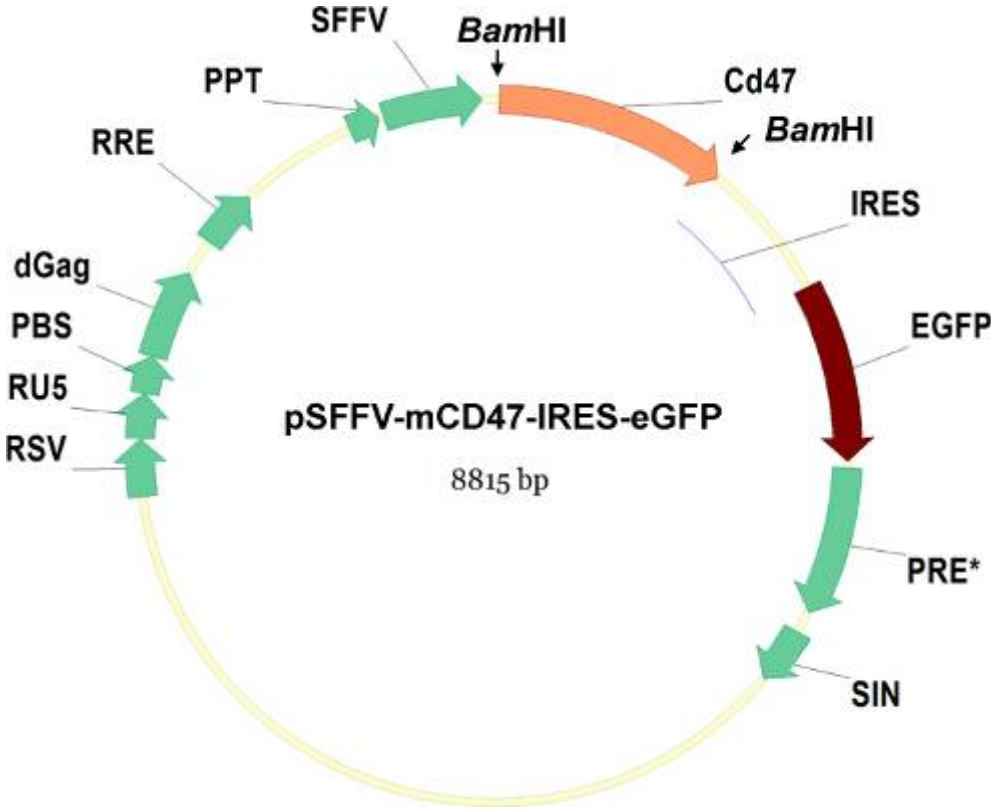
Supporting information

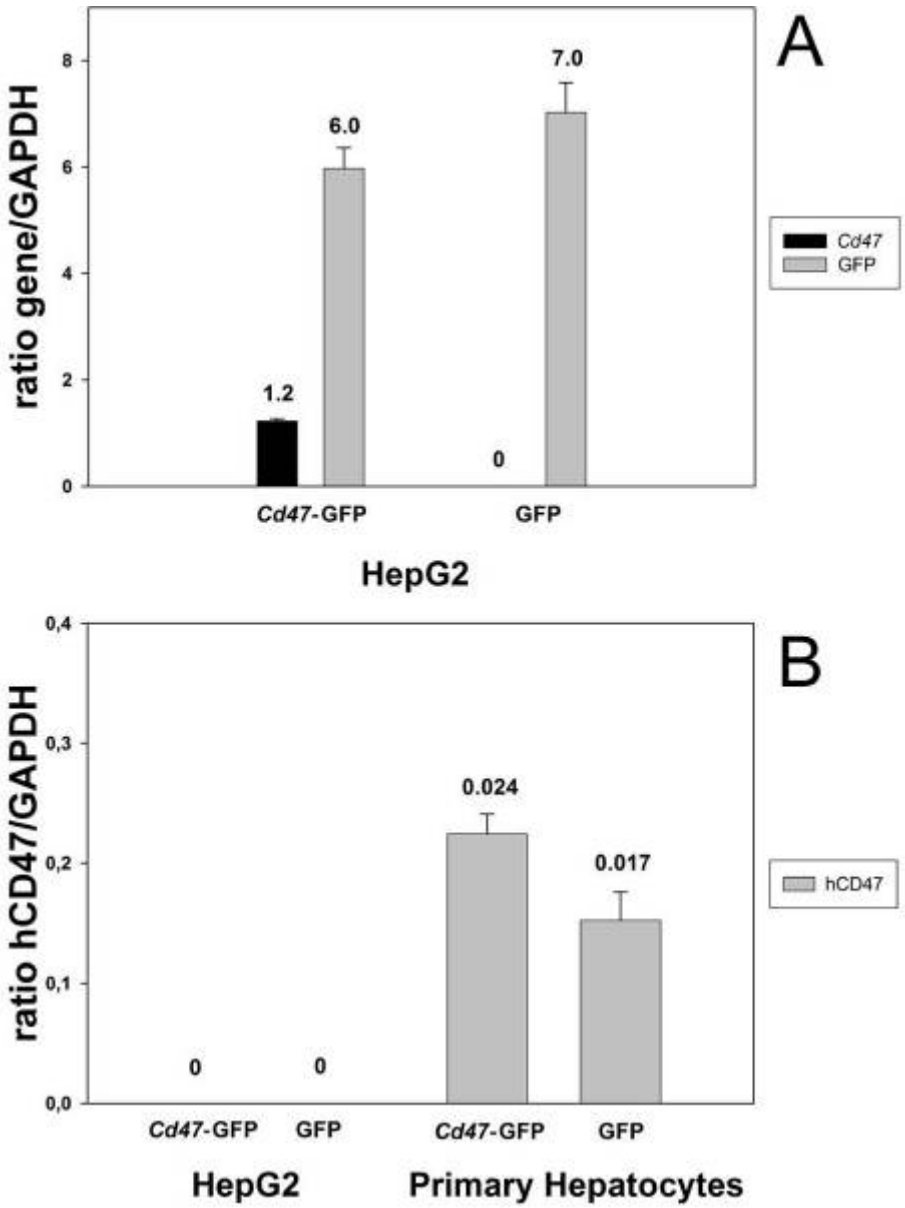
Supplementary Figure 1



Supplementary Fig 1. Lentiviral vector map, pSFFV-Cd47-IRES-EGFP.
Cd47 was cloned into a lentiviral transfer vector with *Bam*HI restriction sites and placed under control of a strong SFFV promoter. eGFP was placed downstream of the ribosome binding site IRES to ensure parallel expression with Cd47.

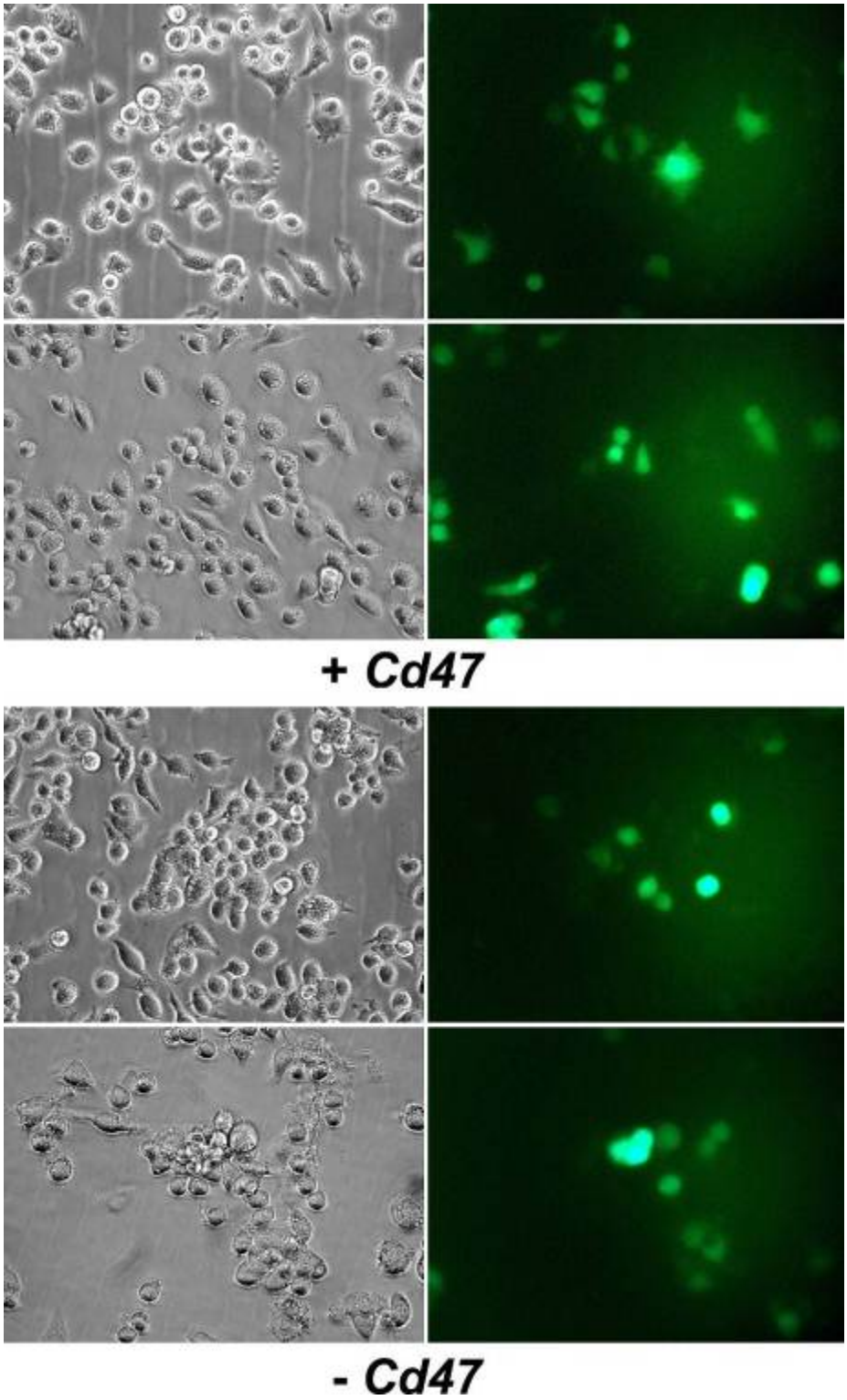
SIN: Self-Inactivating 3' Long Terminal Repeat (LTR);
PRE*:post-transcriptional regulatory element
IRES: EMCV internal ribosome entry site
SFFV: strong internal promoter of SFFV (Spleen focus forming virus)
PPT: central polypurine tract
RRE: Rev responsive element
dGag: portion of the HIV-1 *gag* gene with a closed reading frame
PBS: primer binding site
RU5: 5'-HIV-1 repeat and U5-region of the LTR
RSV: rous sarcoma virus promoter

