Reviewer's report

Title: The Role of p19 and p21 c-H-Ras Proteins and Mutants in miRNA Expression in Cancer and a Costello Syndrome cell model

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Reviewer: Tuoyu Geng

Reviewer's report:

In the manuscript titled ‘The Role of p19 and p21 c-H-Ras Proteins and Mutants in miRNA Expression in Cancer and a Costello Syndrome cell model’ by Roseli García-Cruz et al., authors describe force-overexpressed p19, p21 and their mutants regulate the expression of some miRNAs and the ratio of p19 to p21, and miR-206 mediates effect of p19 H-Ras on cell growth, suggesting p19 plays a role in RNA world. However, the authors may consider the following:

Major Compulsory Revisions
1. The authors need to examine whether p19 and its mutants regulate expression of miRNAs at basal level.
2. The authors provide some evidence indicating miR-206 regulate cell growth. It is better to provide data showing which targeted gene mediates the effect of miR-206. At least, the potential targeted gene should be discussed in the manuscript so the readers could logically understand how miR-206 regulates cell growth.
3. The context where p19 or its mutants are upregulated should be provided. This information provides the logic for why overexpression of p19 or its mutants was applied in this study. Is the expression of p19 and its mutants elevated in cancer or Costello Syndrome? If not, it is better to remove cancer and Costello Syndrome from the title of the manuscript, otherwise, regulation of miRNAs by p19 should be performed without overexpression of p19 and its mutants.
4. The criteria for selection of candidate miRNA regulated by p19 should be provided in the manuscript. Please provide this for why those candidate miRNAs rather than other miRNAs were selected from microarray analysis?
5. It seems that validation of microarray data by real-time PCR was only based on the expression of miR-206. Validation of more miRNAs is needed.
6. In Figure 2, as E4A-E4B was used to determine total H-RAS including p19, so how to distinguish the endogenous p19 from exogenous p19?
7. The authors should clearly state the limitations of this study.

Minor Essential Revisions
1. In Figure 1, it is better to use histogram to present the expression of selected miRNAs. The standard deviation or error should be presented with each mean value. The cells used in this assay should be indicated in the legend. For G1
cells, there was not much difference between GFP-p19 and GFP-p19+anti-miR-206, why is it still labeled as statistically significant (p<0.001)?

2. In the Results section, the subheading ‘p19:p21 splicing ratio alters miRNAs expression and cell growth’ is not properly stated, as the splicing ratio was only inferred from the observations that overexpression of p19 or p21 regulated the splicing ratio.

3. In Figure 5, there’s no statistical significance indicated in this figure.

4. There are many typos in the manuscript that needs to be carefully corrected.

5. Abbreviations should be spelled out when used in the text for the first time.

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

**Quality of written English:** Acceptable

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