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Title: Characterisation and hepatocyte-like differentiation of mesenchymal stem cells derived from adipose tissue of immunodeficient mice

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Body: Background: Mesenchymal stem cells (MSCs) can differentiate into hepatocyte-like cells both in vitro and in vivo. These cells may represent a new resource for the treatment of distinct liver diseases, such as alpha-1 antitrypsin deficiency (AATD), a metabolic disease that has pathological consequences for the liver. Aims and objectives: To isolate MSCs from mouse adipose tissue and verify their potential for differentiation to explore their use for alleviating the shortage of liver transplantation donors to treat AATD. Methods: MSCs were isolated from immunodeficient mice and cultured to 90% confluency. After DNA demethylation, a differentiation medium was applied and cellular morphology was assessed by microscopy after 0, 7, 14 and 21 days. Flow cytometry was used to detect mesenchymal (CD13, CD29, CD44, CD105) and haematopoietic (CD34, CD45) cell surface markers to estimate the number of differentiated cells. Gene expression of the hepatocyte-specific markers transferrin, albumin, CK18, CD26 and CYP3A1 was measured by semi-quantitative reverse transcription polymerase chain reaction. Results: During differentiation, the morphology of MSCs changed from a spindle shape into a more polygonal aspect. Mesenchymal markers were expressed at each time point, whereas haematopoietic markers were hardly detectable after 21 days. The relative gene expression of the hepatic markers was increased at each time point. Conclusion: MSCs derived from mouse adipose tissue may differentiate into hepatocyte-like cells in vitro. Consequently, the potential therapeutic benefit of MSCs in a syngeneic mouse model of chronic AATD can now be analysed.