Supplementary Figure 2



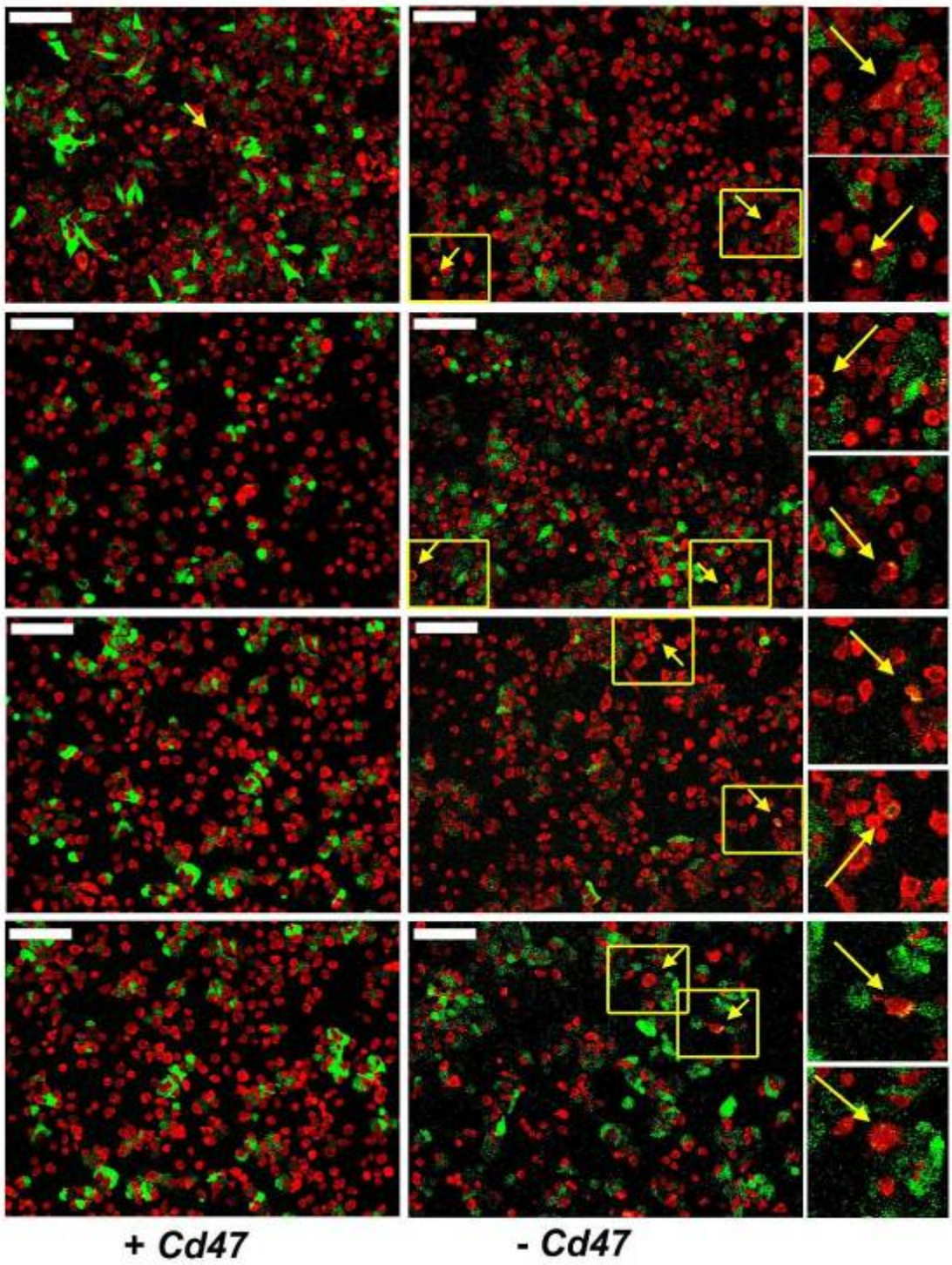
Supplementary Fig. 2. RT-qPCR for expression levels of Cd47, GFP and hCD47.
Total RNA was harvested from HepG2 cells and BD primary hepatocytes transduced with LV-Cd47-GFP (**Cd47-GFP**) or LV-GFP (**GFP**) as control at a multiplicity of infection of 10 and transcribed into cDNA. **a** Transcription levels of Cd47 (black bars) and GFP (grey bars) were determined by primers specifically designed to discriminate Cd47 from human CD47. Cd47 is transcribed 1.2-fold in comparison to GAPDH in HepG2-Cd7-GFP. GFP transcription levels were similar in transduced HepG2 cells. **b** hCD47 transcription levels were analysed in HepG2 cells and primary hepatocytes. HepG2 cell do not express hCD47 while primary hepatocytes show hCD47 transcription. Error bars: SD; n = 3.
Method: Total RNA (RNeasy, Qiagen) from transduced cell lines was DNAse-treated (Ambion) and reverse-transcribed into cDNA (IScript, BioRad). CD47 copy numbers were determined by quantitative PCR using SYBR Green (BioRad) and normalized against human GAPDH copy numbers. Primers used discriminated endogenous hCD47 from Cd47. (Supplementary Material 1)

