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# Role of HGF for reprogramming human liver progenitor cells: Non-essential but stimulative supplement

*To the Editor:*

With great interest, we read the article written by Kim *et al.* in a recent issue of *Journal of Hepatology*.[1] The authors developed a successful HAC culture system for reprogramming mature human hepatocytes into bipotential progenitor cells treated with 2 small molecules A83-01 and CHIR99021 (AC) in combination with hepatocyte growth factor (HGF). Their chemically derived human hepatocyte progenitors could sustain themselves as a population of progenitor cells over a long period while maintaining chromosomal stability and the capacity to differentiate into functional hepatocytes and biliary epithelial cells *in vitro* and *in vivo*.

Kim and colleagues showed that the use of HGF proved to be an essential determinant of the fate conversion process. In their initial work, the authors have adopted the methodology recently described by Katsuda *et al.*[2,3] They confirmed that a cocktail of 3 small chemicals, Y27632, A83-01, and CHIR99021 (YAC), which was very effective in reprogramming mouse and rat hepatocytes, did not support the conversion process in human hepatocytes. The authors observed that YAC-treated human hepatocytes rapidly died off without proliferation. In our laboratory, however, we verified the validity of the YAC cocktail for conversion of human hepatocytes into liver progenitor-like cells. In our identical culture system of YAC initiated by Katsuda *et al.*, the YAC-treated human hepatocytes are slowly converted into stemness state cells with a high ratio of nucleus to cytoplasm. Unlike the rapid expansion of progenitor cells in Kim's HAC culture system, the YAC culture system, without supplemental HGF, takes about 3 to 4 weeks to convert human hepatocytes into the progenitor-like cells expressing high levels of stem cell genes. Our data demonstrate the validity of the YAC cocktail without supplemental HGF for conversion of human hepatocytes into liver progenitor-like cells. Based on the above, we believe that, for reprogramming human liver progenitor cells, exogenous HGF is a non-essential but stimulative supplement or factor, with functions in proliferation and stem cell expansion, but not reprogramming.[4]

Secondly, in the YAC culture system, we observed fibroblast-like cells also proliferated during the reversion process of human hepatocytes but not rat hepatocytes. In our YAC culture system, no FBS were added, however, in Kim's culture system, the authors defined HAC culture system contained 1% of FBS, a

fact which was believed to enhance the cell attachment.[5,6] Hence, we are curious about whether this phenomenon of fibroblast-like cells is also observed in the HAC culture system.

Thirdly, the authors used 6 cases of mature human hepatocytes isolated from healthy and diseased donor livers. To our knowledge, we only isolated the mature hepatocytes from the normal parts of the diseased liver, which are termed non-tumor liver. Strictly speaking, we only use the hepatocytes either from the healthy liver or the non-tumor part of a diseased liver. This is a vital point to consider when interpreting this data, otherwise one may be misled into believing that the culture system can also reprogram the diseased hepatocytes (tumor cells *etc.*) into progenitor cells.

We applaud Kim and colleagues for providing the successful HAC culture system for reprogramming human liver progenitor cells with a rapid expansion rate. It not only overcomes the low conversion efficiency but also reported no other fibroblast-like cells in YAC culture system. Their important work also advances our understanding of the chemical reprogramming of human progenitor cells *in vitro*, offering a different view for the optimization of culture systems in the future.

## Financial support

The authors received no financial support to produce this manuscript.

## Conflict of interest

The authors declare no conflicts of interest that pertain to this work.

Please refer to the accompanying ICMJE disclosure forms for further details.

## Authors' contributions

Conception, design and writing: YH and SE. Review and revision of the manuscript: YH, YS, TM, TH, WG and SE. Study supervision: SE.

## Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jhep.2019.03.017.

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# Reply to: "Role of HGF for reprogramming human liver progenitor cells: Non-essential but stimulative supplement"

*To the Editor:*

We thank Dr. Huang and colleagues for their interest and comments on our recent study, "Small molecule-mediated reprogramming of human hepatocytes into bipotent progenitor cells" published in the *Journal of Hepatology*.[1]

We learnt with great interest from the Dr. Huang's letter that the YAC cocktail (Y27632, A83-01, and CHIR99021) originally developed by Katsuda *et al.*[2] for reprogramming of rodent hepatocytes was also effective in human hepatocytes. The process of conversion of human hepatocytes into stemness state using the

*Received 8 April 2019; accepted 15 April 2019*