Author's response to reviews

Title: 2-Ethoxystypandrone, A Novel Small-Molecule STAT3 Signaling Inhibitor from Polygonum Cuspidatum, Inhibits Cell Growth and Induces Apoptosis of HCC cells and HCC Cancer Stem Cells

Authors:

Wuguo Li (liwuguo@qq.com)
Qing Zhang (qzhang@simm.ac.cn)
Kaotan Chen (2446630@qq.com)
Zhenhua Sima (781775818@qq.com)
Jingli Liu (jlliu@shapb.com)
Qiang Yu (qyu@sibs.ac.cn)
Jiawei LIU (jiawei.liu@ymail.com)

Version: 4 Date: 20 Dec 2018

Author's response to reviews:

Dear Editor,

We are very grateful to Editor and Reviewers for their careful and meticulous reading of our revised manuscript. Their detailed and constructive comments have been very helpful in improving the quality of the manuscript.

We have made careful revisions according to their comments and suggestions. The changes have been marked in blue in the text and our detailed responses to the specific comments are described as follows. We hope that these revisions improve the manuscript such that you and Reviewers will agree that our work is suitable to be published in BMC Complementary and Alternative Medicine.

We look forward to your decision.

Yours sincerely,
Revisions

Manuscript ID:

Title: 2-Ethoxystypandrone, A Novel Small-Molecule STAT3 Signaling Inhibitor from Polygonum Cuspidatum, Inhibit Cell Growth and Induce Apoptosis of HCC cells and HCC Cancer Stem Cells

Authors:

Wuguo Li (liwuguo@qq.com)
Qing Zhang (qzhang@simm.ac.cn)
Kaotan Chen (2446630@qq.com)
Zhenhua Sima (781775818@qq.com)
Jingli Liu (jlliu@shapb.com)
Qiang Yu (qyu@sibs.ac.cn)
Jiawei Liu (jiawei.liu@ymail.com)

Editor Comments:

1. In discussion, page 21, "The our previous preliminary structure-activity relationship..." please remove "the"

Answer: We thank Editor for his careful reading of the revised manuscript. We have corrected it in the revised manuscript. Please see page 21 line 13 in the manuscript.
2. In page 21, "2-Ethoxystypandrone (1) as a STAT3 signaling inhibitor might be considered as a potential STAT3 signaling inhibitor for developing an anticancer agent moleculely targeting CSCs in HCC” please rephrase as sentence is weird

Answer: We agree with Editor’s comments, and we have reorganized this sentence as following:

“2-Ethoxystypandrone (1) might be considered as a potential STAT3 signaling inhibitor for developing an anticancer agent moleculely targeting CSCs in HCC.”

Please see page 21, lines 17-19 in the Discussion Section.

3. In discussion, you mention that "2-Ethoxystypandrone (1) might not only target STAT3 signaling pathway but also bind to other apoptosis-related cellular signaling pathways such as PARP,14 Caspases, PI3K/AKT and RAS/ERK" , please provide evidence for this statement.

Answer: We are very grateful to Editor for his serious and meticulous reading of our revised paper. We have reorganized and rewritten this sentence as follows:

“2-Ethoxystypandrone (1) could be able to block STAT3 activity either via directly targeting STAT3 protein or by binding to the upstream receptor tyrosine kinases such as JAKs in the STAT3 signaling pathway.”

Please see page 24, lines 20-22 in the Discussion Section.

4. In conclusion, page 25, "Therefore, it is worth of further consideration..." consider rephrase

Answer: We thank Editor for his careful reading and for his valuable suggestions. We have reorganized this sentence as following:

“Further investigations are underway in our laboratory to develop more efficient derivatives, identify its precise target proteins and finally determine clinical potentials of 2-ethoxystypandrone (1) as a cancer stemness STAT3 signaling inhibitor.”

Please see page 25 lines 11-14.

5. Authors should provide clearer Western blot photo for Figure 2D (p-Stat3 background too strong, GAPDH loading is not consistent, especially 100uM)

Answer: According to Editor’s suggestion, we have redone Western Blot experiment to improve the quality of the Blots. Please see Figure 2D.
Reviewer reports:

Tomoharu KUBOYAMA (Reviewer 1): Please mention some comments in the revised manuscript about the time lag, because the cells were not dead at 6.5 h after the treatment in Fig. 2B, but they were dead at 24 h after the treatment in Fig. 3A. At 6.5 h after the treatment, STAT3 phosphorylation would be enoughly inhibited and PARP would be already cleaved.

Answer: Authors thank Reviewer for his constructive suggestions. We have added some sentences describe the time lags, which make them easier to understand. Please see page 16 lines 20-24, page 17 lines 12-16, lines 23-25, and page 18 lines 1-2 and lines 5-8 in Results Section.