Enhanced homing of HGF-rAAV-transfected BMSCs and its recovery performance in chronic damaged liver rats with ultrasound in combination with microbubble

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Background, Motivation, and Objective:

Bone marrow mesenchymal stem cell (BMSCs) have demonstrated great potential in the cytotherapy of liver fibrosis and its end-stage outcome, cirrhosis. However, the insufficient homing efficiency and the undefined differentiation direction of BMSCs in injured liver limited its application value. Thus, we employed UTMD to promote the transfection of recombinant adeno-associated virus (rAAV) encoding hepatocyte growth factor into BMSCs. Meanwhile, we investigated the therapeutic effect of combined the transfected BMSCs with UTMD positioning radiation in rats' liver in this study.

Statement of Contribution/Methods:

We selected optimized UTMD parameters to mediate HGF-rAAV transfection of BMSCs, then the transfection efficiency was observed. Liver fibrosis model of rats was acquired via intraperitoneal injection of carbon tetrachloride (CCl4). Model rats were assigned into four groups: normal, CCl4, US+ HGF-rAAV-BMSCs and UTMD+HGF-rAAV-BMSCs. The number of BMSCs distributed in liver were quantified under fluorescence microscope at 2w, 4w after treatment. Furthermore, liver tissues hematoxylin-eosin (H- E) staining and van gieson (VG) staining were used to determine the morphological changes of damaged liver. And biochemical liver function indices were evaluated by a series of serum indicators, including alanine aminotransferase (ALT), aspartate aminotransferase (AST), total protein (TP), albumin (ALB) and alkaline phosphatase (ALP) at every 2 weeks for a total of twelve weeks. The degree of improvement in hepatic fibrosis was quantified by the following fibrosis index, collagen I, vimentin and α smooth muscle actin (α-SMA) using immunohistochemistry immuno-histochemistry and real time polymerase chain reaction (qPCR).

Results/Discussion:

The results showed that the US treatment at 1.0 W/cm^2 or 1.5 W/cm^2 for 30 s or 60 s conditions generated good hepatocyte directional differentiation capacity. In vivo experiment, the average fluorescence intensity of DAPI-labeled BMSCs homing to injured liver was the highest in the UTMD+ HGF-rAAV-BMSCs group at different time points. Among all the treated groups, histopathological analysis of the liver tissue in UTMD+ HGF-rAAV-BMSCs group improved optimally in the detection of various time points. In addition, in response to liver morphology improvement, the reduction of ALT, AST, TP, ALB and ALP gradually returned to normal levels in UTMD group. Compared with the other groups, the anti-fibrotic effect was the most visible in UTMD+HGF-rAAV-BMSCs group when combining the immunohistochemical and the genetic level of fibrosis indexes. Conclusion: The combination of UTMD with HGF-rAAV-transfected BMSCs effectively promoted the homing and stable differentiation of BMSCs in rats of chronic liver injury. The increased BMSCs maintained stable expression of HGF and then stimulated the proliferation of hepatocytes, accelerated reversal of liver fibrosis.

Key words: ultrasound, microbubble, stem cell transplantation, chronic liver injury