

Role of macrophages in disease development and progression

Frank Tacke

3rd Paris NASH Symposium
July 6, 2017

Disclosures Frank Tacke

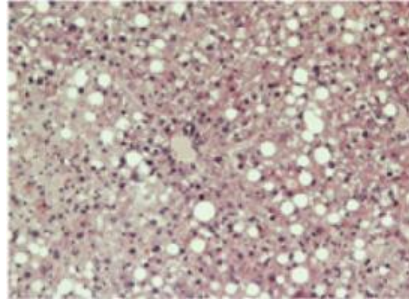
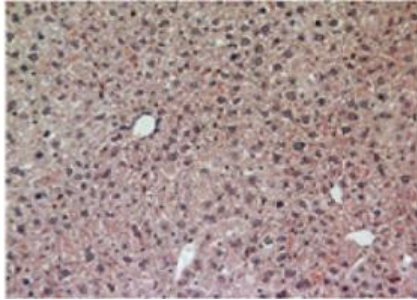
- *Research support (materials, funding):*
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- *Speaker/Advisory Board:*
Tobira/Allergan, Gilead, AbbVie, BMS, Boehringer, Galapagos, Intercept, Falk, Inventiva

Macrophage accumulation is a hallmark of progressive non-alcoholic steatohepatitis

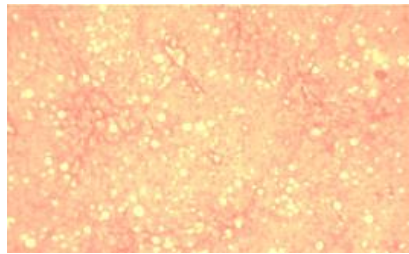
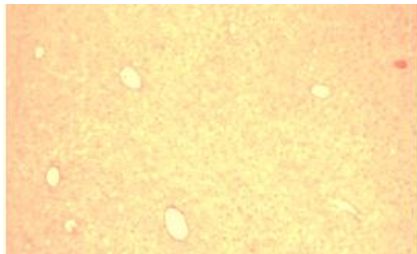
control

NASH model

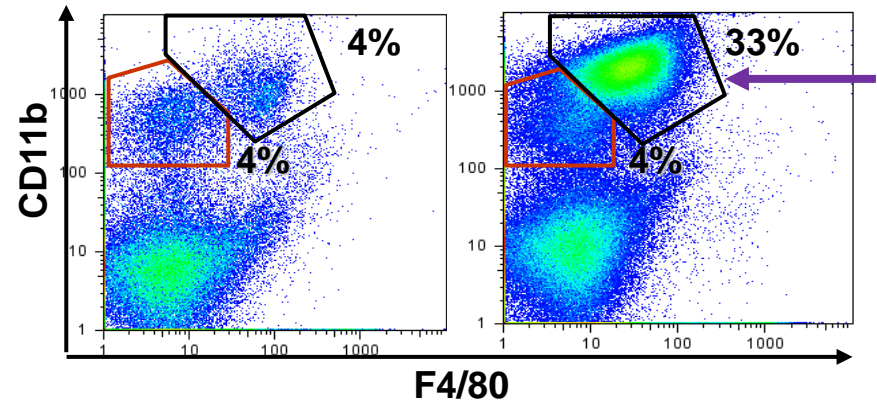
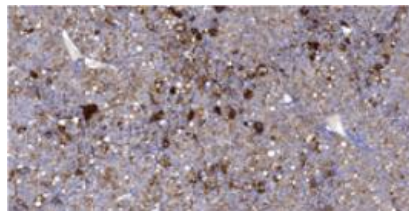
H&E



fibrosis



F4/80



Baeck C / Tacke F. *Hepatology* 2014

Ehling J / Tacke F. *Gut* 2014

Wehr A / Tacke F. *J Immunol* 2013

Baeck C / Tacke F. *Gut* 2012

Karlmarm KR / Tacke F. *Hepatology* 2009

Tacke F / Randolph GJ. *J Clin Invest* 2007

Monocyte / macrophage heterogeneity in liver inflammation, fibrosis and cancer

selected examples
for recent research

Origin
homeostasis vs.
inflammation

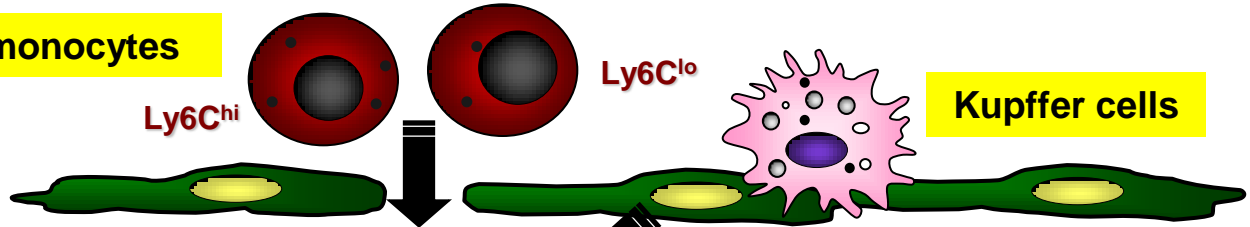
Mass E / Geissmann F. *Science*. 2016
Bleriot C, et al. *Immunity* 2015
Scott CL / Guilliams M. *Nat Commun* 2016
Wang J, Kubes P. *Cell*. 2016

monocytes

Ly6C^{hi}

Ly6C^{lo}

Kupffer cells



macrophages

Differentiation
microenvironmental signals
shape phenotype

Xue J / Schultze J. *Immunity* 2014
Beattie J / Kane PM. *J Hepatol* 2016
Bartneck M / Tacke F. *Hepatology* 2016

„M1 polarization“

inflammation ↑
tumor ↓

„M2 polarization“

inflammation ↓
remodelling ↑
tumor ↑

Function regarding liver injury
pro-inflammatory (can) develop into
restorative macrophages

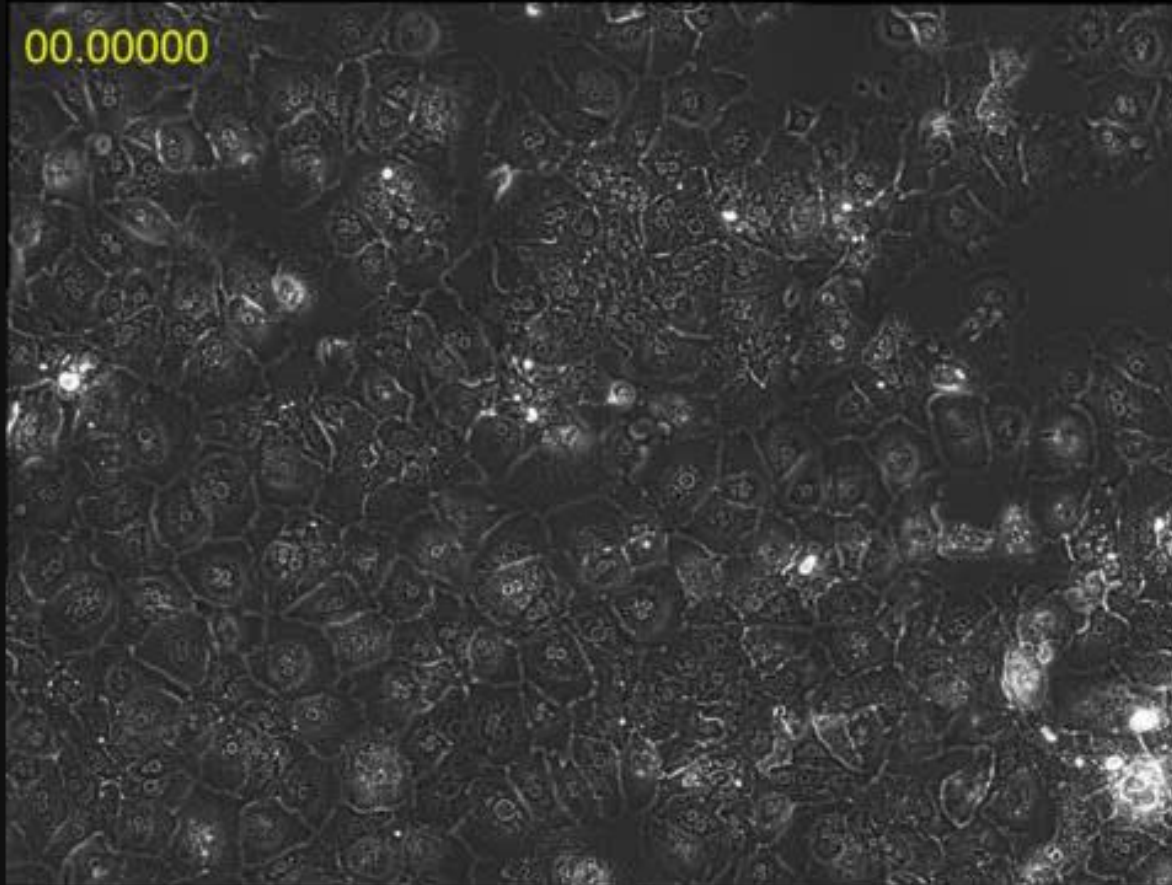
Dal-Secco D / Kubes P. *J Exp Med*. 2015
Ramachandran P / Iredale JP. *PNAS* 2012
Baeck C / Tacke F. *Hepatology* 2014

