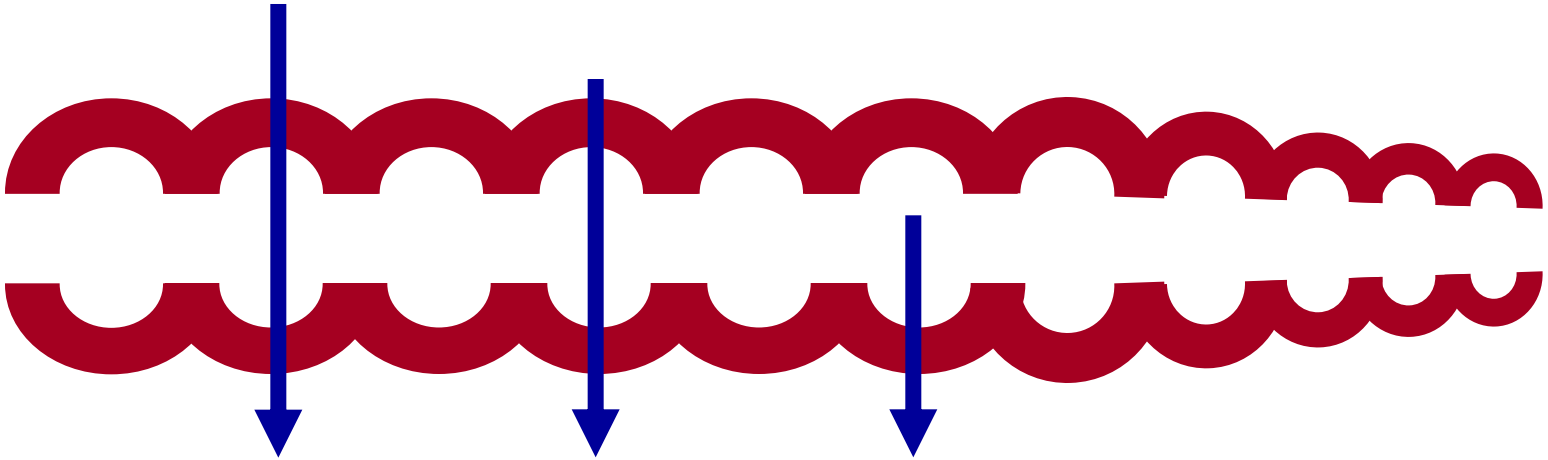
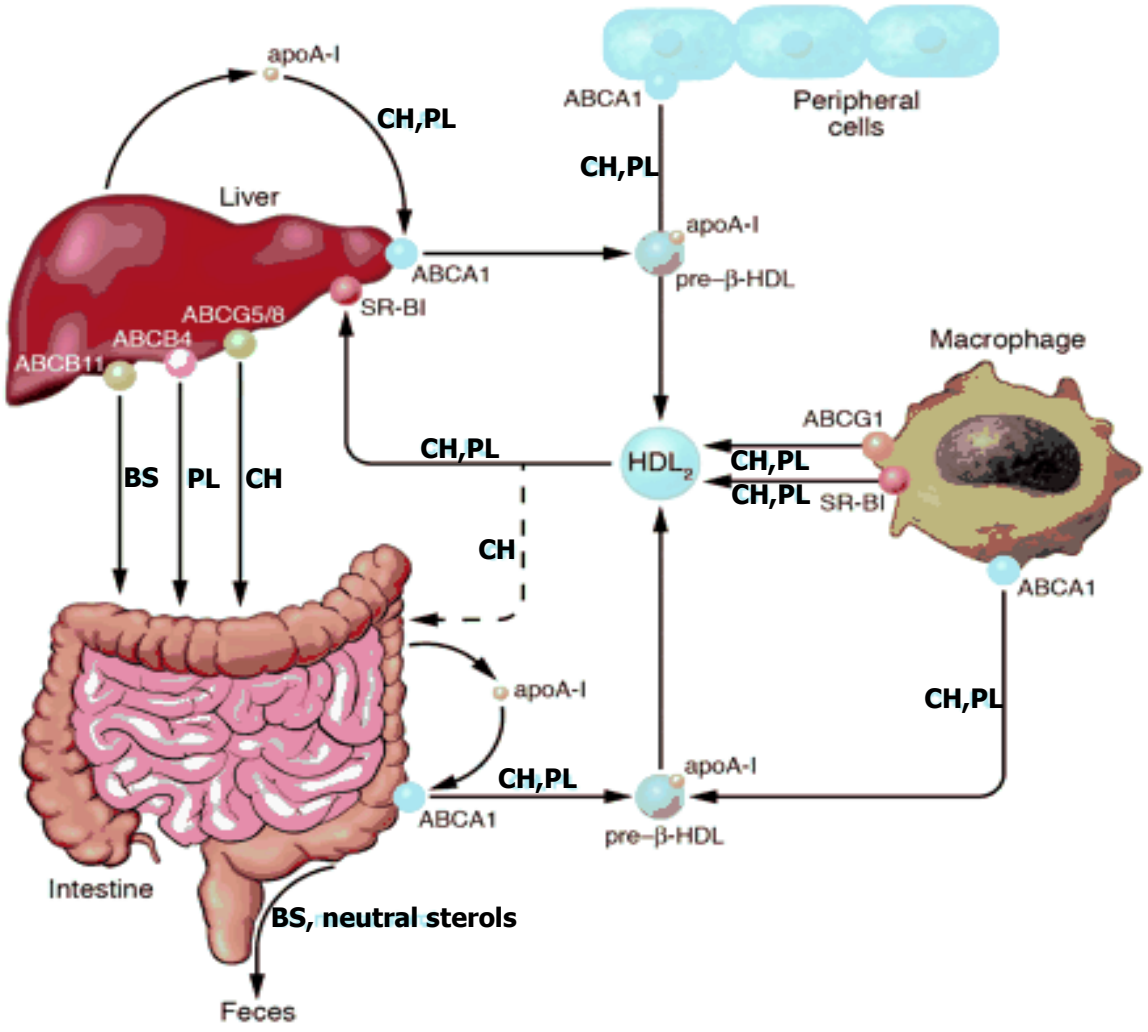
Targeting cholesterol excretion



