

## Supplementary Figure S1: Determination of hepatocyte cell volume and generation of an internal standard for quantitative immunoblotting

**A) Determination of hepatocyte cell volume.** Optical slices obtained from fluorescence microscopy were binarised using ImageJ Software. The amount of black pixels was calculated and multiplied by a device dependent scaling factor to receive hepatocyte cell volume. This method was validated by measurements on fluorescent microspheres of known size.

**B)** *In vitro* **phosphorylation of recombinant p38**<sup>MAPK</sup>. p38<sup>MAPK</sup> and MK2 (not shown) was recombinantly expressed in E. coli, purified by affinity chromatography using Glutathion S Sepharose (GE Healthcare) and *in vitro* phosphorylated as described above. Protein was then applied to SDS-PAGE, transferred to nitrocellulose membrane by western blotting and detected by specific antibodies. (-) Protein without *in vitro* phosphorylation, (+) protein after *in vitro* phosphorylation.

**C** and **D**) Determination of  $p38^{MAPK}$  protein concentration in solution. *In vitro* phosphorylated  $p38^{MAPK}$  and MK2 (not shown) was applied to SDS-PAGE, co-separated with a serial dilution of BSA. After staining with Coomassie Brilliant Blue staining intensity was calculated to determine the concentration of  $p38^{MAPK}$  and MK2 (not shown) in solution.

**E** and **F**) Calibration of the internal standard. Primary mouse hepatocytes were treated for 20 minutes with 20 ng·mL-1 of IL-1 $\beta$  and lysed in triton buffer. The lysate was applied to SDS-PAGE and Western Blotting as a serial dilution together with the *in vitro* phosphorylated protein. Signal intensity was calculated to determine the concentration of Phospho-p38<sup>MAPK</sup>, total p38<sup>MAPK</sup> (not shown), Phospho-MK2 (not shown) and total MK2 (not shown), respectively.



## Supplementary Figure S3: Determination of BMDM cell volume

**A)** Optical slices from a representative measurement on bone marrow derived macrophages (BMDM) obtained from fluorescence microscopy were combined to a 3D illustration with the software Volocity (Improvision). B) Furthermore, optical slices from 54 independent measurements were binarised using ImageJ Software. The amount of black pixels was calculated and multiplied by a device dependent scaling factor to receive BMDM cell volume (2.07 pL, S.E.M.: 0.07).