Monocytes and macrophages in liver diseases

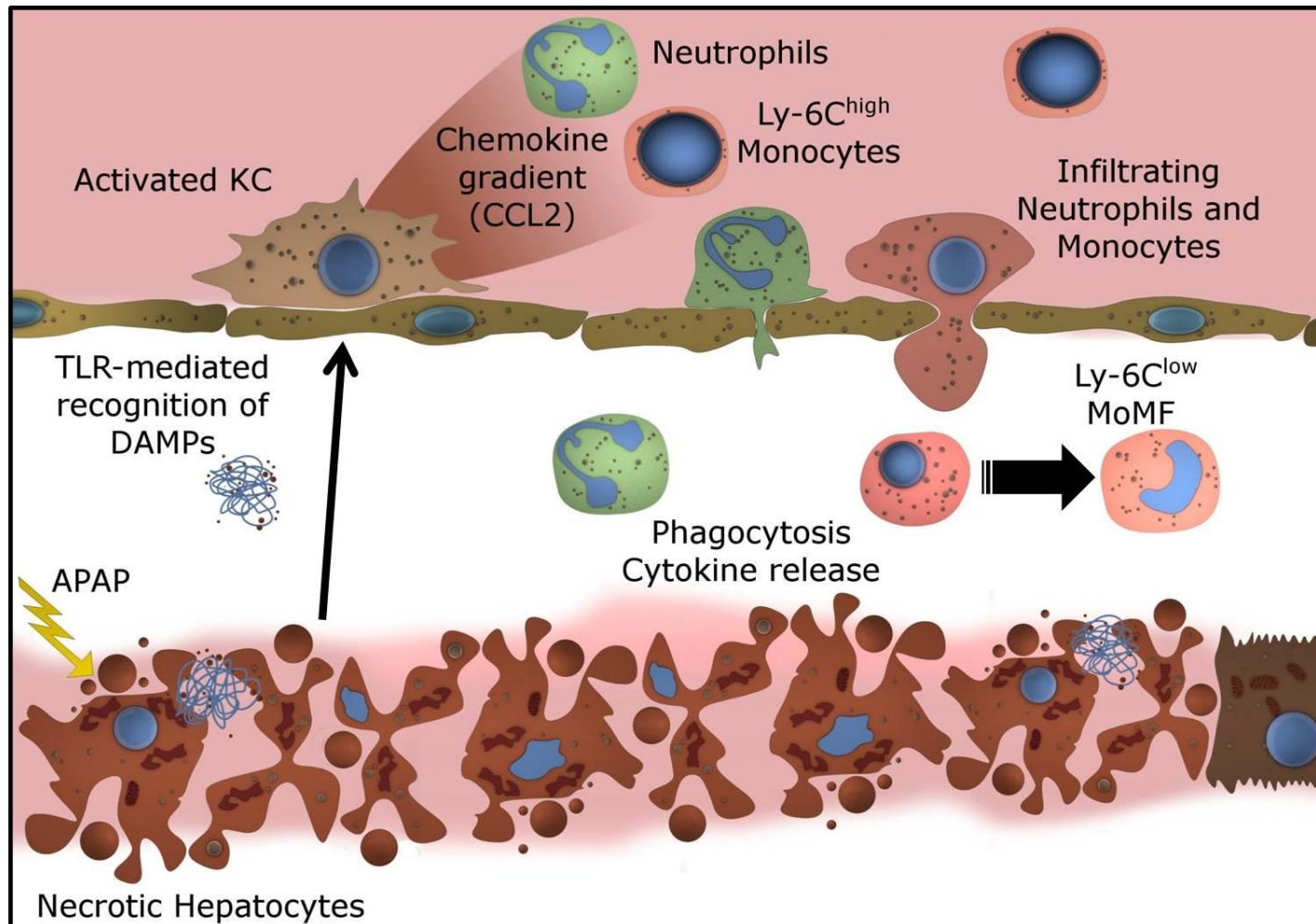
- **Acute liver injury**
- **NASH and fibrosis**
- **Immunometabolism**

Cell Death triggers immune cell homing: acetaminophen injury

Acetaminophen (APAP) 7mg/ml on primary mouse hepatocytes



Role of infiltrating monocytes: acetaminophen induced acute liver injury

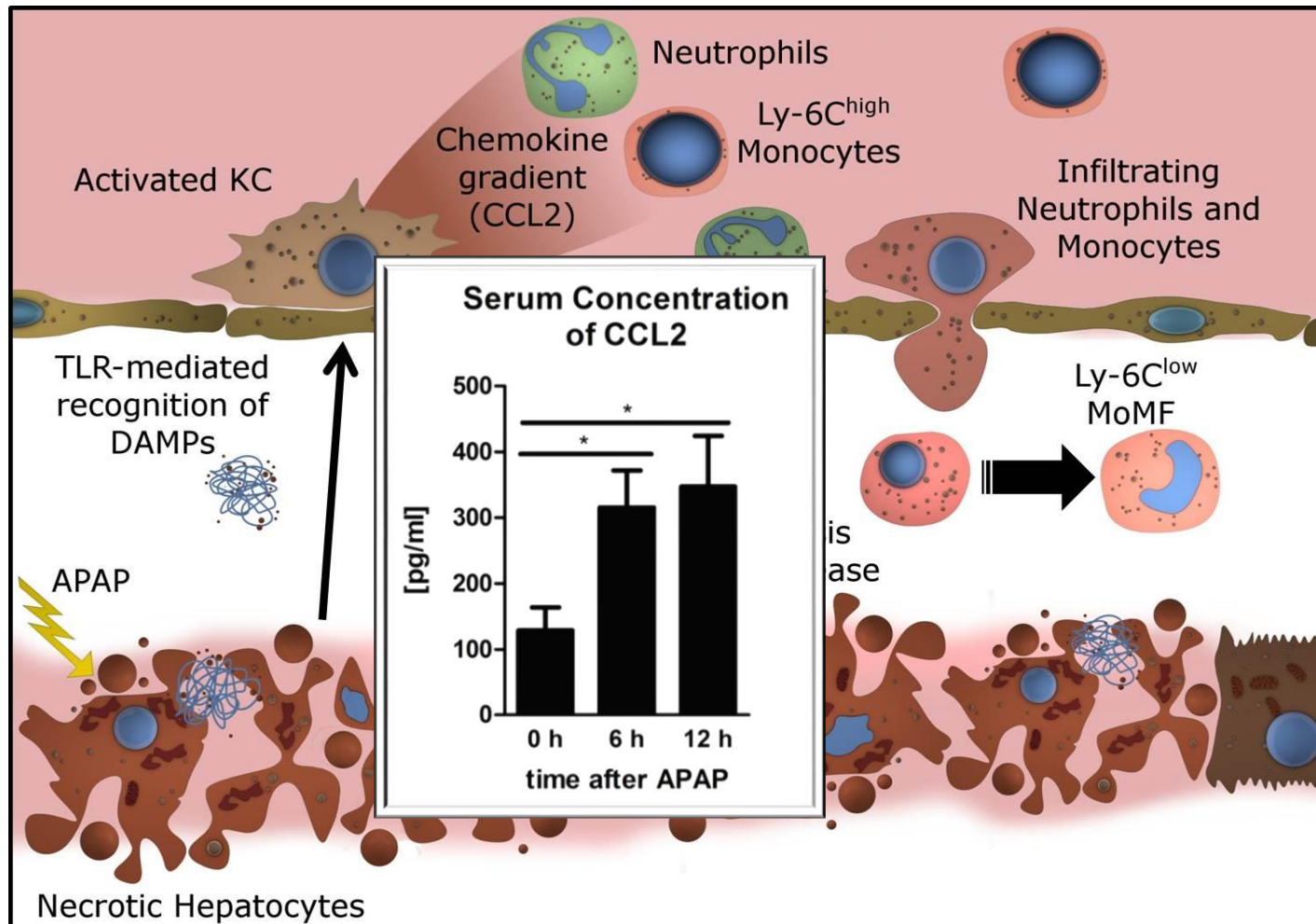


Krenkel O, Mossanen JC, Tacke F. *Hepatobiliary Surg Nutr.* 2014; 3(6):331–43.

based on: Huebener / Schwabe RF. *J Clin Invest* 2015; You Q / Ju C. *Biochem Pharmacol.* 2013;

Zigmond E / Varol C. *J Immunol* 2014

Role of infiltrating monocytes: acetaminophen induced acute liver injury



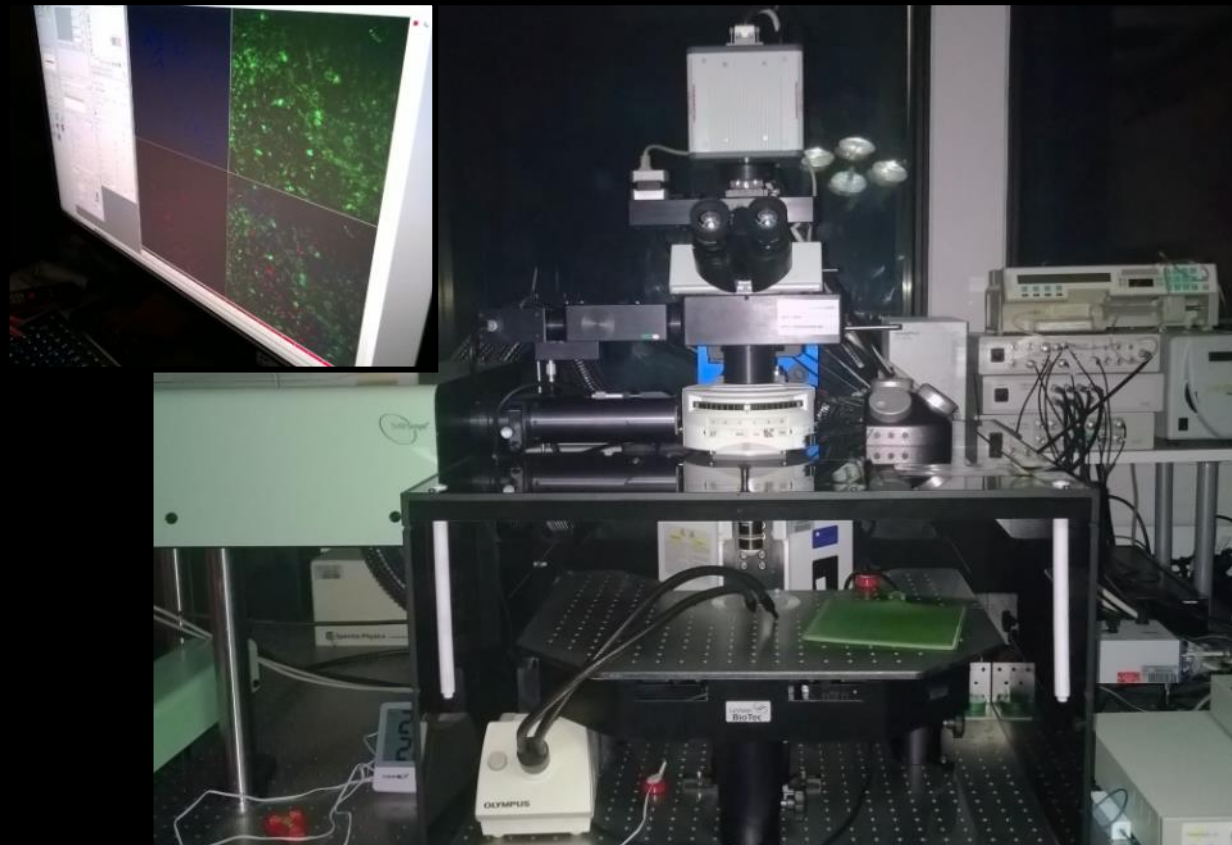
Krenkel O, Mossanen JC, Tacke F. *Hepatobiliary Surg Nutr.* 2014; 3(6):331–43.

based on: Huebener / Schwabe RF. *J Clin Invest* 2015; You Q / Ju C. *Biochem Pharmacol.* 2013;

