

ANSWERING REVIEWERS

November 6, 2013

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name:5764-review.doc).



Title: Cancer stem cell markers correlate with early recurrence and survival in hepatocellular carcinoma

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Name of Journal: *World Journal of Gastroenterology*

ESPS Manuscript NO: 5764

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

1 Format has been updated

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

(1) Reviewer 00503516

Comment 1: As CD133 is a transmembrane glycoprotein (page 7 line 5 from top) it is unclear why the authors detect it in the cytoplasm. The same problem exists also for the marker CD90 which, although being a membrane protein, is detected by the authors exclusively in the cytoplasm (page 7 line 8 from top).

Response: Any protein having different subcellular localization may have a specific function, although the physiological role of CD133 (or CD90) is yet unknown. Absence of membranous CD133 (or CD90) expression, as observed in HCCs in this study, may probably indicate the accumulation of truncated CD133 (or CD90) protein, internalization of CD133 (or CD90) protein on membranes that may result in rapid degradation^[1]. This phenomenon has been already observed for the epidermal growth factor receptor in cultured cells^[2].

[1] Sasaki A, Kamiyama T, Yokoo H, Nakanishi K, Kubota K, Haga H, Matsushita M, Ozaki M, Matsuno Y, Todo S. Cytoplasmic expression of CD133 is an important risk factor for overall survival in hepatocellular carcinoma. *Oncol Rep* 2010 2010-08-01; 24(2): 537-546.

[2] Beguinot L, Lyall RM, Willingham MC, Pastan I. Down-regulation of the epidermal growth factor receptor in KB cells is due to receptor internalization and subsequent degradation in lysosomes. *Proceedings of the National Academy of Sciences* 1984; 81(8): 2384-2388.

Comment 2: Substitute the word "role" with "correlation" in the text.

Response: This has been revised in the paper.

(1) **Reviewer 00068250**

Comment 1: There is bias in the baseline data between the two groups. More patients with liver cirrhosis and lower platelet counts are found in the ER group. As cirrhosis is an important factor influencing overall survival of HCC patients after hepatectomy, the authors need to explain in the discussion section how to manage this bias. Propensity-score matching will cover this bias

Response: In this study, we aimed to analysis the expression of CD133, CD90, EpCAM in patients and to search for association with early recurrence. So clinicopathological characteristics related to tumor recurrence should be balanced between the ER and NER groups. Then we used propensity-score matching to generate balanced pairs of ER and NER patients. The explanations have been added into the results and discussion sections.

Comment 2: Some abbreviations in the abstract are presented without complete phrase.

Response: These have been added into the paper.

Comment 3: Edmondson grade and tumor size are significantly associated with survival time of HCC patients, which is widely known conclusion. Log-rank analysis of them is unnecessary

Response: This part has been deleted in the paper.

Comment 4: CD90 expression is significantly associated with higher Edmondson grades, and this result should be explained in detail.

Response: These have been added into the paper.

3 References and typesetting were corrected

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

Sincerely yours,

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