page 14 and fig4C. Inn lane 8, there is no significant supershift as compared to lane 3. Therefore the CBF-A subunit binding to the oligomer in the nuclear extract from CEM cells may be stronger than NALM6 extracts.

It would be more informative if titrations with different concentrations were performed, and the two complexes better resolved.

It should be noted that NFY-b not only reduced complex II formation, but also formed a new complex with a higher mobility that that of the other two complexes.Mutations in the NFY binding site in the oligomer should have been made to ensure that NFY was indeed binding to the CCAAT sequence. Left unanswered is the following:

B-cells contain the shorter transcript that encodes for the cytosolic enzyme. Is there a difference in message processing between the two lineages?

Minor Essential Revisions (such as missing labels on figures, or the wrong use of a term, which the author can be trusted to correct)

Page 13., Fig. 4b. Standard deviations would be helpful.

Page 20, fig.4c. The lanes should be numbered staring with the negative control, or the last sentence in the legend of fig. 4 corrected.

There are several typos. For example,page 2, line 17: led, not lead; line 20: significantly, not significant.

The trivial name, raltitrexed, rather than the trade name should be used.

Discretionary Revisions (which the author can choose to ignore)
What next?: Unable to decide on acceptance or rejection until the authors have responded to the major compulsory revisions

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

Quality of written English: Acceptable

Statistical review: No

Declaration of competing interests:

'I declare that I have no competing interests'