Zigmond E / Varol C. *J Immunol* 2014

Monocyte and macrophage populations in experimental liver injury

intravital multiphoton-microscopy



Heymann F / Tacke F. *Hepatology* 2015
Heymann F / Tacke F. *J Vis Exp* 2015

Analyses of migration and cell-cell-
interactions in real-time *in vivo*

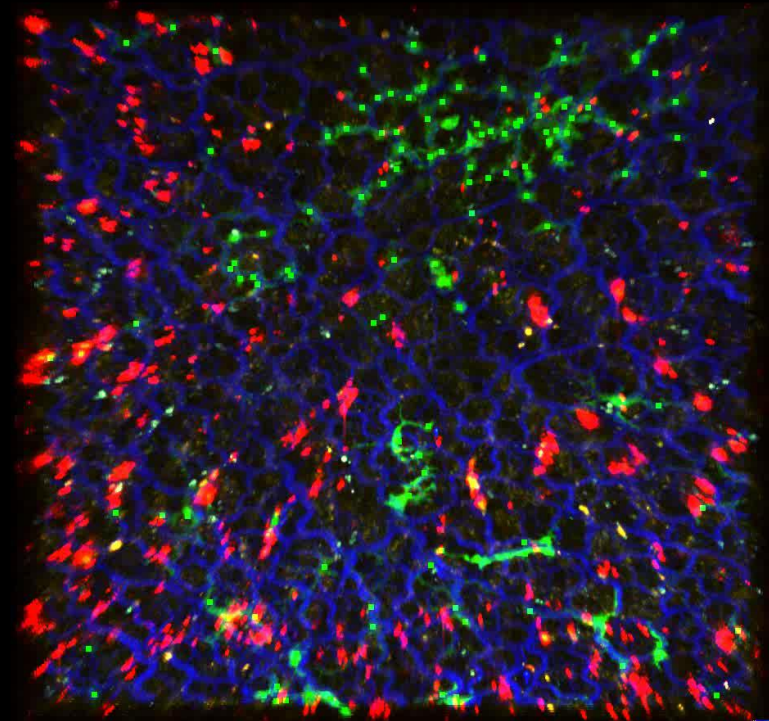
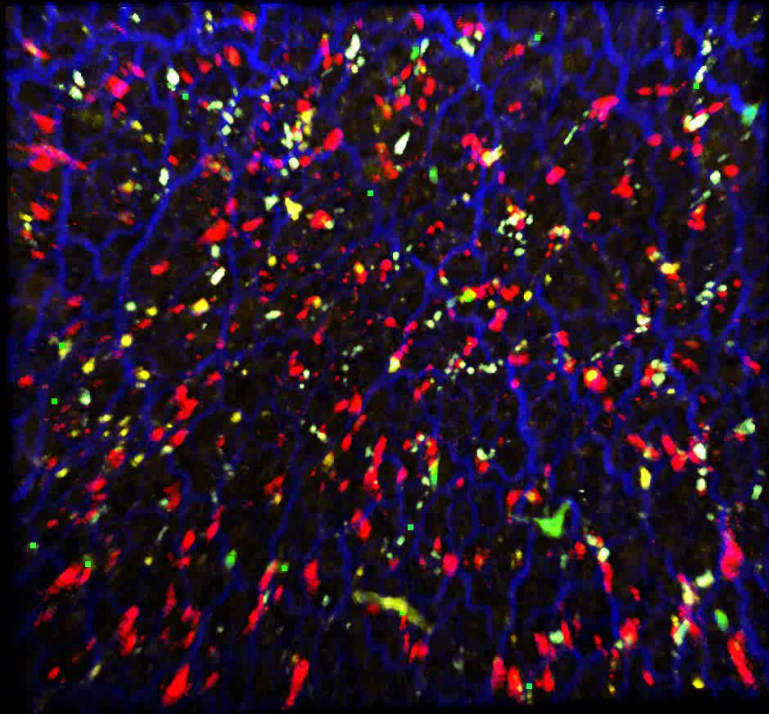
CCR2⁺ inflammatory monocyte recruitment after acetaminophen overdose

control

CCR2-eGFP
(reporter)

Latex particles
SHG (collagen)

APAP 9 hours



50 μ m

0.000 Speed 0.400

Time
0.00.00.0000

50 μ m

0.000 Speed 0.400

Time
0.00.00.0000

- progressive accumulation of CCR2⁺ monocytes at 9-12h after APAP injury

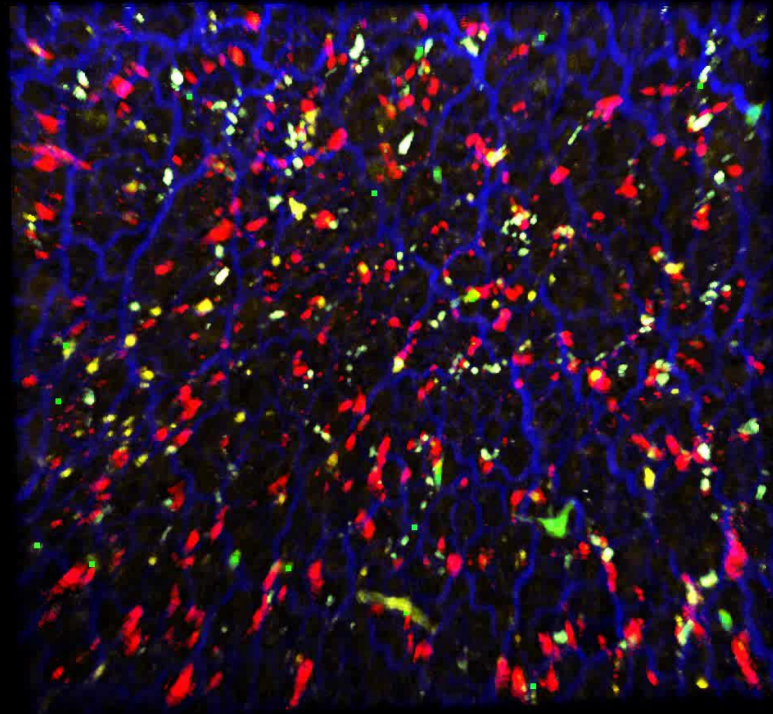
CCR2⁺ inflammatory monocyte recruitment after acetaminophen overdose

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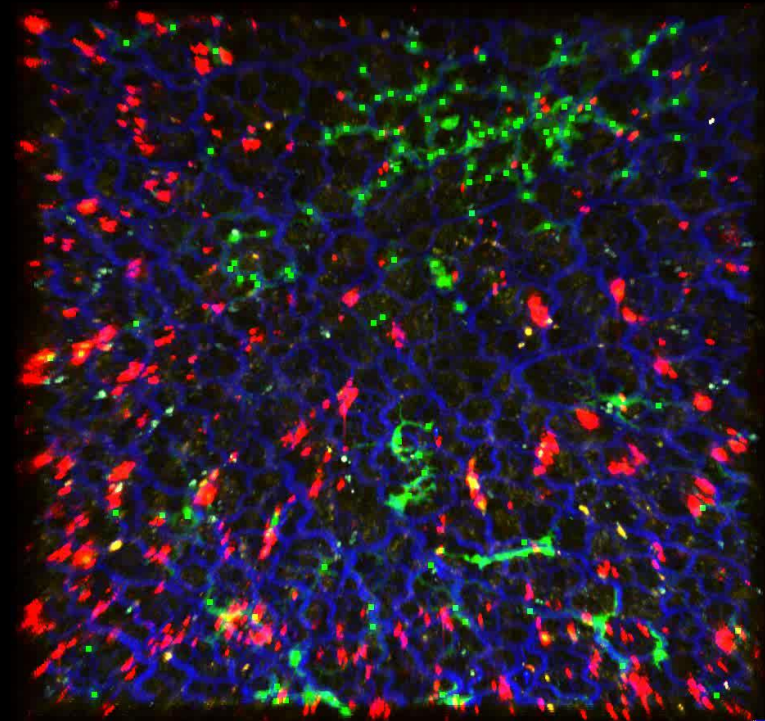
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0.00.00.0000

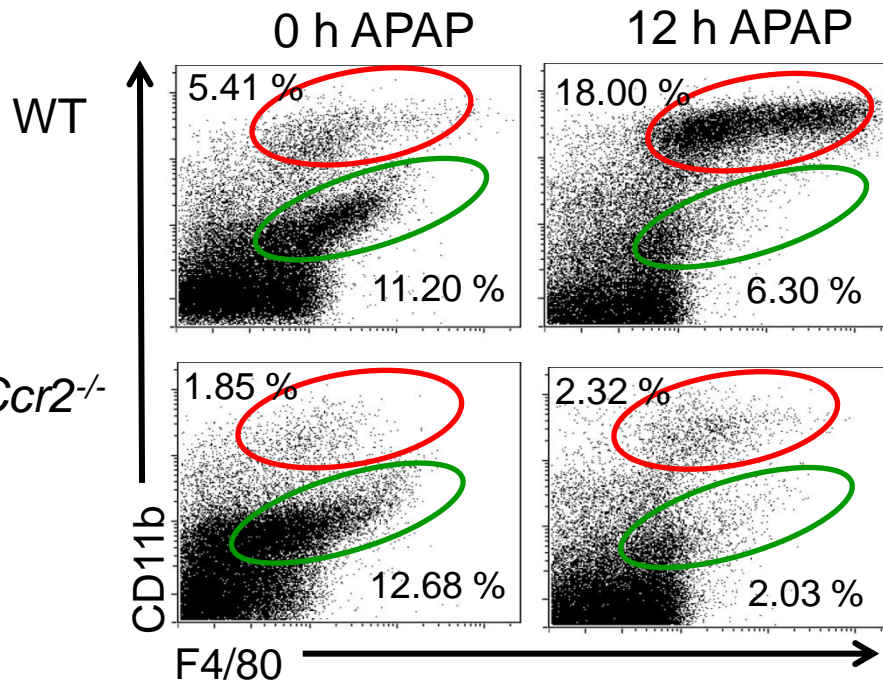


0.000 Speed 0.400

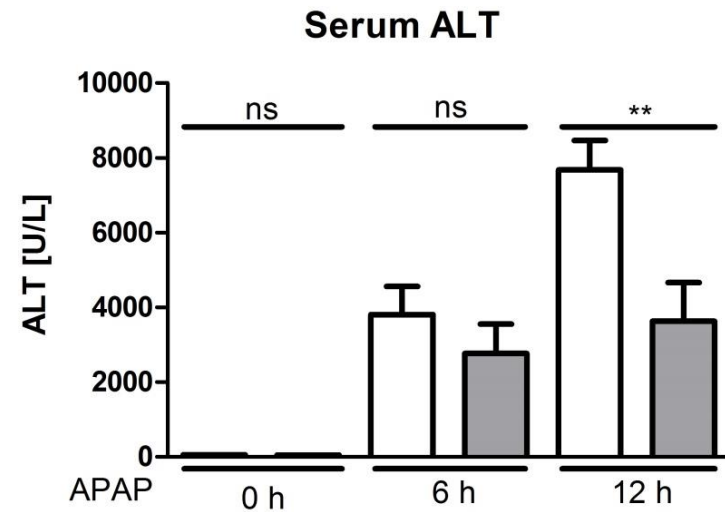
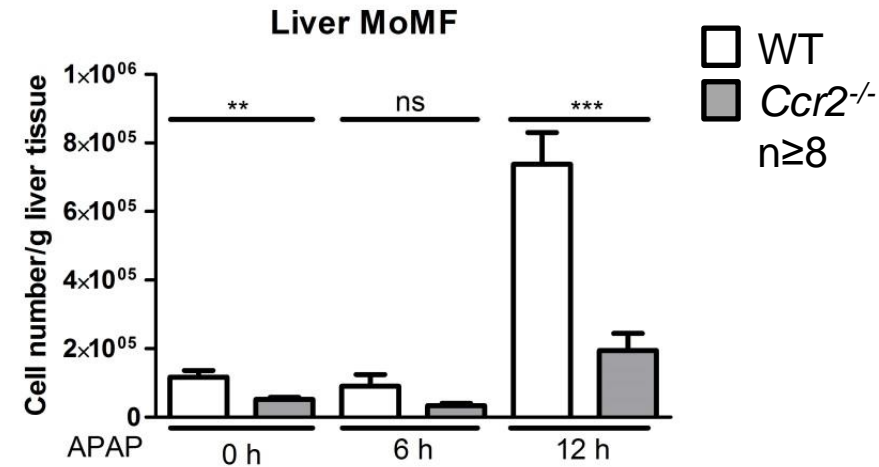
Time
0.00.00.0000

