

## VEGF improved liver regeneration monitored by MRI

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### Background/ Aims:

The liver is a metabolic powerhouse and is unique in its capability to regenerate and restore the metabolic demand after injury as partial hepatectomy (PHx). PHx is usually required in numerous pathological conditions such as cirrhosis and liver metastasis. Under these circumstances liver regeneration is not efficient as in the normal liver. There are many factors affecting proliferation and regeneration in the liver and VEGF is suspected to induce proliferation. The aim of this study was to find a way to improve liver regeneration in mice. Using MRI we were able to follow angiogenesis and liver regeneration non-invasively in VEGF conditional transgenic (Tgs) mice.

### Methods:

**Animal models:** Tgs mice expressing VEGF in the liver under Tetracycline regulation were used, as described previously<sup>1</sup>. 30% PHx was performed on these adult mice. The VEGF Tgs mice were either switched 'on' 1 day before PHx by removal of tetracycline (VEGF 'on') or were kept with Tetracycline through the whole experiment (VEGF 'off').

**MRI:** MRI experiments were performed on a 4.7 T Bruker Biospec spectrometer on days 0-6 after PHx (n=3-4 per each time point per group). T<sub>1</sub> weighted SE images were acquired (TR/ TE =400/18ms) for liver volume estimations. Changes in hepatic hemodynamics were evaluated from GE images acquired during breathing of air, air-CO<sub>2</sub> (95% air and 5% CO<sub>2</sub>), and carbogen (95% oxygen and 5% CO<sub>2</sub>) as described<sup>1</sup>. Four images were acquired at each gas mixture ( slice thickness = 1.5 mm; TR/TE= 100/10 ms; FOV = 5.8 cm; 256 × 128 ; 4 averages). Other experimental details were as reported<sup>1</sup>. The change induced by carbogen (VF) signifies vascular density and tissue perfusion. The change generated by CO<sub>2</sub> (VD) corresponds to the distribution of blood nourishment to the liver. Data is presented in color maps overlaid on the original baseline image for |VD| and |VF| >0.005<sup>1</sup>. Data was compared to histology.

### Results:

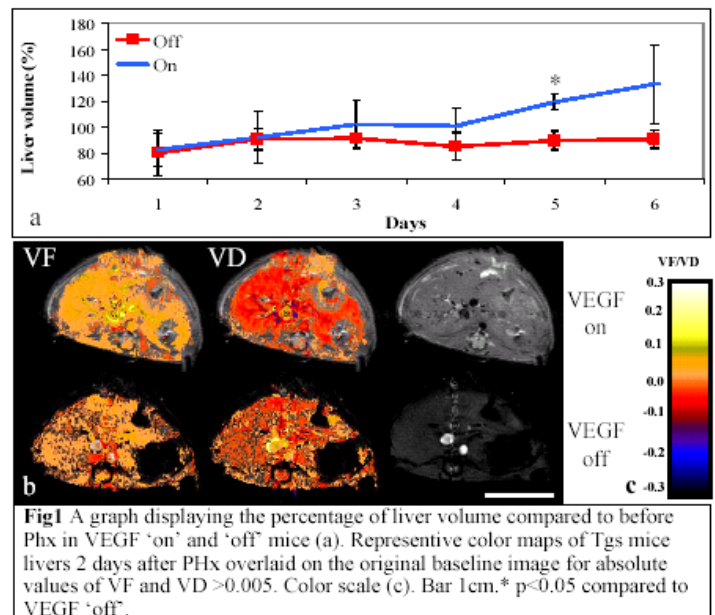
After PHx in VEGF 'on' mice, regeneration of the liver appeared more advanced and the size of the scar was reduced. This was demonstrated by volume measurements from MRI scans (fig1a) and amplified proliferation (4 fold) determined by BrdU staining of histology. Normally liver volume is regulated tightly however, the volume of the VEGF 'on' liver expanded to 130% approximately (fig1a, p<0.05). In the initial regenerative process, perfusion usually declines, since hepatocyte proliferation precedes endothelial proliferation. MRI functionality scans revealed ability of VEGF to improve liver perfusion illustrated by higher VF values (fig1b, p<0.05). Normally in regeneration, VD values ascend and become positive signifying a change in the source of nourishment. Since there is a growing need of energy in the liver the hepatic artery becomes the main source of blood enriching the liver with oxygenated blood. However, with VEGF, VD values were more negative (fig1b, p<0.05) as perfusion was better and there was no requirement for change in nourishment.

### Discussion:

VEGF induced early and improved liver regeneration. This could be exploited in pathological conditions, such as cirrhosis and cancer, in which PHx is required and regeneration is not as efficient as under normal conditions. More research needs to be done in order to define the exact therapeutic window of VEGF for liver regeneration. In future, assessment of VEGF therapy should be made on a cirrhotic model. MRI provides us with unique information about the exceptional process of liver regeneration. It enables us to follow this process non-invasively providing functional information of the tissue.

### References

1. Dor Y., Djonov V, Abramovitch R, Itin A, Fishman G, Carmeliet P, Goelman G and Keshet E. EMBO J. 21:1939-47, 2002.
2. Abramovitch R., Dafni H., Smouha E., Benjamin L.E. and Neeman M. Canc Res 59:5012-5016, 1999.



**Fig1** A graph displaying the percentage of liver volume compared to before Phx in VEGF 'on' and 'off' mice (a). Representative color maps of Tgs mice livers 2 days after PHx overlaid on the original baseline image for absolute values of VF and VD >0.005. Color scale (c). Bar 1cm. \* p<0.05 compared to VEGF 'off'.