Manuscript NO: 49148

Title: Circular RNA PIP5K1A promotes colon cancer development through inhibiting miR-1273a

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Dear editor,

Thank you for your letter with regard to our manuscript (49148). We greatly appreciate for your time in editing our manuscript and the valuable comments and suggestions from reviewers. Accordingly, we have revised the manuscript in track changes mode in MS word. The point-by-point answers to the comments are listed as below. We would like to re-submit this revised manuscript to ***World Journal of Gastroenterology*** and hope it is suitable for publication in the journal.

Yours sincerely,

Jiao-Zhen Xu

Reviewer #1

(Comment) The writing needs editing; it is full of errors. My comment concerns the RT-PCR. Is there any data that demonstrate whether the RNA used is DNA free? Trizol isolation is not necessarily going to get rid of all the DNA, and I do not see any indication that any subsequent DNase step was performed. Therefore, is there any negative control, such as doing the RT-PCR but skipping the RT step? If the RNA is really DNA free, then performing PCR directly on the RNA should not yield a band. Can the authors comment on this, at least? If they have any of these RNA samples left over, showing that no product is formed without the RT step would be helpful.

(Response) Thanks for your comment. We are very sorry for uncomplete RT-PCR description. RNA concentration and purity had been measured by UV spectrophotometer, and RNA with A260/A280 ratios of 1.8–2.0 was considered as high-quality RNA without protein and DNA contamination. In addition, RT step has been supplemented. According to your suggestion, we have supplemented RT-PCR description as following: "Total RNA was obtained by Trizol (Invitrogen) according to manufacturer's instruction. RNA concentration and purity were measured by UV spectrophotometer (BD, Franklin Lakes, NJ, USA), and RNA with A260/A280 ratios of 1.8–2.0 was considered as high-quality RNA without protein and DNA contamination. Then high-quality RNA was reverse transcribed into complementary DNA (cDNA) with a Reverse Transcription Kit (TaKaRa, Dalian, China). PCR amplification was performed by a SYBR Premix Ex Taq TM II (Takara). The primers sequences are listed in Table 1. The PCR program was: 95°C for 3 min, 40 cycles of 95°C for 10 s and 59°C for 20 s, using the ABI Stepone plus Real-time PCR system (Applied Biosystems, Foster City, CA, USA). Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) and U6 were served as internal control of measuring circPIP5K1A and PIP5K1A levels. Data were analyzed with $2^{-\Delta\Delta Ct}$ method." In addition, we have made a careful check all through the paper and substantially improved language both in spelling and grammar by a native English speaker. The "Non-Native Speakers of English Editing Certificate" has been provided as an attachment.

Reviewer #2

(Comment) This is an interesting manuscript investigating the role of circPIP5K1A in colon cancer and its effects on modulation of cancer development. Besides some minor grammatical errors, this is a well written article with a robust outcome. Minor language corrections should be made by a native English speaker.

(Response) Thanks for your comment. We have made a careful check all through the paper and substantially improved language both in spelling and grammar by a native English speaker. The “Non-Native Speakers of English Editing Certificate” has been provided as an attachment.