Supplementary Figure 3



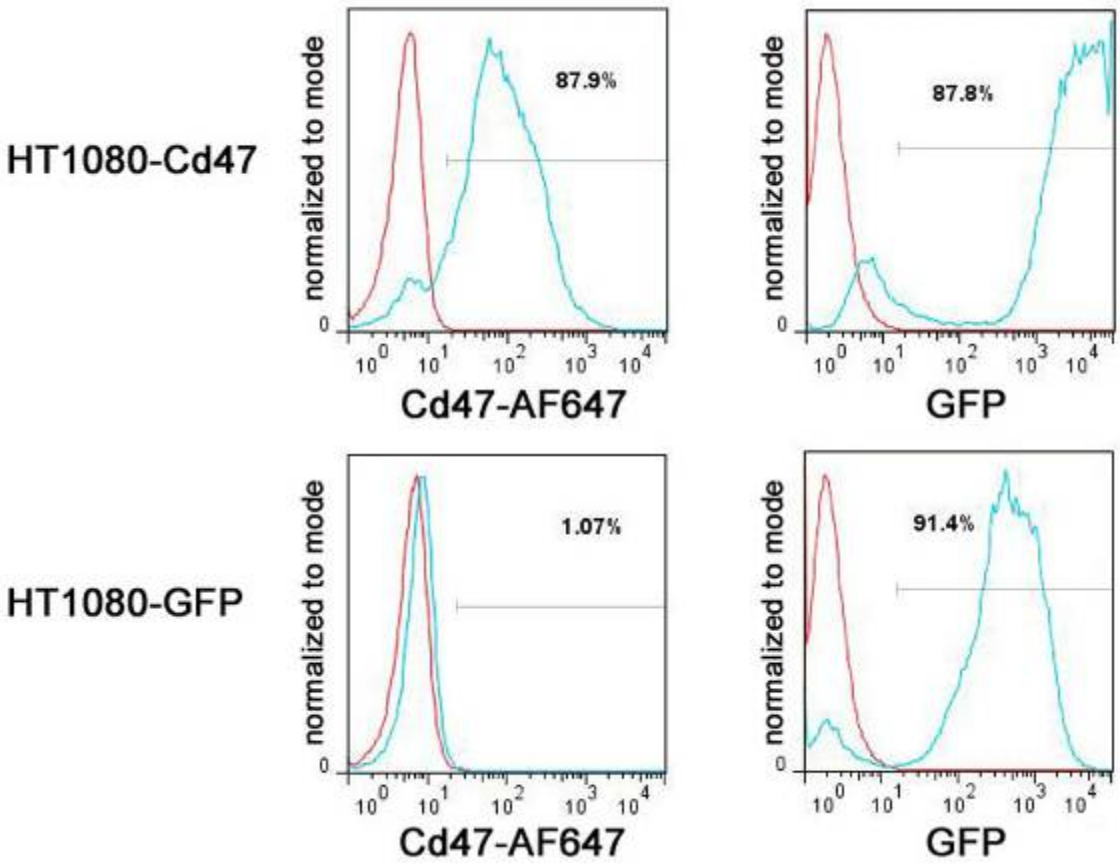
Supplementary Fig. 3. Additional examples for contact assay.
HepG2-Cd47-GFP cells were incubated with RAW264.7 macrophages for 14 hours. Contact of HepG2-Cd47-GFP (+**Cd47**) and HepG2-GFP (**-Cd47**) with RAW264.7 macrophages was monitored and percentage of contacting macrophages within total macrophage counts was calculated to determine a phagocytotic index. These images demonstrate that RAW264.7 macrophages appear randomly scattered between HepG2 cells with surface expression of Cd47 while macrophages cluster around HepG2 with no Cd47 expression.

Supplementary Figure 4



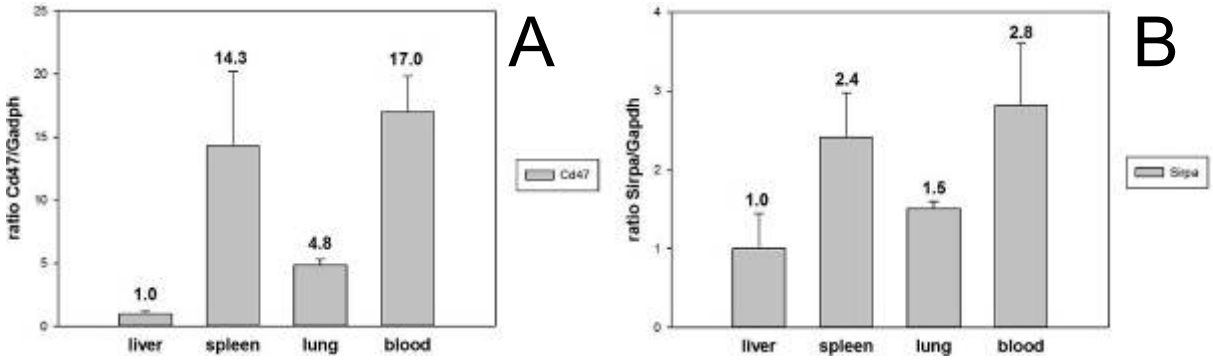
Supplementary Fig. 4. Additional examples for phagocytotic assay.
HepG2-Cd47-GFP **(+Cd47)** and HepG2-GFP **(-Cd47)** were incubated with TAMRA-labelled RAW264.7 murine macrophages for 14 hours and phagocytotic events were counted. Phagocytotic events occur more often when RAW264.7 macrophages encounter HepG2 cells not expressing Cd47 (**-Cd47**). Areas of phagocytotic events are magnified in the right panel for identification of double positive yellow macrophages (**right panel**). Scale bars, 100 μm.

