

In vitro transformation of peripheral monocytes to progenitor cells with putative hepatopoietic potential and their labeling with supraparamagnetic iron oxides for MR-analysis

Andreas Benesic¹, Frank Berger², Markus Bystron¹, Enrico de Toni¹, Veit Gülberg¹ and Alexander L. Gerbes¹

¹Medizinische Klinik und Poliklinik II, Klinikum Großhadern, Ludwig-Maximilians-Universität München, Marchioninstr. 15, 81377 München

²Institut für Klinische Radiologie, Klinikum Großhadern, Ludwig-Maximilians-Universität München, Marchioninstr. 15, 81377 München

Introduction:

Shortage in donor organs and side effects of immunosuppressive therapy necessitate the search for alternatives to liver transplantation. Considerable effort is focussed at cell based therapies. Transformed peripheral monocytes have been described as a possible source of hepatocyte-like cells.

Aim of this study was to establish the generation of peripheral blood monocyte-derived hepatocytoid cells and labeling of these cells with supraparamagnetic iron oxides (SPIO) to enable their further use and visualisation in transplantation experiments.

Methods:

Peripheral monocytes of healthy donors were isolated and incubated in presence of M-CSF, IL-3 and mercaptoethanol, followed by culture in the presence of FGF-4. Marker expression was investigated by FACS analysis. The specific enzyme activities of DPP IV, γ GT and ALT were determined and compared to HepG2 cells. Cells were labeled using Resovist® and MR-scanned in gelatin.

Results:

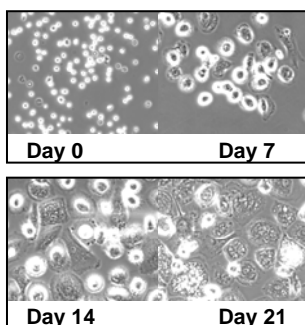


Figure 1: Morphologic changes of monocytes during the culture period (Mag X 32)

Marker-expression

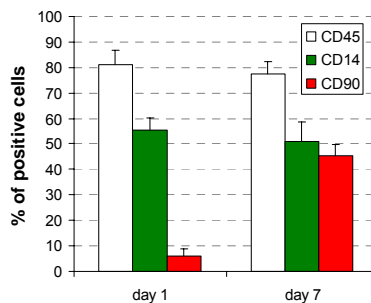


Figure 2: Expression patterns of CD45, CD14 and CD90 on cells on day 1 and day 7 (n=6 for each bar).

Enzyme-activities

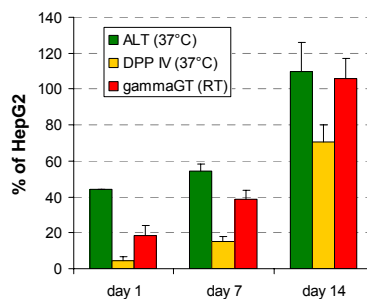


Figure 3: Specific enzyme activities of GGT, ALT and DPP IV increase during the transformation process. Values as % of HepG2 specific activities (n= 4-12).

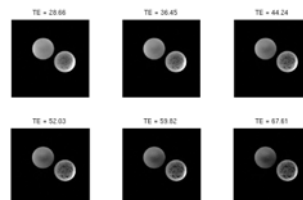


Figure 4: MR-analysis (T2) of iron-labelled cells cast in gelatin in Falcon-tubes.

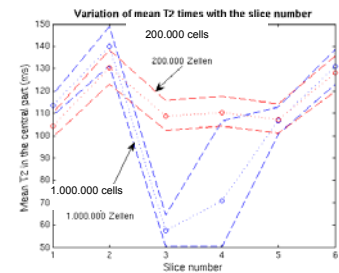


Figure 5: Variation of T2-times (mean and 95% CI) by 2×10^5 and 1×10^6 iron-labelled cells cast in gelatin in Falcon-tubes (coronary slices).

Conclusions:

- Transformed peripheral monocytes show enhanced CD90-expression after 7 days in culture
- During the transformation process, there is an increase in specific enzyme activities, reaching values comparable to HepG2 cells
- Labelling of transformed monocytes using supraparamagnetic iron oxides enables detection of these cells by MR-analysis
- Thus, SPIO-labelling can be used in further transplantation experiments using transformed monocytes in models of liver damage

Plans/work in progress:

- Comparison of transformed Monocytes and primary Hepatocytes concerning functional parameters
- Labelling and in vivo imaging of transplanted transformed monocytes in animal models of liver damage