Peer Review File

Reviewer A

1. Line 390 miR361-3p is error.

Reply: Thank you for your comment, we check the manuscript, have correct the miR-3613-5p.

2. Introduction and discussion states HCC has "high degree malignancy" and "extremely poor prognosis". Correct these details. 5 yr survival is good if a patient gets some treatment, and survival is improving with immunotherapy and adjuvant treatments.

Reply: Thank you for your comment, we change to the advanced HCC. See page 3, line 64.

3. I find it paradoxical. KMO is bad. HCC has less KMO. Then why is HCC prognosis bad? Please explain this.

Reply: Thank you for your comment. KMO is low expression in the HCC, further by the KM plotter, we find the low expression KMO with bad OS; finally, by the cell experiment, we verified that knockdown KMO in HCC cells promote the cell proliferation, then say KMO is bad.

4. You had so many details in method section. No money was needed? How come no funding for this study?

Reply: Thank you for your comment, these are written in order to clarify the article.

Reviewer B

The authors introduced the kynurenine 3-monooxygenase (KMO) as the novel biomarker to promote the tumor development and progression in hepatocellular carcinoma (HCC). They found that the expression of KMO is decreased in HCC compared to non-tumor liver tissue, and hsa-miR-3613-5p was found to be highly expressed in HCC cells and could negatively regulate the expression of KMO. They conclude that KMO plays an important role in the early diagnosis, prognosis, occurrence, and development of liver cancer, and may target miR-3613-5p to function. The paper is unique and well written, but I am afraid such conclusion is too much, because the actual mechanism and contribution of KMO to HCC progression has not been fully explained yet. Also, many hepatologists might be interested in the relationship between these markers and previous well-known risk factors such as tumor markers (AFP(L3), PIVKA-II), tumor differentiation, vascular invasion, and so on. It should be better to discuss them even with hypothesis.

1. There have been numerous well-known and convenient markers which are related to HCC progression, such as tumor markers, differentiation, vascular invasion, and so on. What do the authors regard the relationship between KMO and such markers? Also, the tumor stage and treatment should be shown as the patient characteristics.

Reply: Thank you for your comment, in the introduction section we declare why we choose the KMO. See page 4, line 92-101.

2. The expression of miRNA are written two types as "hsa"-miRNA and "has"-miRNA throughout the manuscript. Which is correct?
Reply: Thank you for your comment, hsa-miR is correct, we will correct it in the manuscript. Page 10, line 301-320, line 358.

Reviewer C

HCC represents an unmet medical need. The authors describe the role of KMO and its signaling network in HCC cell lines in vitro.
Overall, the role of KMO in HCC has been described previously by other authors (first publication from 1963) so this is not a novel finding. The authors confirmed KMO by an in silico approach and then in a very small subset of cell lines in vitro only (other authors have already shown the functional role of KMO in in vivo models, too).

The novelty of the paper is in identification of miR-3613-5p as a downstream mediators. Yet, such findings need be taken carefully as the can sometimes not be confirmed in different experimental settings or when looking into more complex conditions or human data sets. Therefore, functional validation of this miRNA is needed and additional data from human samples/data sets is needed to provide a meaningful novel aspect to this paper. Otherwise, the paper is largely descriptive and recapitulates what is already known on KMO.
Reply: Thank you for your comment, we will do the experiment as you advised in the future study.

Reviewer D

1. The below sentence is grammar mistake. And "extremely poor prognosis" cannot be used for HCC.

32 **Background:** Hepatocellular carcinoma (HCC) one of the most common cancers and
33 an important medical problem with extremely poor prognosis. The role of messenger

Reply: Thank you for your comment,we corrected it to Hepatocellular carcinoma (HCC) is one of the most common cancers and an important medical problem with poor prognosis .

2. Since you obtained genes from GSE101728 and GSE88839 (samples from patients), please add the statement "The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013)." **in both the "Methods" section of Main Text and the "Ethical Statement" section of Footnote.**
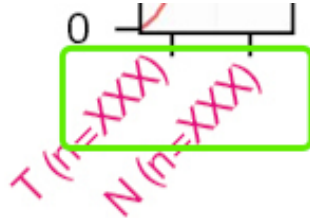Reply: Thank you for your comment, we have added.

2. Figure 3:

Figure 3 is not clear enough. Please resubmit it in higher resolution.

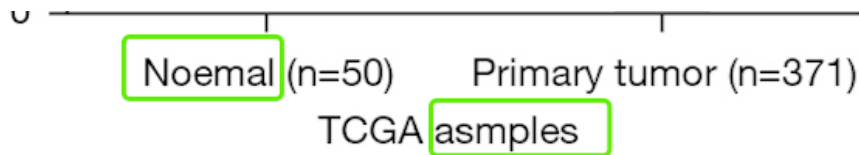Reply: Thank you for your comment,we have corrected as you said.

4 Figure 4:

1) Please check whether the numbers should be completed.

Reply: Thank you for your comment, we have completed.

2) There are spelling mistakes. Please revise.

Reply: Thank you for your comment, we have corrected.

5. Figure 5:

The below word should be "TargetScan".

Reply: Thank you for your comment, we have corrected.

6. Figure 6:

There are no A1-4 parts in Figure 6, but A1-4 were mentioned in the text. Please unify.

301 was applied to analyze their association with OS of the hub genes (Figure 6). We

302 found that low KMO, hsa-miR-302c-5p, hsa-miR-302f, and hsa-miR-6793-3p

303 expression was associated with a shorter OS in patients with HCC (Figure 6A2-4;

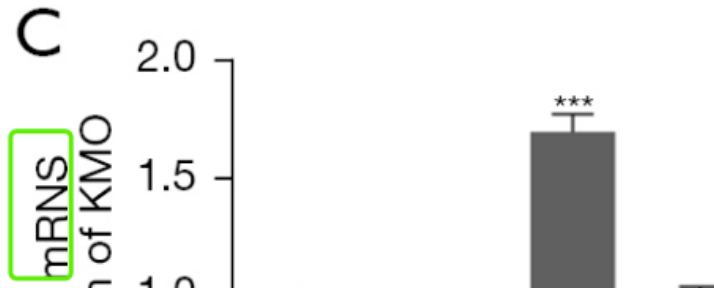Reply: Thank you for your comment, we have corrected.

7. Figure 8:

1) Please indicate the staining method and the scale bar in Figure 8D-E legend.

2) Please indicate the full name of "OD", "Si", "PE-H", "FITC-H" in the legend.

Reply: Thank you for your comment, we have corrected.

8. Figure 9:

There is spelling mistake "mRNS".

9. There are two reference lists in your paper. Please remove the unnecessary one.