Supplementary Figure 5



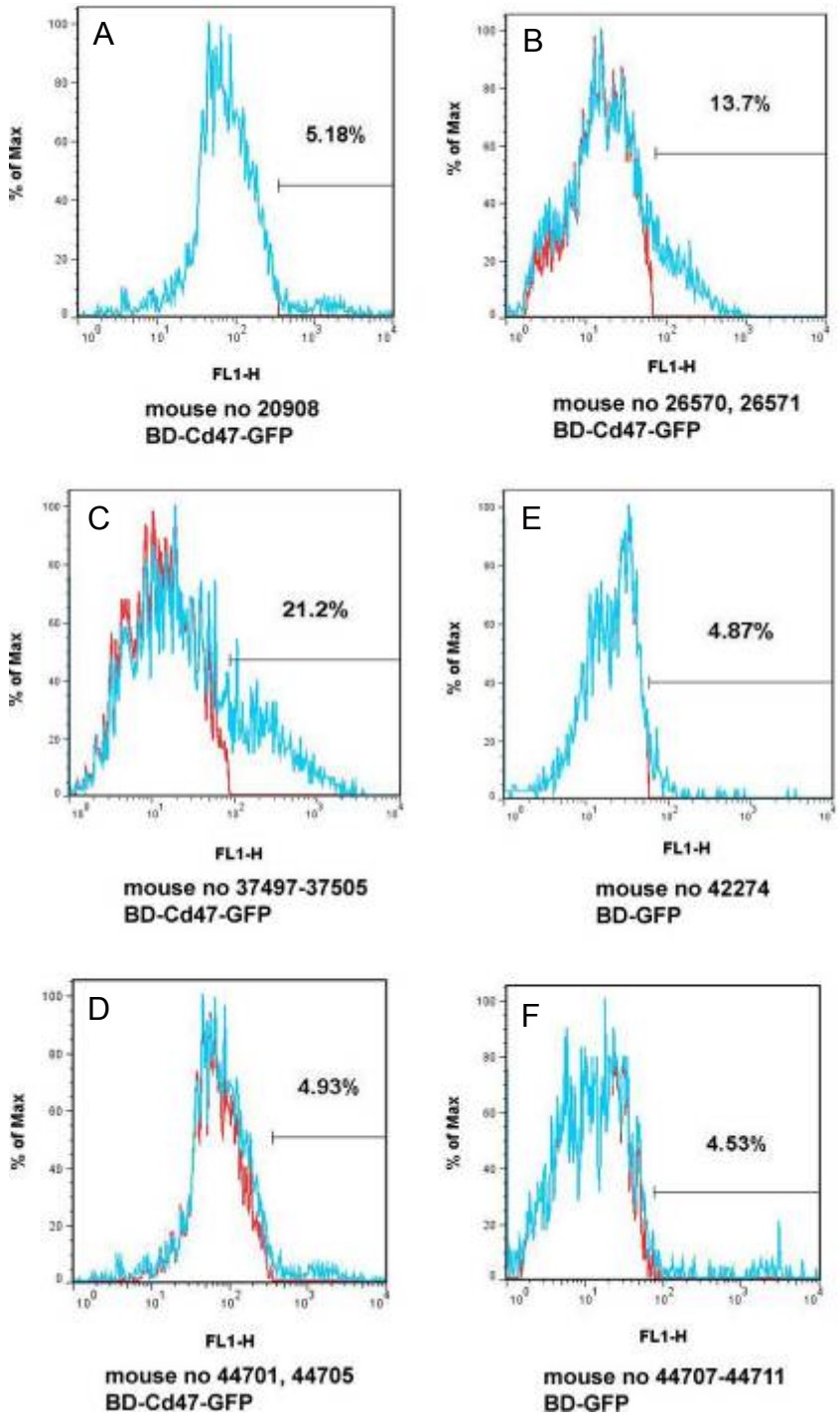
Supplementary Fig. 5. Cd47 and GFP expression in transduced HT1080 cells.
HT1080 cell lines were transduced with LV-Cd47-GFP or LV-GFP at a multiplicity of infection of 10, as titratred on HepG2 cells. Transduction efficiency was determined by measuring GFP- and Cd47-expression by FACS. HT1080-Cd47-GFP show strong co-expression of Cd47and GFP while control HT1080-GFP only show expression of GFP.