- progressive accumulation of CCR2⁺ monocytes at 9-12h after APAP injury

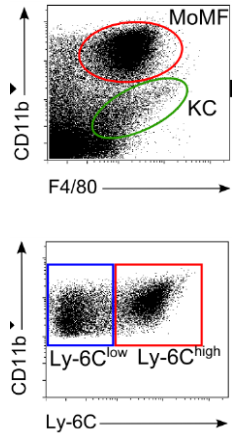
Ccr2^{-/-} mice are protected in the early phase of acetaminophen induced liver injury



Monocyte-derived Macrophages (MoMF)
Kupfer Cells (KC)



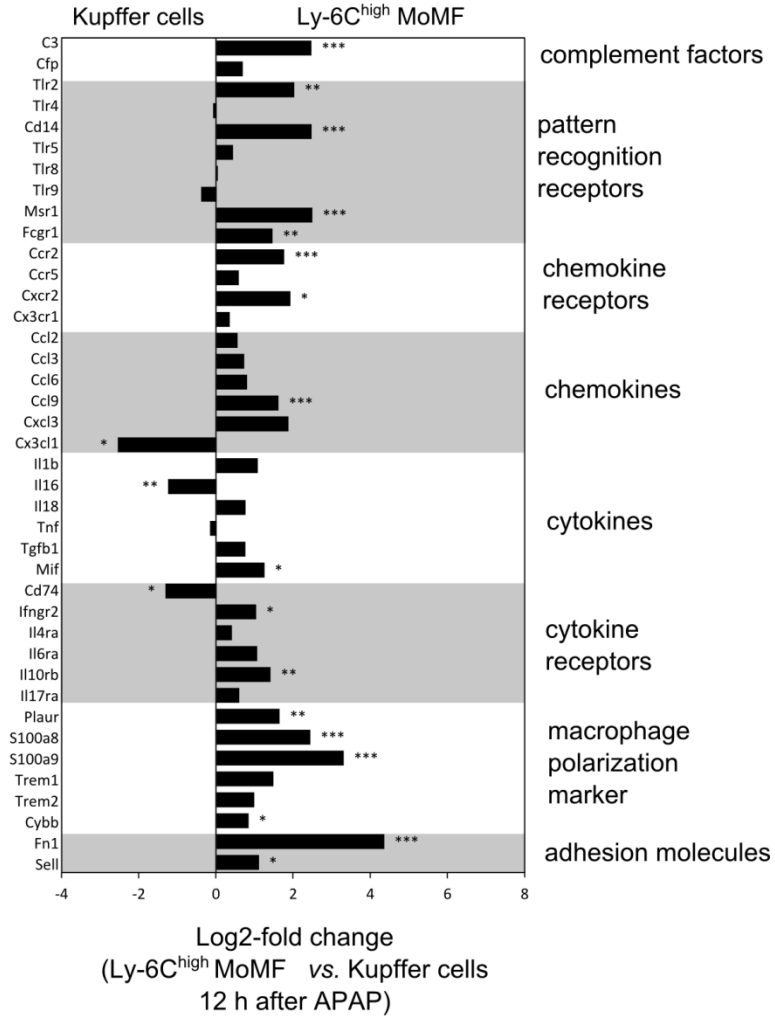
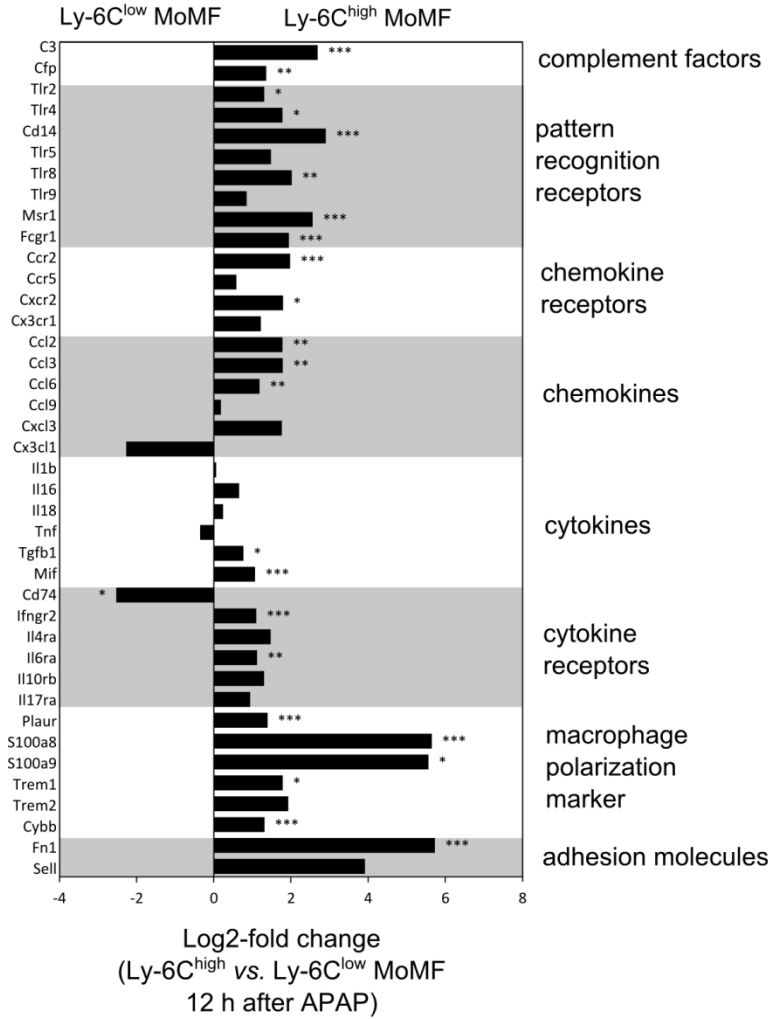
CCR2⁺ monocyte-derived macrophages have an inflammatory phenotype



