Reviewer’s report

Title: MicroRNA-182 Promotes Metastasis of Hepatocellular Carcinoma Through the Down-regulation of Metastasis Suppressor 1

Version: 1 Date: 4 February 2012

Reviewer: Elisa Giovannetti

Reviewer’s report:

The paper by Wang and colleagues investigated the role of miR-182 and its target gene MTSS1 in hepatocarcinoma.

Despite their findings are quite interesting, several key aspects need to be clarified before the manuscript may be suitable for publication.

The Abstract should be rewritten, adding the methods, including the number of samples, and the experimental procedures, as well as shifting the statement “one member of a miRNA cluster in a chromosomal locus (7q31-34) frequently amplified in HCC)” from the results to the background.

In the introduction the Authors should rephrase the statement “Although increasing data indicate miRNAs may play crucial roles in human tumorigenesis, it is still largely unknown for specific deregulated miRNAs involved in individual tumor and the corresponding mechanism”

The description of the clinical characteristics of the patients evaluated in this study should be improved.

Which is the median disease free survival (and 95% CI) in all the patients and in the patients with low and high expression levels of miR-182 (the figure 3 does not report the information about these values)?

How many patients did relapse (e.g. number of events) and which was the range of the follow-up?

Information on chemotherapy after surgery is also missing.

The Authors should perform a multivariate analysis including miR-182 expression levels (high vs. low) and at least the other clinical parameters reported in table 1.

The Authors should provide the information about the number of metastatic lesions where they detected a reduced expression of MTSS1 (again, this information is also missing in the bar-graph of the Figure 1G). Moreover they should specify whether they evaluated multiple lesions from the same patient or whether their data referred to metastasis from different patients.

Regarding the HCC samples used for the evaluation of miR-182 expression by PCR analysis, the Authors should report how they performed tumor micro- or macro-dissection, and report the percentage of tumor cells in their samples.
Regarding the IHC, the authors should report whether they only evaluated negative vs. positive stained samples or whether they could perform a more careful scoring of the staining of MTSS1.

As also reported by the authors, another important issue is “why miR-182 is up-regulated in HCC and other cancers”. Therefore, the Authors should check whether the chromosomal locus 7q31-34 was amplified in a number of their HCC samples.

The authors reported that "Target scan" showed that “MTSS1 is one important target of miR-182”, but they did not report other possible important targets that could be used also in comparison with previous studies.

For example, recent reports evaluated a novel function for miR-182 in the posttranscriptional regulation of BRCA1 expression and in DNA repair after chemotherapy (Moskwa and colleagues, Molecular Cell 2011)

Moreover, they cited the article on profiling of microRNA expression in hepatocellular carcinoma, which revealed microRNA-224 up-regulation, but they did not study also mir-224 as possible “control”.

Finally, they should check the targets of miR-182 at least also in another database, such as “microcosm” http://www.ebi.ac.uk/enright-srv/microcosm/htdocs/targets/v5/

Most data presented are obtained in 2 cell lines (Hep2 cells) with no functional data in the other hepatocellular in vitro models and particularly in vivo mouse models, which can be helpful to evaluate the metastatic role of miR-182 and MTSS1. The Authors should at least add some comments on the limitations of their study on these aspects.

Although partially reported in the legend to the figure 6, no appropriate controls are shown in the bar-graphs neither described in the methods/results for the analysis of the in vitro invasion assay.

There are several typos, such as "Lipifectamine", "metastatic leisions", that should be corrected

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

**Quality of written English:** Needs some language corrections before being published

**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