Supplementary Figure 6



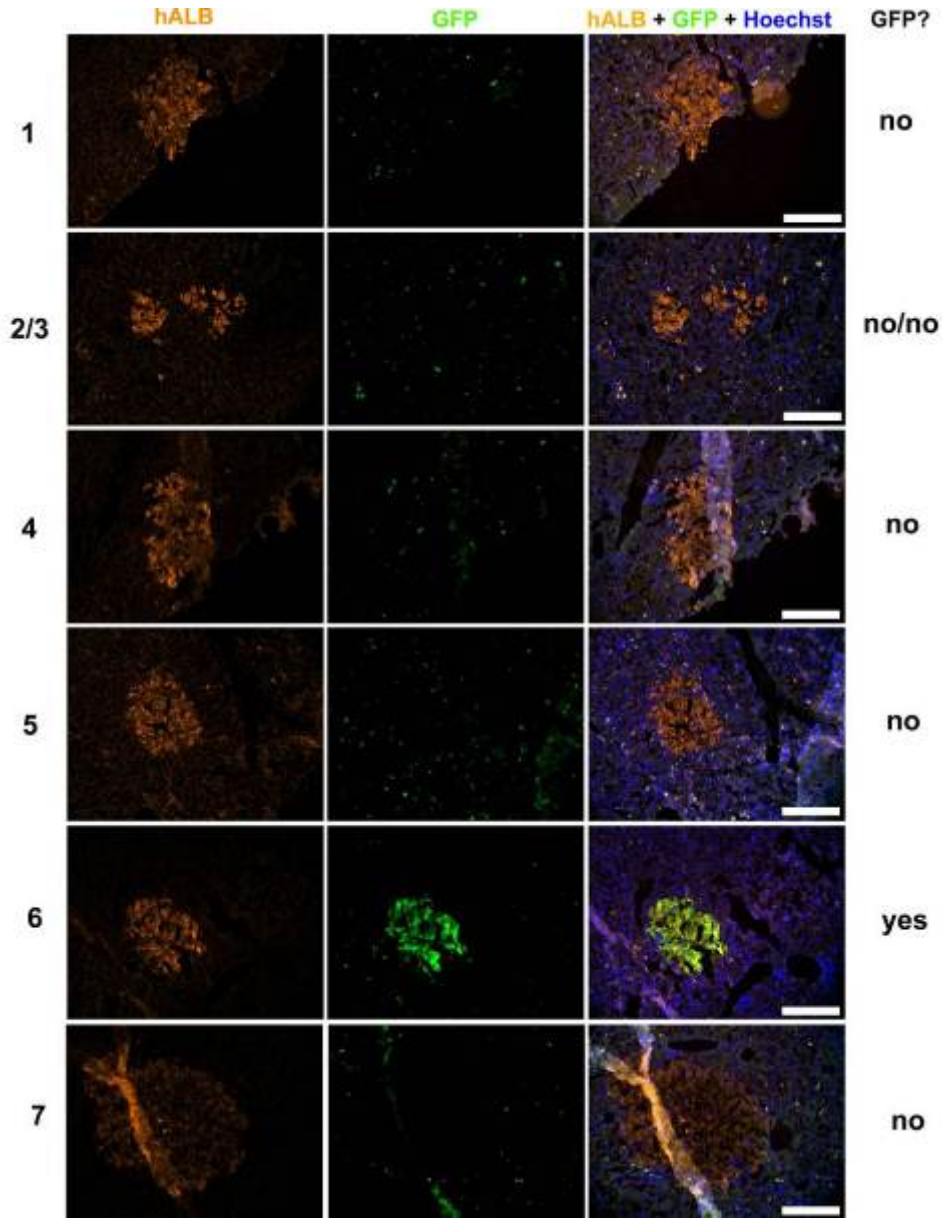
Supplementary Fig. 6. Cd47 and Sirpa expression in BALB-ΔRAG/γ_c-uPA mice.
Relative mRNA expression levels quantified by RT-qPCR of A) Cd47 and B) Sirpa in various macrophage enriched tissues of BALB-ΔRAG/γ_c-uPA mice compared to the liver (set arbitrarily to 1). Mean values of whole organ mRNA-samples of three mice are shown. Error bars: SD.