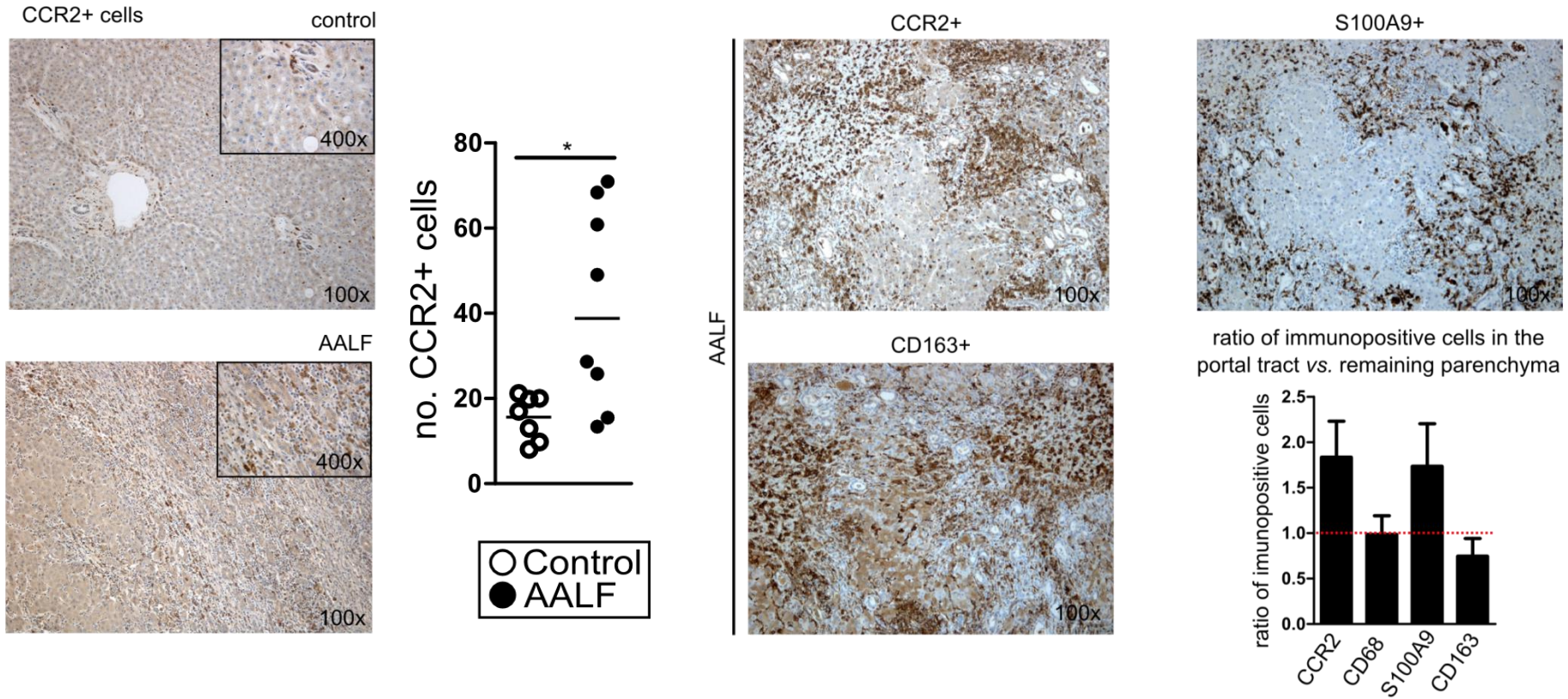
FACS sorting



Nanostring[®]
analysis for 561
gene targets
associated with
inflammation



Inflammatory CCR2⁺ macrophages in human acetaminophen acute liver failure

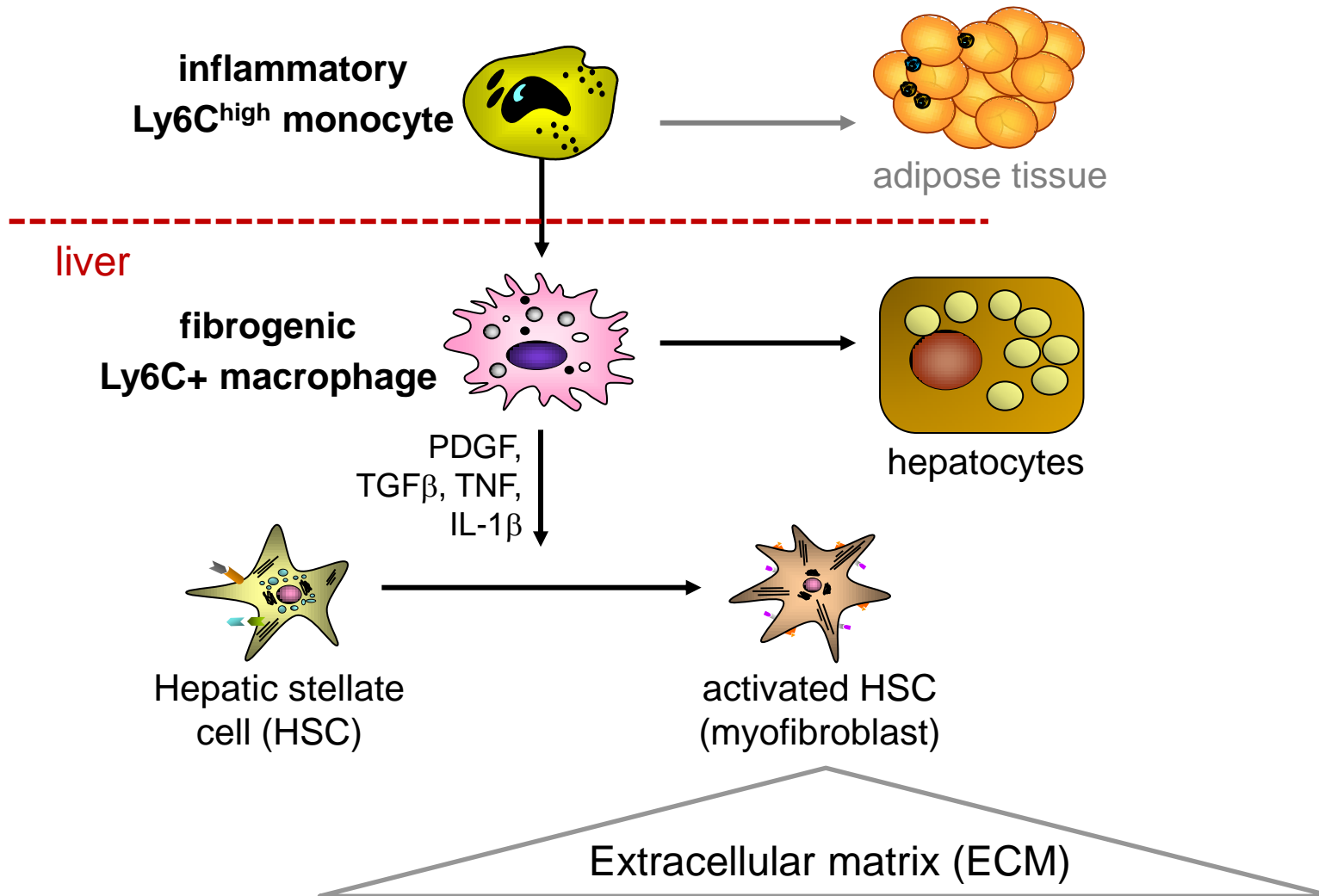


Mossanen JC, Krenkel O / Tacke F. *Hepatology* 2016; 64(5):1667-1682
 similar data by Antoniadis CG / Wendon J. *Hepatology* 2012; 56(2):735-46

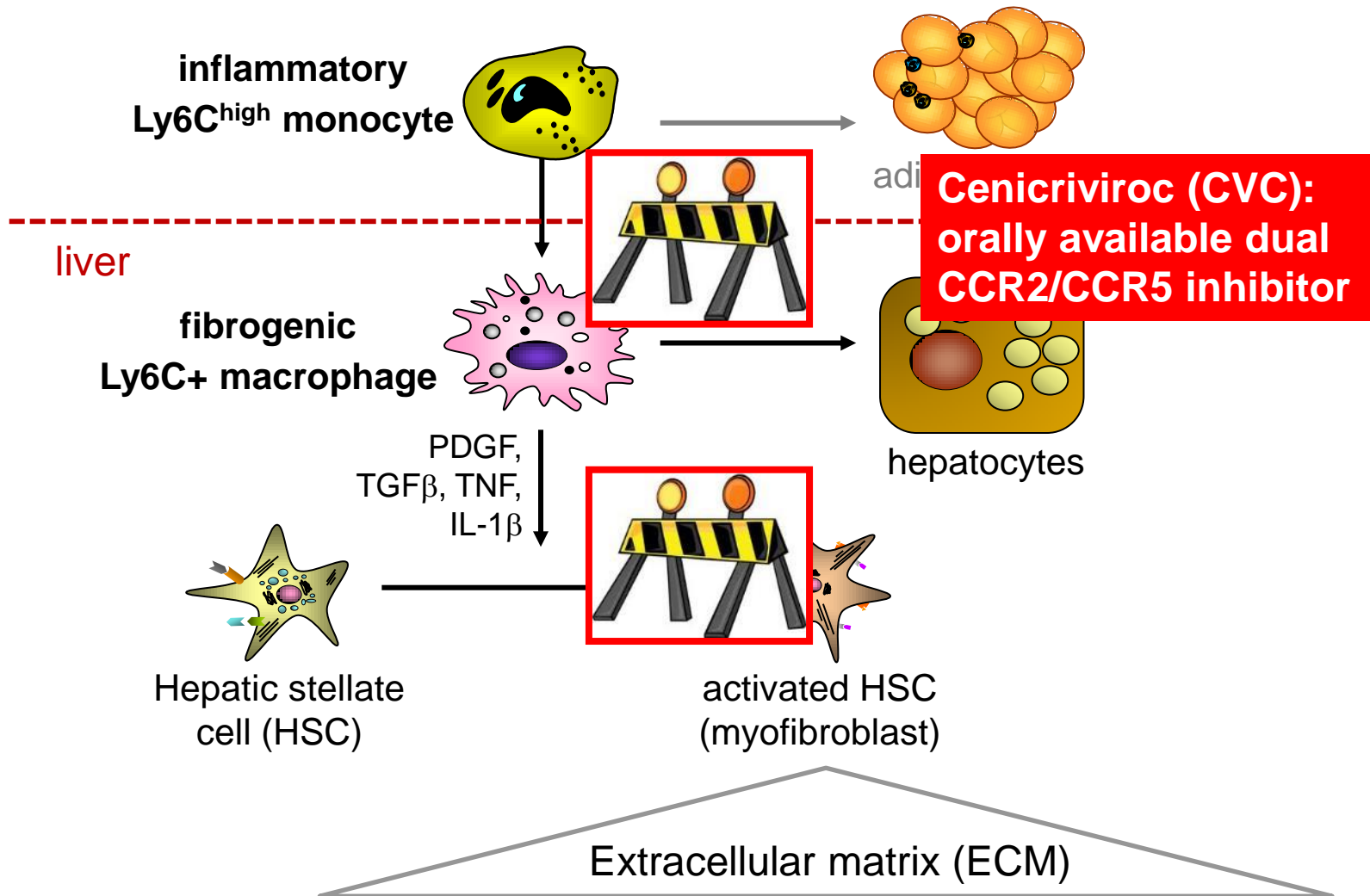
Monocytes and macrophages in liver diseases

- **Acute liver injury**
- **NASH and fibrosis**
- **Immunometabolism**

Monocyte / macrophage subsets during regression of liver fibrosis



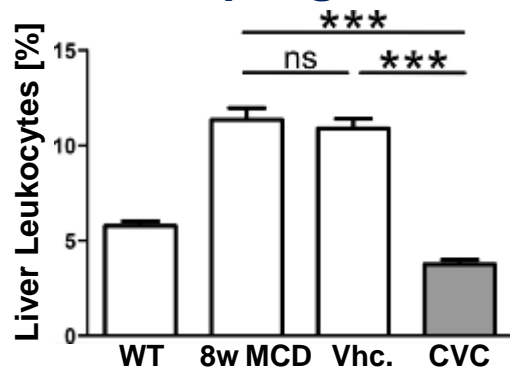
Monocyte / macrophage subsets during regression of liver fibrosis



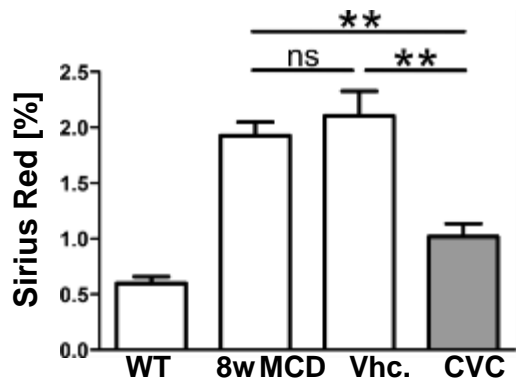
Chemokine receptor CCR2/5-inhibitor CVC in experimental NASH and fibrosis

Mouse model of NASH (MCD)

liver macrophages reduced



liver fibrosis reduced



clinical trial phase 2b (*ongoing*)

patients with NASH + fibrosis (n=289)
enriched for co-morbidities

CVC 150mg
(n=145)

Placebo
(n=144)

liver biopsy after 1 year

CVC 150mg
(n=145)

CVC 150
(n>63)

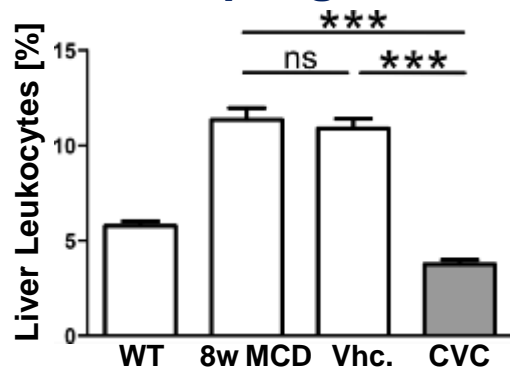
Placebo
(n>63)

liver biopsy after 2 years

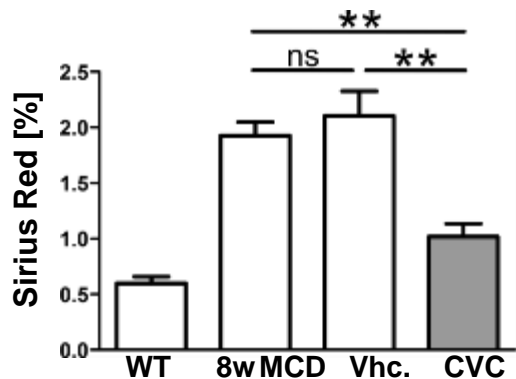
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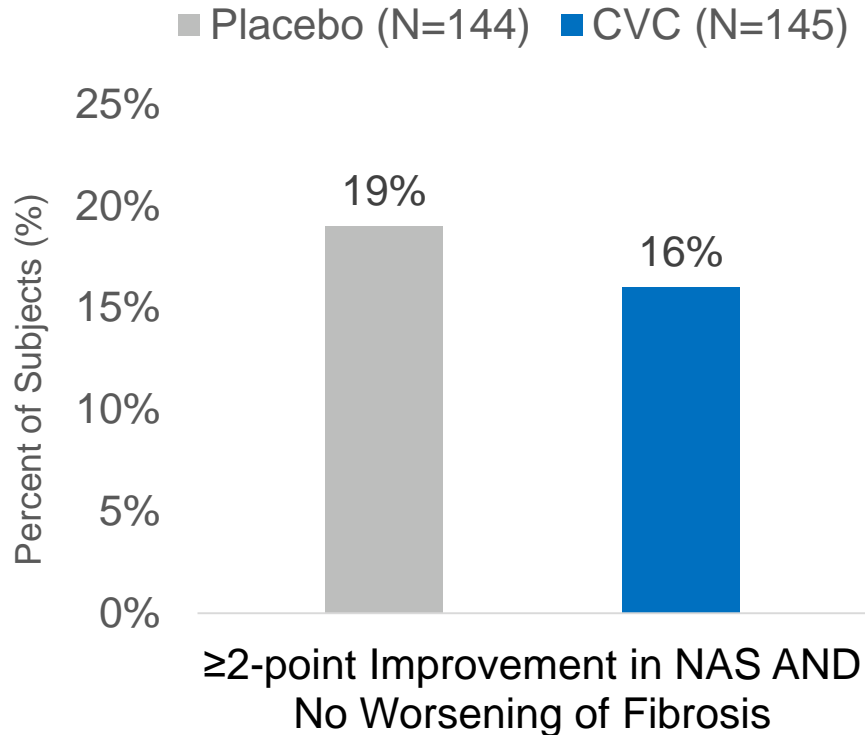
interim analysis after 1 year treatment:

