Reviewer's report

Title: Cross platform analysis of methylation, miRNA and stem cell gene expression data in pediatric germ cell tumors highlights characteristic differences by tumor histology

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Reviewer: Daniel O Vidal

Reviewer's report:

The paper reports very interesting results about GCT biology, mainly regarding the potential role of stem cell factors and miRNAs in the disease.

Minor Essential Revisions

1. Section: Methods; Topic: Stem Cell Factor Expression - Authors should make clear how data normalization was carried out for PCR array analysis. Although the authors described the use of 5 endogenous housekeeping genes, how the normalization was performed? They used the average expression of the 5 endogenous genes, or only the data of the endogenous gene that showed the lowest variation, or an algorithm considering a set of best endogenous genes? This is important, because other reports can reproduce this data for comparison using different samples.

2. Section: Results - When I finished reading the results, I missed the methylation and genotyping data. It was not clear if the authors conducted the analysis in this paper or if the samples were the same used and analyzed in the reports previously published by the group and referenced in references 11 and 47. Authors should make clear if they used data from methylation and genotyping from the previously published works. If novel data was generated, I believe that it is important to include a topic in the results reporting both methylation and genotyping results.

3. Section: Results; Topic: Characteristics of the Study Samples - I suggest that authors describe the histologies present in the mixed tumors as well as its prevalence (%) in the tumor. It could be helpful to understand the clustering of these samples (Figure 2) in the stem cell factor expression analysis. I also note that in the figure 2, regarding the samples it was described 7 YSTs, 19 Teratomas, 7 germinomas and 6 mixed/other cases. It is discrepant from the description of the population study in this topic and need revision.

4. Section: Discussion - The 6 main hubs identified in the cross platform analysis are linked to stem cell factors or miRNAs. So, in page 15, lines 8-12, the authors presented a short paragraph about the lack of methylation differences appearing as important hubs in the cross-platform analysis. I understood that the authors also incorporated the genotyping data in the cross-platform analysis, so I think the authors should also included a short paragraph in the discussion about the
lack of hubs associated to the four molecular markers evaluated.

5. Figures - The number of the figures need revision. Figure 3 in the text was referenced as figure 2b; Figure 4, in the text as Figure 3.

6. Table 1 - I suggest, in the first column, the inclusion of (%) in the variable sex and location. For example, Male (%).

**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 that I have no competing interests