Supplementary Figure 7

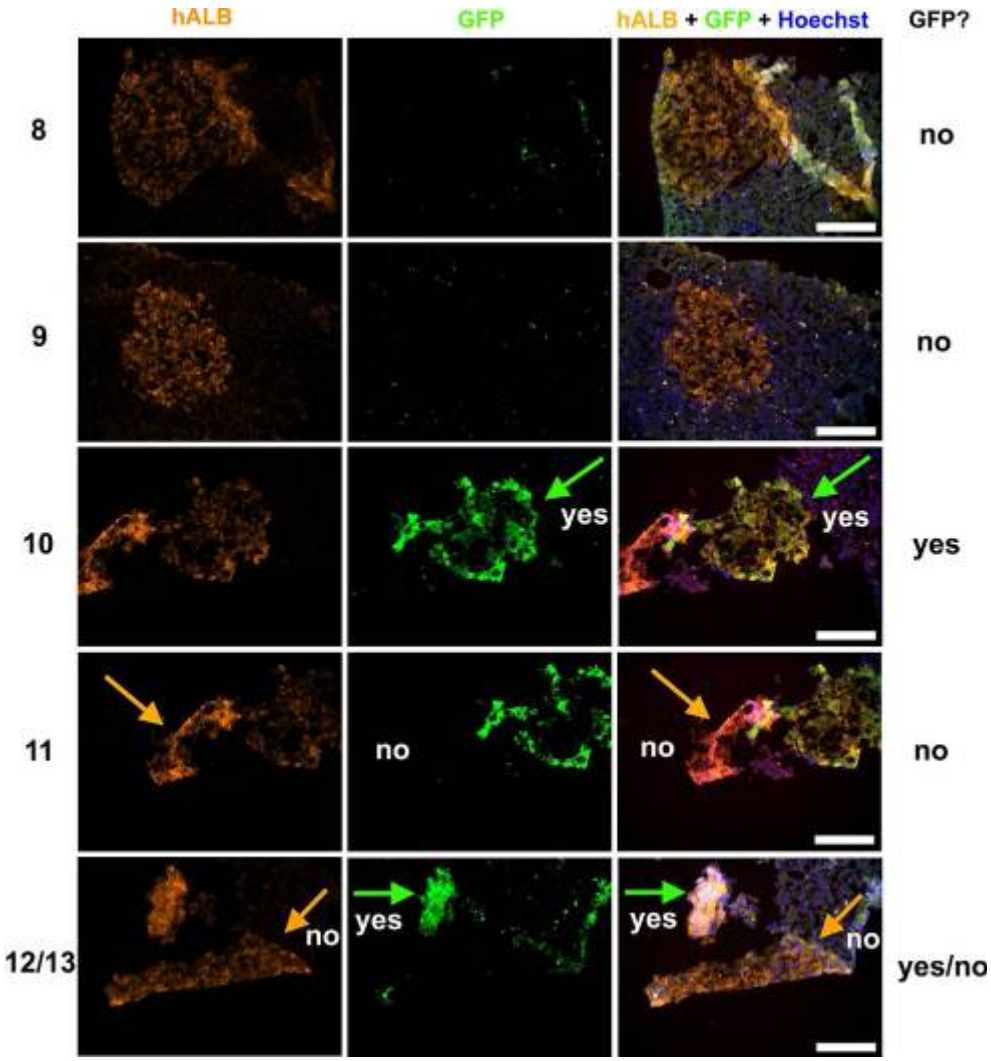


Supplementary Fig. 7. Initial transduction efficiencies in transplanted BD hepatocytes. FACS profiles of all transduction batches A-F (see also Table 1 column 6)
BD cryopreserved hepatocytes were transduced with LV-Cd47-GFP or LV-GFP and transplanted into uPA mice. Additional samples were kept in culture for 5 days and analysed by FACS for GFP expression to determine pre-transplantation transduction efficiency. Transduction efficiency was used for comparison with *in vivo,* post-transplantation percentages of GFP expressing clusters within the total $hALB^{+}$-cluster population in tissue samples.

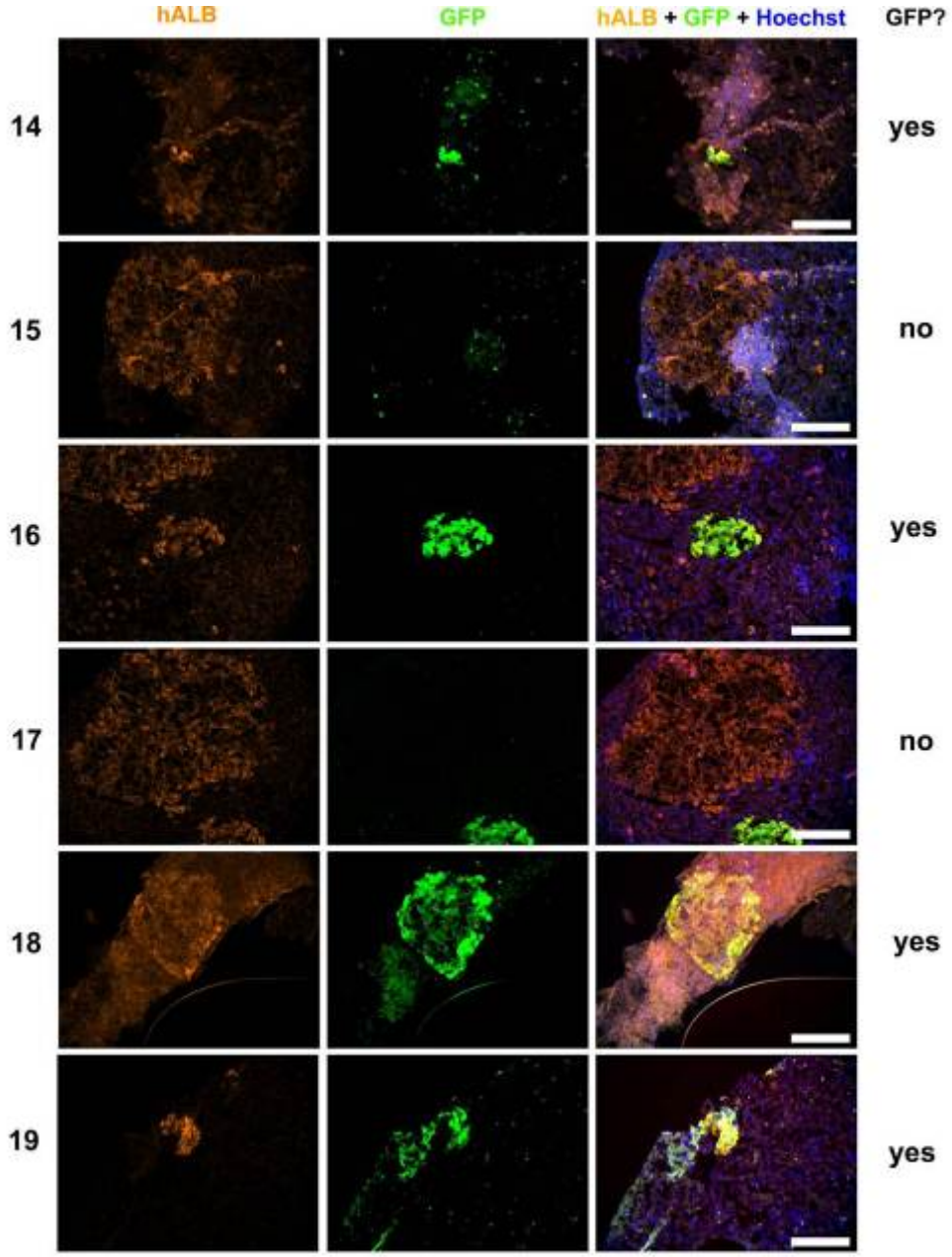
Supplementary Figure 8 (1)



Supplementary Figure 8 (2)



Supplementary Figure 8 (3)