- 126 + 126 sufficient paired biopsies
- baseline characteristics:
33% F1, 29% F2, 38% F3 fibrosis
52% type 2 diabetes, BMI ~34 kg/m²

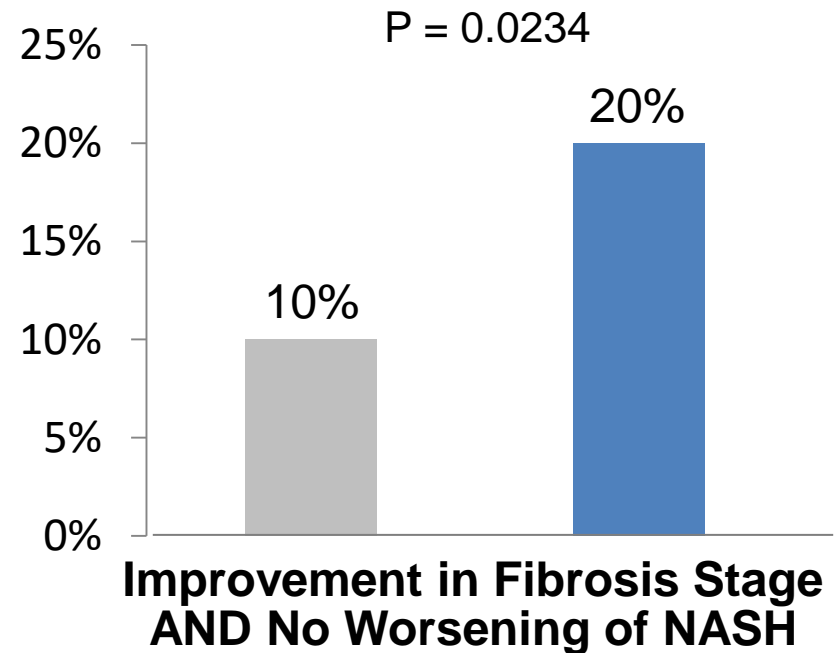
Sanyal A et al. #LB-1, AASLD 2016

Chemokine receptor CCR2/5-inhibitor CVC in patients with NASH and fibrosis

Steatohepatitis-Score



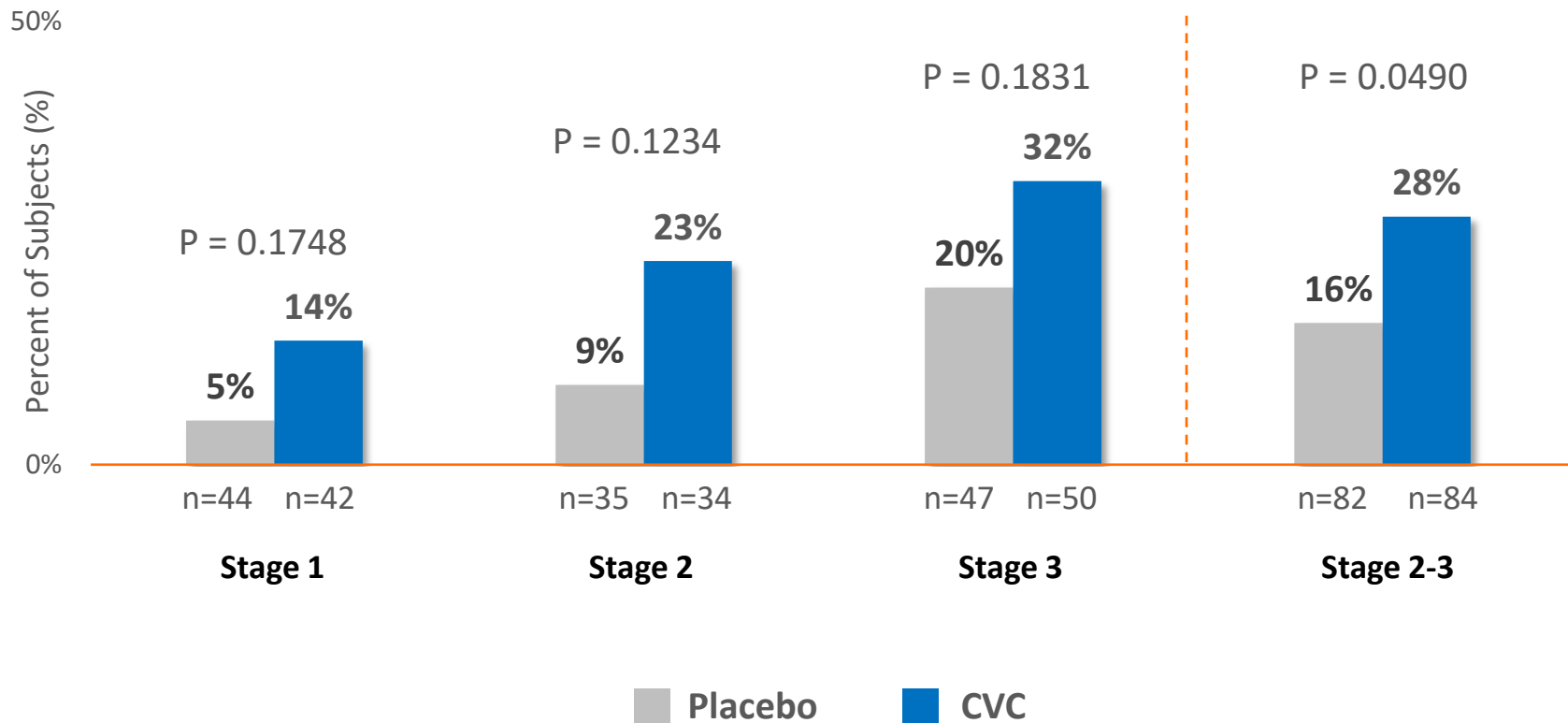
Fibrosis-Score



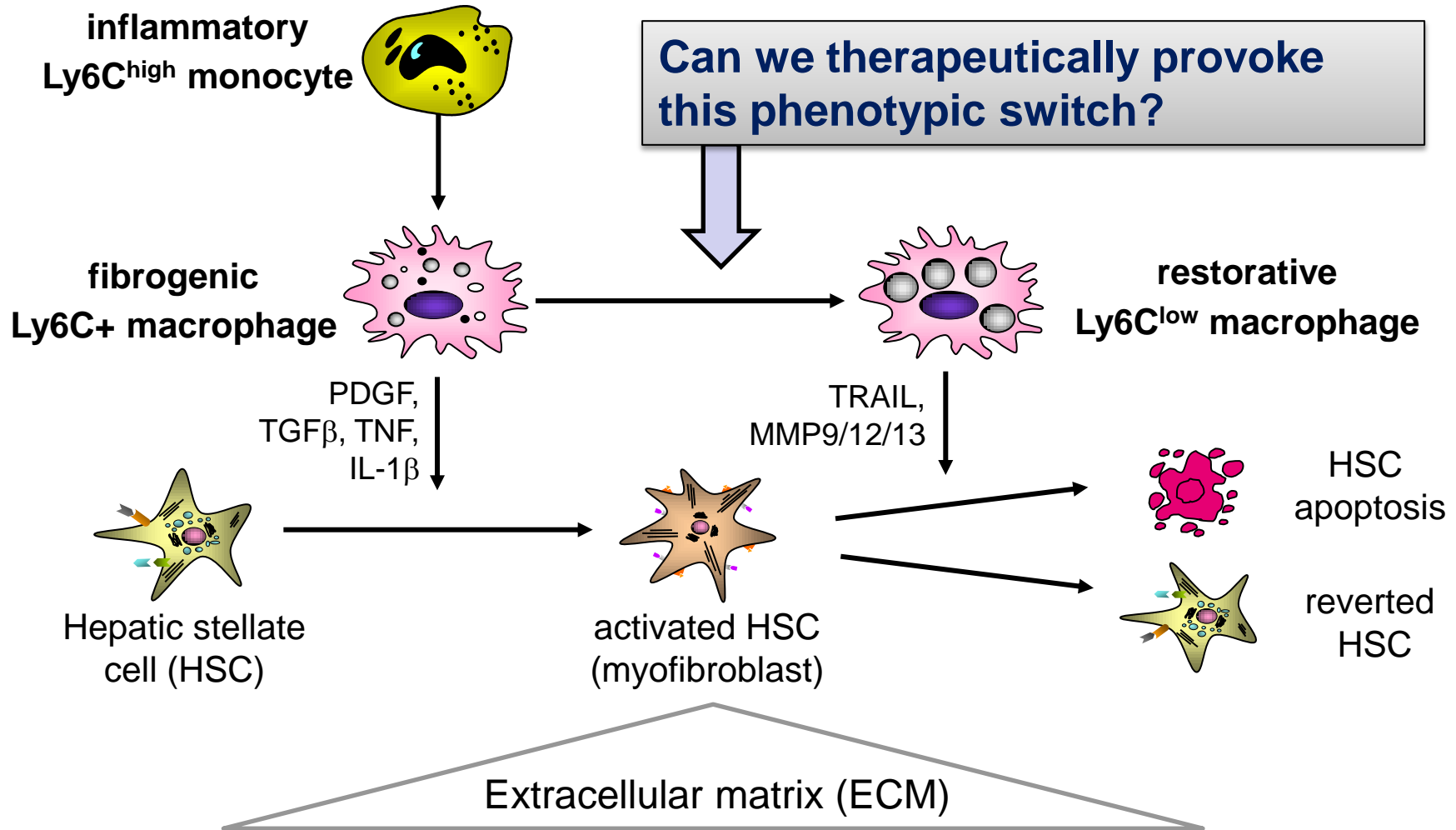
NASH resolution: 8% vs. 6% (n.s.)

