Author's response to reviews

Title: Combined mutations of asxl1, cbl, flt3, idh1, idh2, jak2, kras, npm1, nras, runx1, tet2 and wt1 genes in myelodysplastic syndromes and acute myeloid leukemias

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Author's response to reviews: see over
Marseille, June 15, 2010

Dear Editors,

Thank you very much for your help in the review of our manuscript (MS: 4308194333591616). We are also much grateful to the reviewers for their time and for their helpful comments.

Please find here a new version of the manuscript, modified as follows (new sentences are underlined):

1 - Answer to reviewer One:

We thank the reviewer for his praise of our study.
- We have modified the sentences concerning R140Q IDH2 mutation.
- We have modified table 1 (Glu960Ser is a frameshift mutation).
- Legend to Table 2 was present but for some reason probably got deleted in the submitted pdf.

2 - Answer to reviewer Two:

- We thank this reviewer for his positive appreciation of the results and we agree with him about the discussion. We eliminated the mention of initiator, selector and amplifier. We have also deleted the mention of 8 other class 1 genes and put less emphasis on the “missing culprits”.

However, we have maintained some of our propositions since they derive from the strict interpretation of the data. Our study is about gene combinations and cooperation and it may be legitimate to propose some hypotheses.

We have taken into account the fact that some mutations never occur together, the observed combinations, and the known or potential biological functions of the genes. We agree that the grouping of IDH and WT1 is flimsy but it is not without complete meaning either (we listed three reasons), based on the little we currently know of the function of these genes. RUNX1 could be seen as a tumor suppressor since mutations have been shown to truncate and inactivate the protein and the locus is often deleted or broken. This of course does not mean that a fusion with RUNX1 may not make it an oncogene. It was clear in our study that the RUNX1 mutations were strictly exclusive of TET2, which is considered as a tumor suppressor. We agree with the reviewer that RUNX1 does not behave like a classical Knudson-like tumor suppressor (although we found in a paired case one mutation of RUNX1 in MDS and two mutations in the corresponding AML). There may be more to it, as for the other genes, and we have mentioned that also.

Actually our model is just adding some complexity to the famous model of Gilliland and this is justified by the number of new genes identified recently. If we anticipate what deep sequencing will reveal we may even not be close.
We have hesitated to follow the advice of the reviewer to remove Figure 2. However, we decided to keep it because we feel that it may have its value. Besides, it derived from the data and cannot be contradicted by them. Moreover, that the model was deemed interesting by reviewer one has comforted us that it may be somehow useful.

- Basically a figure that would show individual mutations would reproduce the tables where we have put in gray each mutation.
- We have reworded the second sentence.
- We have now defined what we intend for a complex karyotype.
- We have now mentioned the exons we screened in the main text.

3 - Answer to reviewer Three:

We thank the reviewer for his positive appreciation of the study. Indeed, ours is the first to combine this very set of genes, one of which we have identified ourselves. We agree with the reviewer that data on fusion genes (and KIT) would have been highly interesting. Indeed, we had contemplated doing this analysis and include it in the manuscript, but are still in the process of collecting patients.

4 – Answer to reviewer Four:

We thank the reviewer for his positive appreciation of the study.

Finally we have added a recent reference (Boulwood et al., 2010 – ref 26)

Thank you again for this thorough evaluation of our work.

Sincerely,

Marie-Joelle Mozziconacci