Supplementary Fig. 8. Judgement examples for tissue immunofluorescence stainings.
Cluster images from the liver of mouse 37501 transplanted with LV-Cd47-GFP transduced hepatocytes, cryo-sectioned and stained for hALB (left panel) and GFP (middle panel). Each human albumin positive cluster was analysed for GFP expression (yes and no). Cluster 10-13 are in close proximity to each other and GFP expressing clusters are marked with a green arrow. GFP negative clusters are marked with an orange arrow. The percentage of GFP-positive human albumin clusters was divided by the percentage of GFP positive cells detected in the initial FACS 6 days after transplantation to determine the ratio. In this mouse 19 hALB clusters were identified. 7 of them were GFP positive, resulting in 36.8 %, to be compared to 21.2 % transduction efficiency measured by FACS analysis of transduced hepatocytes in culture five days after transplantation (see Fig. 6B). The ratio is calculated to 1.7 in mouse 37501 (see Table 1). Scale bars, 250 µm.

Supplementary Material 1

Primer	Sequence	comment
forward cloning primer murine Cd47	5’ -GCGGATCCGGAGATGTGGCCCTTGGCG- 3’	*Bam*HI site
reverse cloning primer murine Cd47	5’ -CGGGATCCCACCTATTCCTAGGAGGTT- 3’	*Bam*HI site
forward qPCR primer murine Cd47	5’ -TAGCACTACTACAGATCAAA- 3’	Specific to murine Cd47
reverse qPCR primer murine Cd47	5’ -CACCATGGCATCGCGCTTAT- 3’	Specific to murine Cd47
forward qPCR primer human CD47	5’ -CAAGTCCACTGTCCCCACTG- 3’	Specific to human CD47
reverse qPCR primer human CD47	5’ -TGTGTGTGAGACAGCATCACT- 3’	Specific to human CD47
forward and reverse murine Sirpa,	Mm_Sirpa_1_SG QuantiTect Primer Assay, Qiagen	
forward qPCR primer GFP	5’ - ACTTCTTCAAGTCCGCCATG – 3’	--
reverse qPCR primer GFP	5’ - AGCTCGATGCGGTTCACCAG -3’	--
forward qPCR primer human GAPDH	5’ -CTCTGGTAAAGTGGATATTG- 3’	Specific to human GAPDH
reverse qPCR primer human GAPDH	5’ -CTCCCCCCTGCAAATGAG- 3’	Specific to human GAPDH

Supplementary Material 1. Cloning and real-time PCR primers used.

Supplementary Material 2

atgtggcccttggcggcggcgctgttgctgggctcctgctgctgcggttcagctcaactactg
tttagtaacgtcaactccatagagttcacttcatgcaatgaaactgtggtcatcccttgcatc
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gggatcatagctctagcagaactacttggattagtttatatgaagtttgtcgcttccaaccag
aggactatccaacctcctaggaataggtga

Supplementary Material 2. Corrected nucleotide sequence of Cd47.
Murine Cd47 was amplified from embryonic mouse liver and Hepa 1.6 cells, cloned into pCR4-TOPO and sequenced. In comparison to AK_151922, there is a base exchange at bp 716 from adenine to guanine of the open reading frame (underlined). The nucleotide sequence was accepted under the accession number HQ585874 at Genebank.

Supplementary Material 3

MWPLAAALLLGSCCCGSAQLLFSNVNSIEFTSCNETVVIPCIVRNVEAQSTEEMFVKWKLNKS
YIFIYDGNKNSTTTDQNFTSAKISVSDLINGIASLKMDKRDAMVGNYTCEVTELSREGKTVIE
LKNRTVSWFSPNEKILIVIFPILAILLFWGKFGILTLKYKSSHTNKRIILLLVAGLVLTVIVV
VGAILLIPGEKPVKNASGLGLIV**V**STGILILLQYNVFMTAFGMTSFTIAILITQVLGYVLALV
GLCLCIMACEPVHGPLLISGLGIIALAELLGLVYMKFVASNQRTIQPPRNR

Supplementary Material 3. Corrected amino acid sequence of Cd47.
Cd47 amino acid sequence from embryonic mouse liver cells and Hepa 1.6 cells. The amino acid exchange I213V is underlined. The amino acid sequence has been accepted as protein ID ADQ12919.1 at Genebank.

Supplementary Material 4.

Antigen	raised in	label	dilution	Company	catalog #
CD47, murine	goat		1/250 (WB: HepG2) 1/100 (IF)	R&D Systems	Baf 1866
CD47, murine	goat		1/200 (WB: Hepatocytes)	Santa Cruz	SC-7059
Albumin, human	goat		1/200 (IF)	Bethyl	A80-229A
GFP	rabbit		1/100 (IF)	Invitrogen	A11122
Actin	rabbit monoclonal		1/1000 (WB)	Sigma	A5060
Goat IgG - H&L - F(ab)2	rabbit	HRP	1/1000 (WB)	Abcam	ab5755
Rabbit IgG - F(ab)2	goat	HRP	1/1000 (WB)	Abcam	ab6112
rat IgG whole	goat monoclonal	HRP	1/1000 (WB)l	Acris	R1614HRP
Goat IgG (H+L)	donkey	AF647	1/500 (IF)	Invitrogen	A-21447
Rabbit IgG (H+L)	donkey	AF488	1/500 (IF)	Invitrogen	A21206
SIRPα, murine	rat	PE	1/100 (FACS)	BD Pharmingen	560107

Supplementary Material 4. Antibodies used.