Chemokine receptor CCR2/5-inhibitor CVC in patients with NASH and fibrosis

Response by Baseline Fibrosis Stage (mITT)



Monocyte / macrophage subsets during regression of liver fibrosis



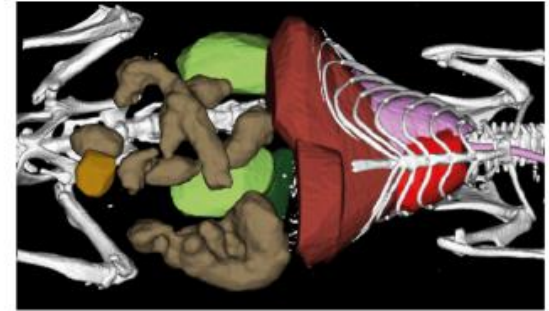
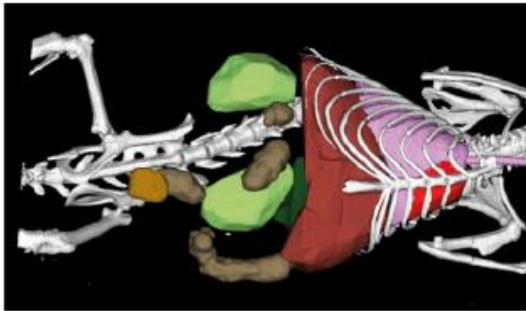
Targeting macrophages in liver inflammation and fibrosis

Microbubbles

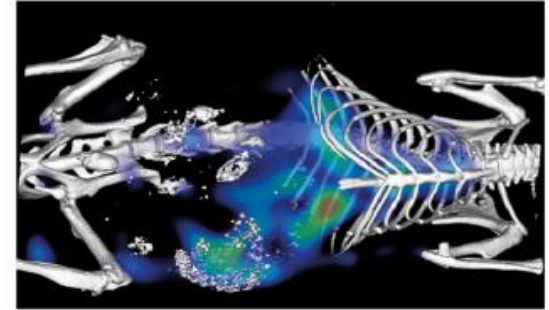
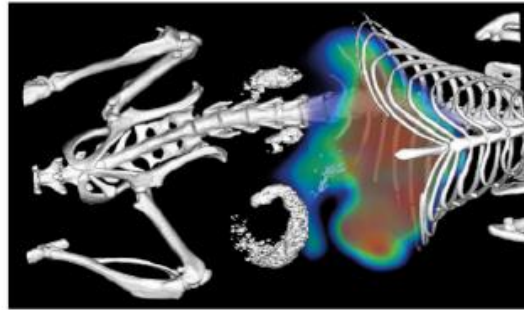
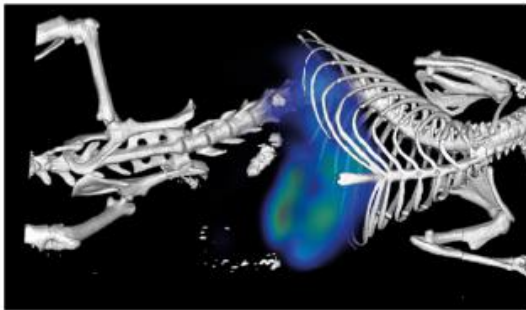
Liposomes

Polymers

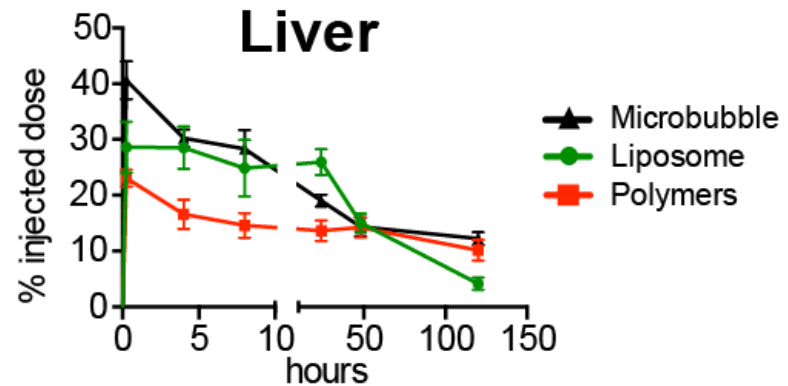
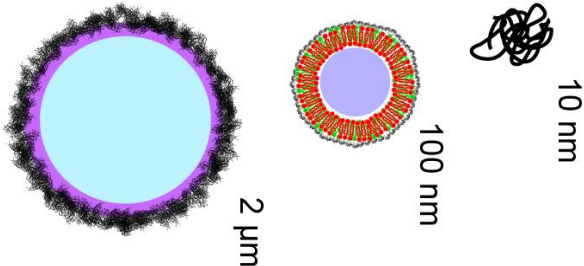
CT+Seg.



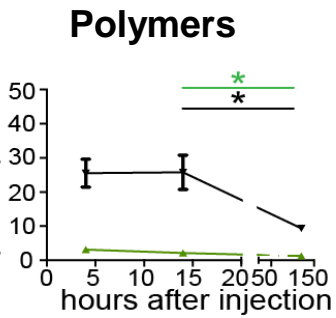
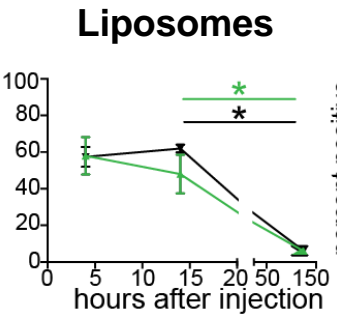
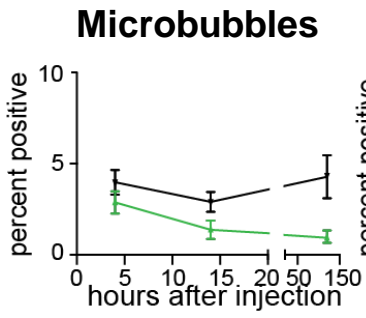
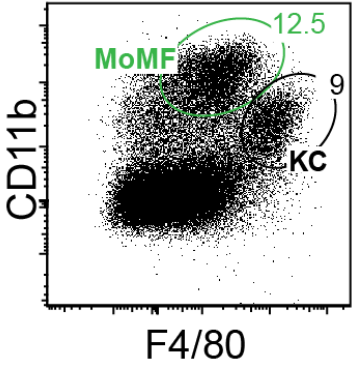
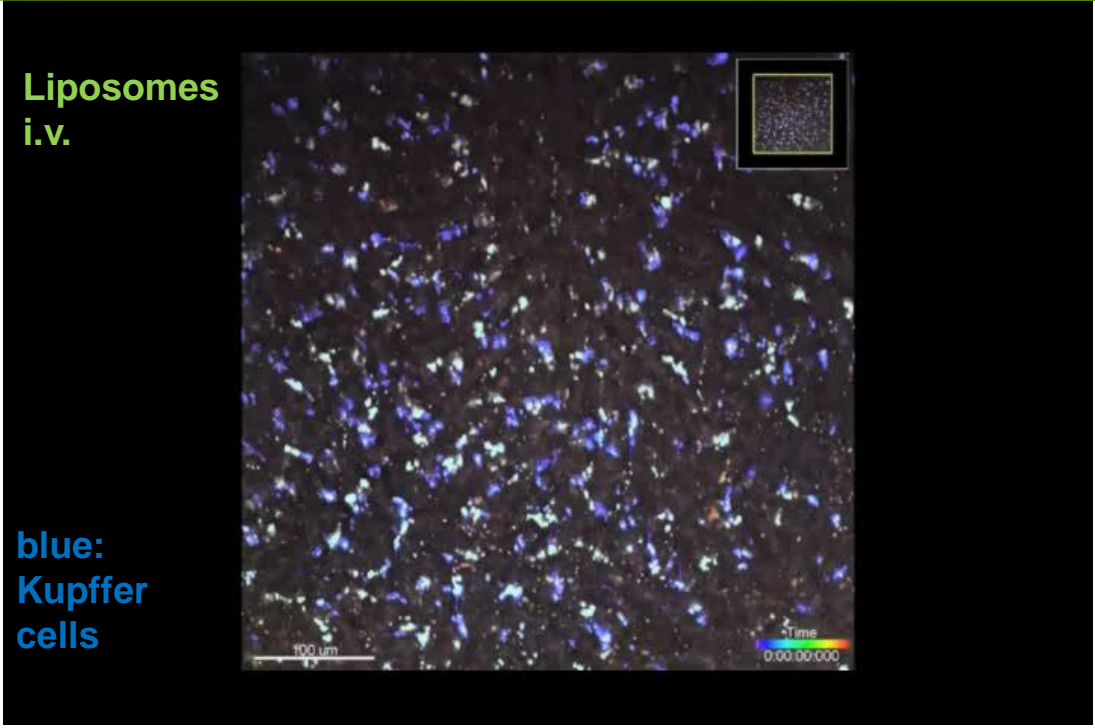
CT+FMT



Microbubbles Liposomes Polymers



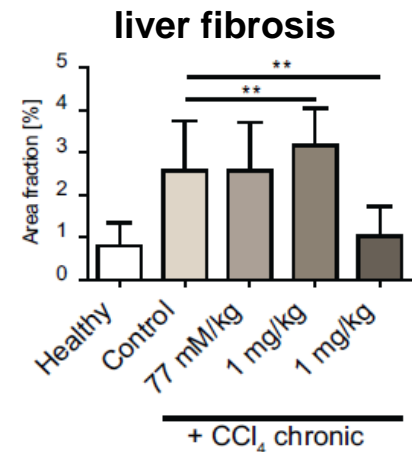
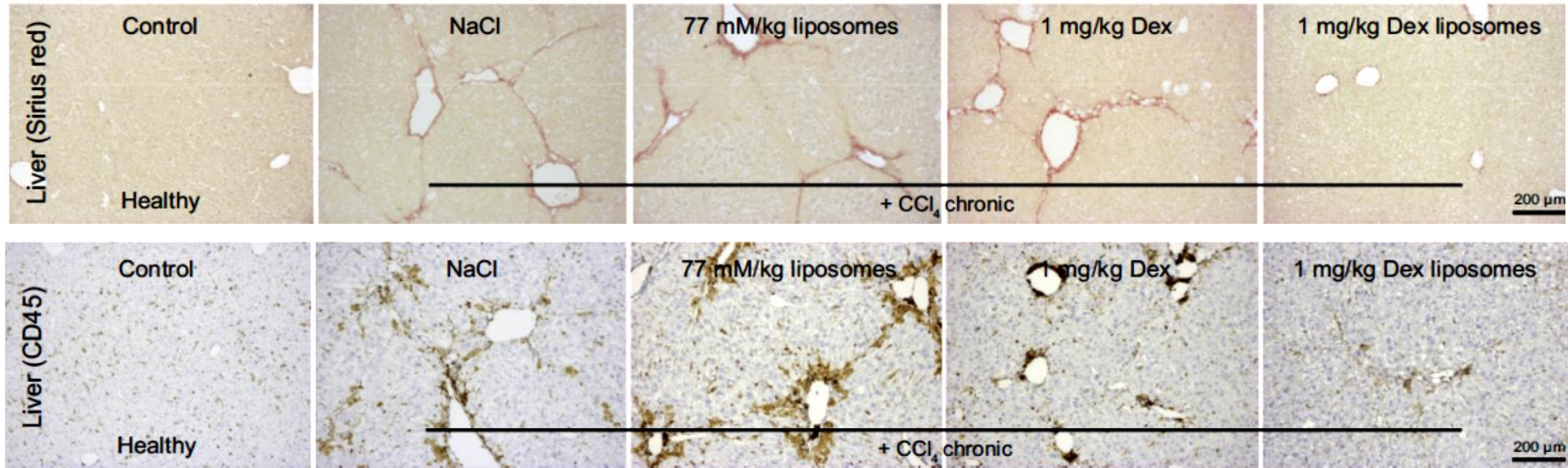
Targeting macrophages in liver inflammation: Drug delivery systems



