

ANSWERING REVIEWERS



Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 7581-revised.doc).

Title: Radiobiological characteristics of cancer stem cells from esophageal cancer cell line

Author: Jian-lin Wang, Jing-ping Yu, Zhi-qiang Sun, Su-Ping Sun

Name of Journal: World Journal of Gastroenterology

ESPS Manuscript NO: 11091

The manuscript has been improved according to the suggestions of reviewers:

(1) In their article "A Study on Radiobiological Characteristics of Cancer Stem Cells Derived from Esophageal Cancer Cell Lines" the authors characterize sphere formation and radio sensitivity of two esophageal cancer cell lines. The article is well written and conclusive, however, some minor points need to be addressed. In particular, the authors should mention the cancer stem cell marker CD44 and CD271 in their part 1 (Introduction). In Figure 1, size bars are missing in all pictures. Error bars seem to be missing from Fig 1D. Analysis of BMI1 and SOX2 should be performed using real time PCR, or at least semi-quantitative PCR. Labels are missing from Fig.3B, it is not clear which one is the parental vs the sphere cell line, and which cell line represents which graph. Similarly, labels are missing from Fig.4A, which one is the parental or sphere cell line? Fig.4B does not correlate with Fig.4A. In Fig.4A it seems that around 2% of KYSE150 parental cells are positive for CD44 (0Gy), but on the graph in 4B it says 40%, which is not supported by the FACS data. Similarly, for TE1 cells at 0Gy 16.9% of cells are positive for both CD44 and CD271, but on the graph it says less than 1%. Fig.4B needs to be re-analysed completely. In Fig.4C it is not clear what "a" is supposed to mean. Apart from that a good article, short but focused..

(2) This is an interesting study of the radiobiology of sorted cells from esophagus, human esophageal cancer cell lines KYSE-150 and TE-1 obtained from a hospital in China. No fresh human esophagus cancer specimens were tested. No fresh human stem cell populations were studied. The studies looked at self-renewal of sphere-type cultures and looked at cell cycle and cell surface markers. The radiobiology is not complete. The authors should do full radiation dose response curves and calculate a D0 and ?. They should look at the textbook by Hall and Gaccia to look for classic radiobiological properties. Figure 1 shows biological properties of the sphere. Figure 2 shows radiation dose on sphere formation. The survival fraction is shown, but D0 and ? should be calculated. The authors should represent the data as either linear quadratic or single hit, multi-hit statistical evaluation. Figure 3 shows influence of irradiation on cell cycle changes, and this needs to be evaluated more completely and described in the text. Figure 4 looks at expression of the stem cell marker genes on the cells in culture at various stages. The results are interesting and provocative and with attention to appropriate radiobiological parameters this will be a useful contribution to the literature. The discussion should be revised in view of the new data.

1 Format has been updated

2 Revision has been made according to the suggestions of the reviewer

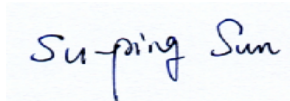
3 Thanks for your carefully review and valuable suggestions, and we would like to give some interpretation about the questions you proposed. CD44 and CD271 were mentioned in the introductory part. the charts were made changes according to your suggestion. Radiobiology indicators

D0 , Dq and SF2gy data were added in the concluding part of the article. If there is problem please let us know. This study mainly focused on the Cancer Stem Cells Derived from Esophageal Cancer Cell Lines, as to the relation of radiobiological Characteristics is another worth research point, and maybe we would do some work in our future study.

References and typesetting were corrected

Thank you again for publishing our manuscript in the *World Journal of Gastroenterology*.

Sincerely yours,

A handwritten signature in blue ink that reads "Sun SP". The signature is written in a cursive, flowing style.

Sun SP, ,PhD

Department of Radiation Oncology

The Affiliated Hospital of Nanjing Medical University, Changzhou Second People's Hospital,
Changzhou 213003, China

Tel.: +86 519 88118723; fax: +86 519 88115560

Email: ssp56@126.com