

Dear reviewers,

Thank you for your valuable comments on our paper which we have amended according to your suggestions. And we reply to the specific comments below.

Reviewer #1

Comment: This paper is best defined as a "case series" rather than a "pilot study". Please change the title accordingly.

Response: Thank you for your advice. And we have modified the title.

Comment: In the text it should be said that the stem cells were "infused" rather than "transfused".

Response: Modified.

Comment: For the sake of consistency, either "stem cells injection" or "bone marrow injection" should be written in the text. How was the aspirated bone marrow treated and harvested? Was it used as a whole for injection, just right after aspiration? On the other hand, was it somehow treated and preserved?

Response: Thank you for your advice and questions. The autologous bone marrow was performed under local anesthesia, and the right posterior superior iliac spine was selected as the puncture site. About 30 ml of the patients' bone marrow was collected in a syringe containing 10 ml of heparin saline (62.5 U/ml), which was used as a whole for injection, just right after aspiration.

Comment: Endoscopic procedure. You mentioned that 30 ml of bone marrow + 10 ml of heparin were injected at a 1ml/min velocity. This means that 40 mins were required for the whole injection. Can you confirm? How was the infusion rate determined beforehand? How was it possible to maintain the needle stability inside the PV for such a long time?

Response: Thank you for your questions. It usually takes about 30-40 minutes for the whole injection, and the injection is administered under the guidance of endoscopic ultrasound, which helps us to maintain the the infusion rate and needle stability.

Comment: Patients. To understand the results, concomitant treatments should be mentioned. Were the patients under antiviral therapy against hepatitis B? Were diuretics either started or adjusted after the diagnosis of ascites?

Response: Thank you for your advice and questions. All patients were under antiviral therapy against hepatitis B, and diuretics were started after the diagnosis of ascites.

Comment: Results. It seems unfair to talk about significant improvement in a study with 5 patients. I think this can be referred to as a feasibility and safety study, but I would refrain from any evaluation of statistical significance.

Response: Thank you for your advice. And we modified the expression of statistical analyses.

Comment: How was ascites measured in mm?

Response: Thank you for your questions. The depth of ascites was evaluated by abdominal ultrasound.

Comment: The sentence "Other indexes showed a slight fluctuation during the follow-up period" is too generic and should be removed. The results of elastography are not reported at follow up.

Response: Modified.

Comment: Abstract. "Compensated cirrhosis" instead of "compensatory".

Response: Modified.

Comment: Endoscopic procedure. Use "echoendoscope" and not "ultrasonic gastroscope". Use "stylet" and not "probe".

Response: Modified.

Comment: Discussion. "Autologous" and not "autogenous".

Response: Modified.

Reviewer #2

Comment: Logical flow to the bone marrow transplantation with EUS was not clear.

Response: Thank you for your advice. We have modified the part of "Endoscopic procedure" to explain the bone marrow transplantation with EUS.

Comment: Did the authors have any proof-of-concept data? Or was the technique performed as a brainstorm?

Response: Thank you for your questions. The stem cell therapy has been extensively studied as a promising treatment for DLC, and EUS offers a potential access to the portal vein, through which the stem cells can be precisely delivered to the liver. The results of this study can be considered to be the proof-of-concept data.

Comment: How would the authors speculate where the transplanted cells reside? Or did they disappear?

Response: Thank you for your questions. We speculate that the transplanted bone marrow resided in the liver, and we will design animal experiment to prove this hypothesis. We have added this question into the part of limitations.

Comment: The authors counted the number of CD34 positive cells. How was "CD34" featured?

Response: Thank you for your questions. The CD34 positive cells are bone marrow cells specialized in forming different blood elements and being stem cells, which have the character of plasticity and can change into hepatocytes.

Comment: The authors probably expected that the transplanted cells resided in the liver. If so, how would the authors obtain evidence of homing of the transplanted cells in liver?  
Response: Thank you for your questions. As described above, this is a limitation of this study, and we will design animal experiment to further prove this hypothesis.

Comment: Bone marrow transplantation to liver insufficiency has been reported. For example, infusion into peripheral vein. Did the authors refer the literatures? What was the significance of the authors' study as compared to the previous studies?

Response: Thank you for your questions. We have referred the literatures of bone marrow transplantation into peripheral vein to liver insufficiency. Bone marrow injection by EUS-guided intraportal FNI offers an accurate targetability, and this study proved that the use of EUS-guided fine needle injection for intraportal delivery of bone marrowstem cells was feasible, safe and appeared effective in patients with DLC.

Comment: Where were the samples obtained as bone marrow aspiration? Were the bone marrow samples subjected to transplantation immediately after aspiration of bone marrow? If so, bone marrow sampling performed in the same room as the EUS transplantation? Was the flowcytometry performed after the transplantation?

Response: Thank you for your questions. The aspiration and transplantation of bone marrow were performed in the sterile operating room, and the bone marrow samples were subjected to transplantation immediately after aspiration. And the flowcytometry was performed to count the total number of nucleated cells and the proportion of CD34+ cells, while the bone marrow was used as a whole for injection.

Comment: The enrolled patients had ascites. Did the ascites cause complications? For example, bleeding after the EUS guided transplantation.

Response: Thank you for your questions. However, no complications such as hemorrhage, hematoma, perforation, fever, pain, infection, acute liver failure, hepatic encephalopathy or bleeding were observed during or after the procedure.

Comment: Figure 3. What is "ascites (mm)? Were there any possibilities that measuring methods affected that (mm)?

Response: Thank you for your questions. The depth of ascites was evaluated by abdominal ultrasound.

Comment: Figure 4. A showed slight change during the observation period. In Figure 4B, percentage of Child-Pugh Scores seemed change dramatically. For example, C disappeared in post 1m, 6m, and 12m. Child-Pugh A occupied majority in post 3m and 6m. How would the authors speculate the trends of the data?

Response: Thank you for your questions. Figure 4. A showed the Child-Pugh scores, and Figure 4. B showed the Child-Pugh classes. Considering only 5 patients included in this study, slight change of Child-Pugh scores can lead to huge change of Child-Pugh classes. And the results of Child-Pugh classes showed that EUS-Guided intraportal

injection of autologous bone marrow could alleviate severity of DLC.

Comment: Table 1. What is “ascites (mm)”?

Response: Thank you for your questions. It means the depth of ascites, which was evaluated by abdominal ultrasound.

Thanks again for the above comment and suggestion.