Kupffer cells
Monocyte-derived macrophages

Targeting macrophages in liver inflammation: *Proof-of-concept* in fibrosis

Dexamethasone-loaded Liposomes

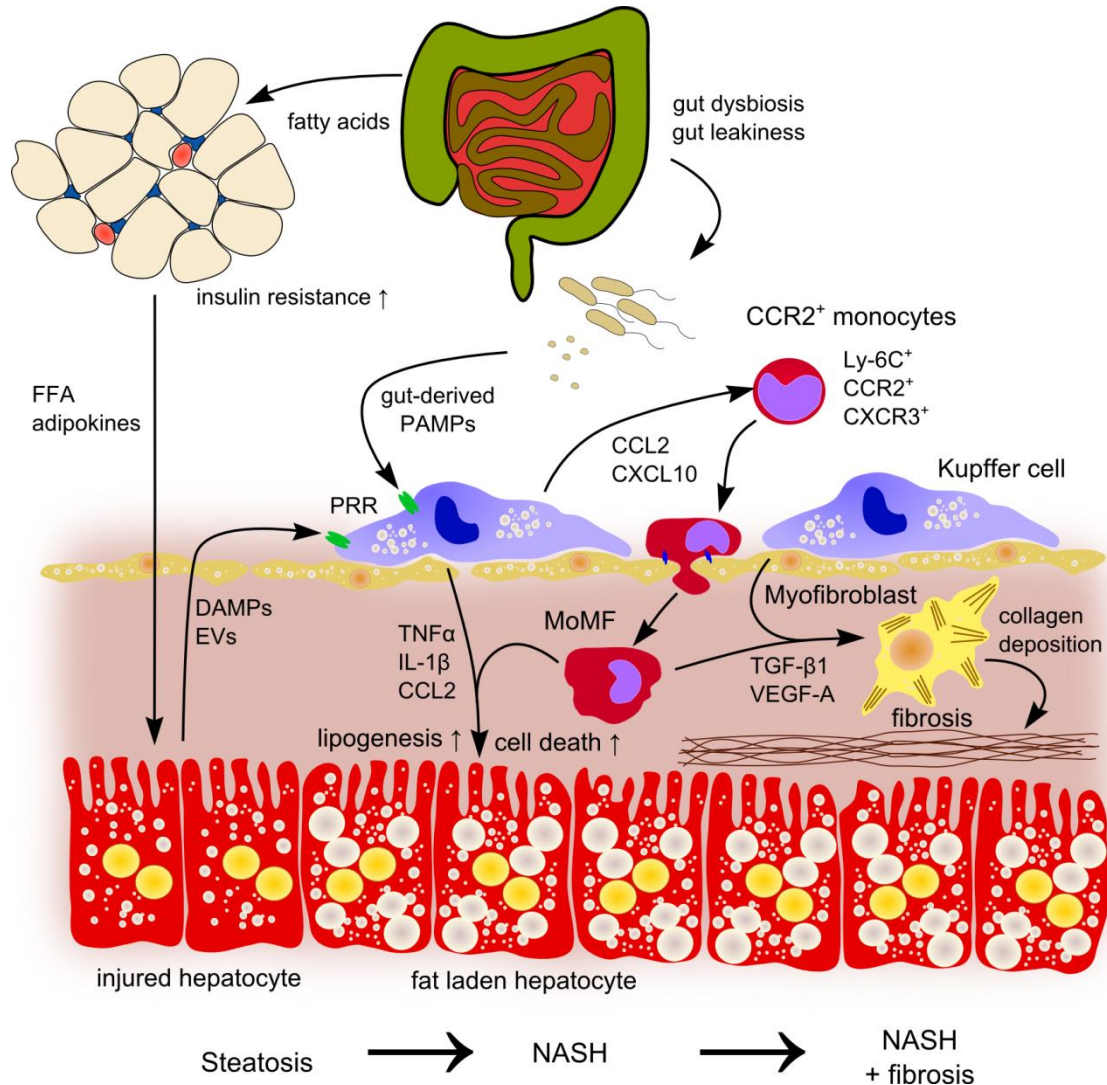


Bartneck M / Tacke F, *Nanomedicine* 2014
Bartneck M / Tacke F, *Biomaterials* 2015
Bartneck M / Tacke F, *ACS Nano* 2012
Topuz F / Tacke F, *Biomacromolecules* 2017

Monocytes and macrophages in liver diseases

- **Acute liver injury**
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Immunometabolism 1: Integration of metabolic signals by macrophages

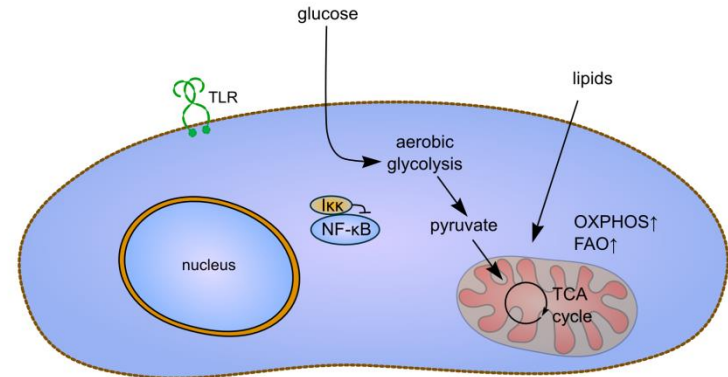


Immunometabolism 2: Adaptation of cellular metabolism by macrophages

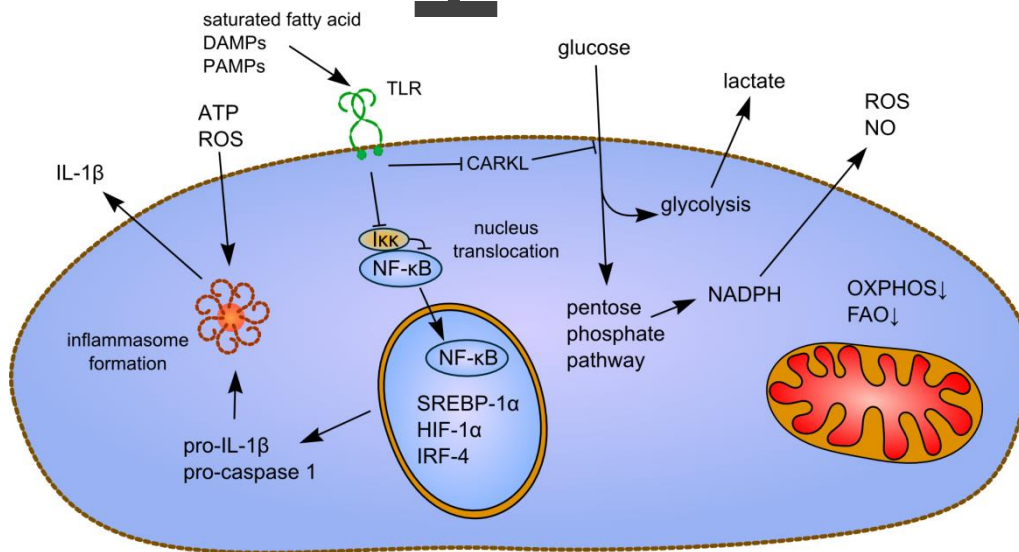
Manifold potential targets...

- PPAR $\alpha / \gamma / \delta$
- ASK1

Macrophage metabolism in homeostasis



Macrophage metabolism upon activation



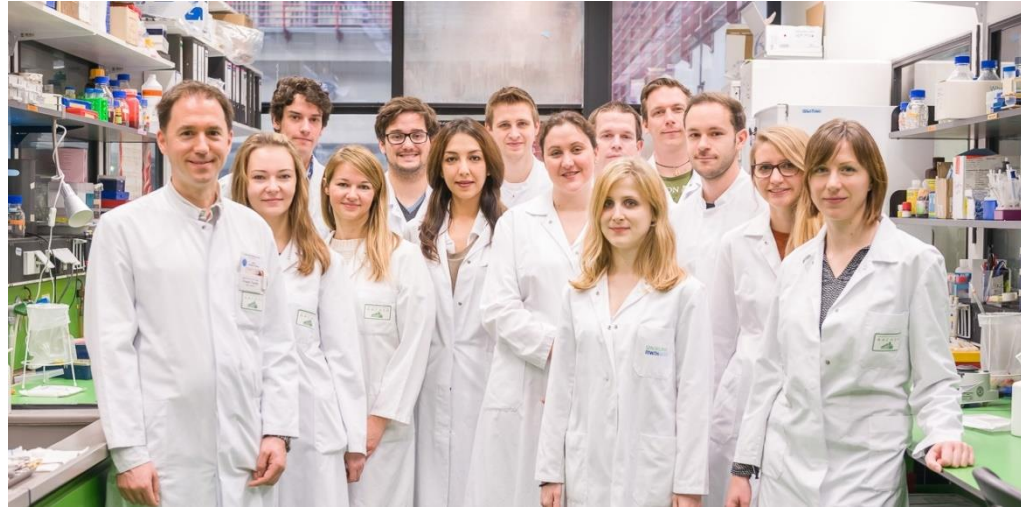
- *NF- κ B activation*
- *glycolysis*
- *ROS and NO production*
- *distinct transcription factors*
- *inflammasome formation*
- *cytokine production*

Thank you!

Tacke Lab

Dr. Jana Mossanen
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 Oliver Krenkel
 Aline Roggenkamp
 Tobias Püngel
 Peter Schrammen
 Can Ergen
 Marlene Kohlhepp

Q3 cell isolation
 Sibille Sauer-Lehnen
 Carmen Tag



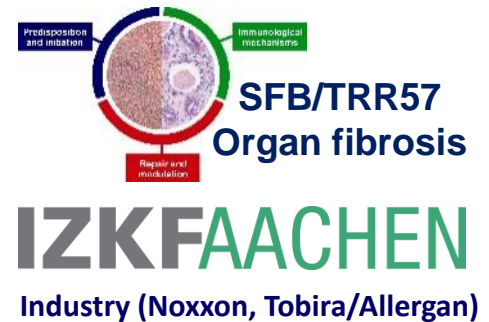
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 Ulf Neumann

collaborators

Tania Roskams
 Olivier Govaere
 Quentin Anstee
 Eric Lefebvre
 Florent Ginhoux
 Steffen Jung
 Jo van Ginderachter
 Gwen Randolph

Funding



Role of macrophages in disease development and progression of NASH

- **Monocyte** and **macrophage subsets** impact inflammation, hepatocyte injury, hepatic stellate cell activation, angiogenesis, but also resolution of injury, in mice and men
- Hepatic **macrophage heterogeneity** includes origin, differentiation/ polarization, immunological properties and functions in disease progression/regression
- Therapeutic application of the **CCR2/CCR5 inhibitor CVC** in mice ameliorates steatohepatitis and fibrosis without impairing tissue repair, supporting the therapeutic potential in patients with NASH
- **Important lines of research** in the field: **regulation** and **balance** of pro- and antiinflammatory subsets, novel **imaging** approaches to dissect immune cell subsets, immune mechanisms in transition from chronic inflammation to **cancer**, targeting mechanisms by **nanoparticles**, **translation** into clinics (biomarkers + therapy)