Reviewer's report

Title: Downregulation of Cyclophilin A by siRNA diminishes non-small cell lung cancer cell growth and metastasis via the regulation of matrix metalloproteinase 9

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Reviewer: Giovanni Luca Beretta

Reviewer's report:

In the present manuscript Zhe Quian and coworkers studied the role of the peptidyl-prolyl isomerase Cyclophilin A (CypA) enzyme in non-small cell lung cancer cell (NSCLC) growth and metastasis. Using siRNA directed to CypA, they demonstrated the involvement of the enzyme in growth and metastasis of NSCLC and reported that CypA increased NSCLC cell invasion by regulating the activity of secreted matrix metalloproteinase 9 (MMP9).

Although the experiments are well performed and the manuscript well written the paper suffers of some imperfections.

Major Compulsory Revisions

1) To dissect the role of the mass protein and enzyme activity it would be interesting to evaluate the effect of small molecule/s targeting the CypA activity. In this regard some inhibitors have been already published by Ni and coworkers (Ni S et al. Discovering potent small molecule inhibitors of cyclophilin A using de novo drug design approach J Med Chem. 2009 Sep 10;52(17):5295-8) and one of these compound (239836 Cyclophilin A Inhibitor) is commercially available. It should be interesting to evaluate the cell growth, motility, invasion and, most importantly, MMP9 activity of NSCLC cell line/s exposed to this inhibitor.

2) The authors refer to different growth-related signaling molecules that are stimulated by CypA in cancer (e.g., ERK1/2, Jak2, p38, Stat5) but they did not evaluate possible changes (expression levels and phosphorylation) of these actors after CypA silencing.

3) In the Discussion section the authors reported that CypA accelerates cell growth by stimulating cell proliferation, tumorigenesis, and metabolism, and by inhibiting apoptosis. However, they do not evaluate apoptosis in CypA silenced cells. What is the effect of CypA silencing on apoptosis in NSCLC cell line/s.

4) The authors report that CypA enhances the activity of secreted MMP9 but they did not elucidate the mechanism/s. In my opinion this is the most important conclusion of the paper that requires explanation. Again, is there a direct interaction between CypA and MMP9? A detailed analysis of the protein sequence of MMP9 could help in defining this point.

Minor Essential Revisions
1) Abstract, conclusions: change the sentence “CypA was correlated with decreased NSCLC cell tumorigenesis and metastasis” to “The suppression of CypA expression was correlated with decreased NSCLC cell tumorigenesis and metastasis”.

2) Figure legends of figure 3 and 4: change + SEM to ± SEM.

Discretionary Revisions
None

**Level of interest:** An article whose findings are important to those with closely related research interests

**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