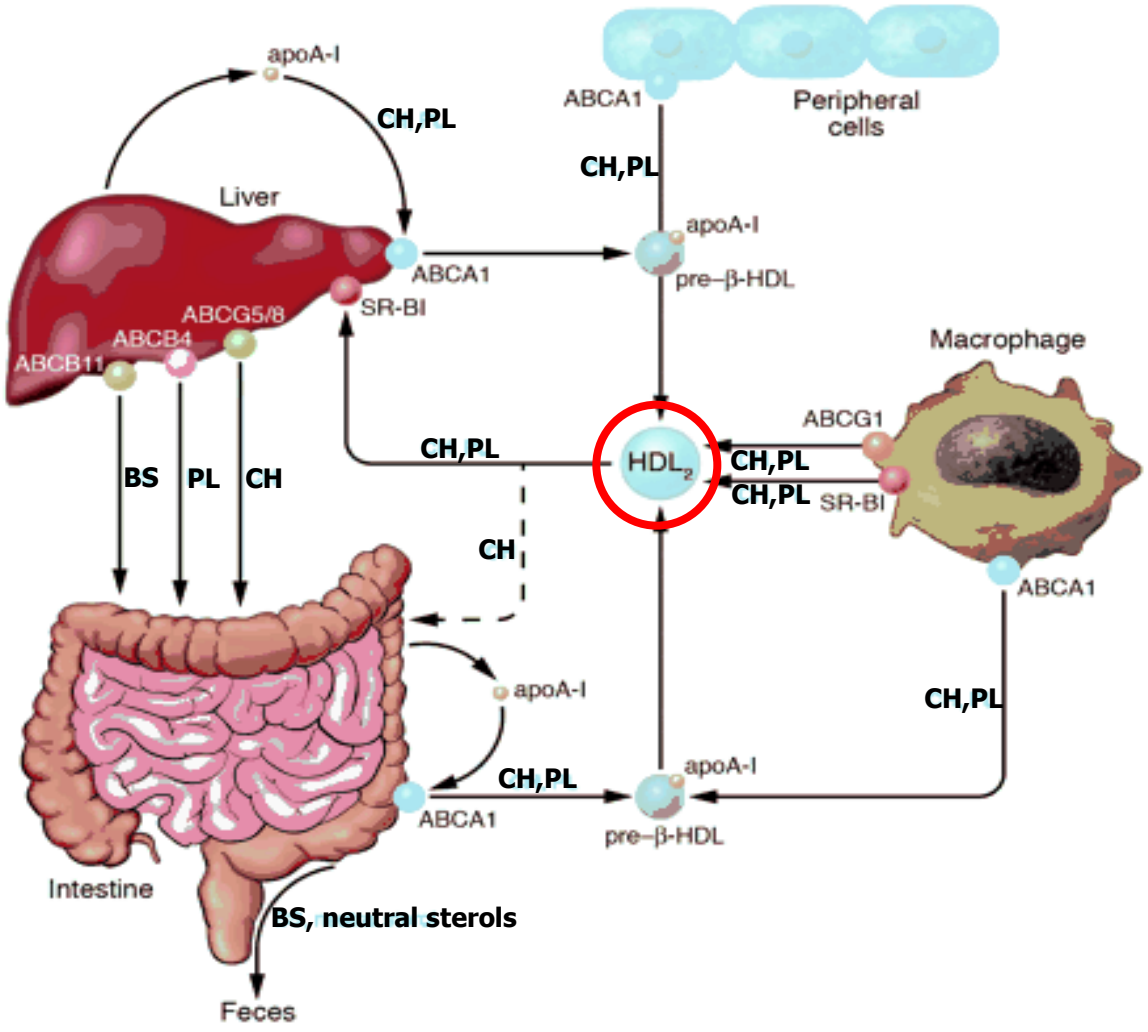
Dutch Atherosclerosis Society

Lily Jakulj, MD

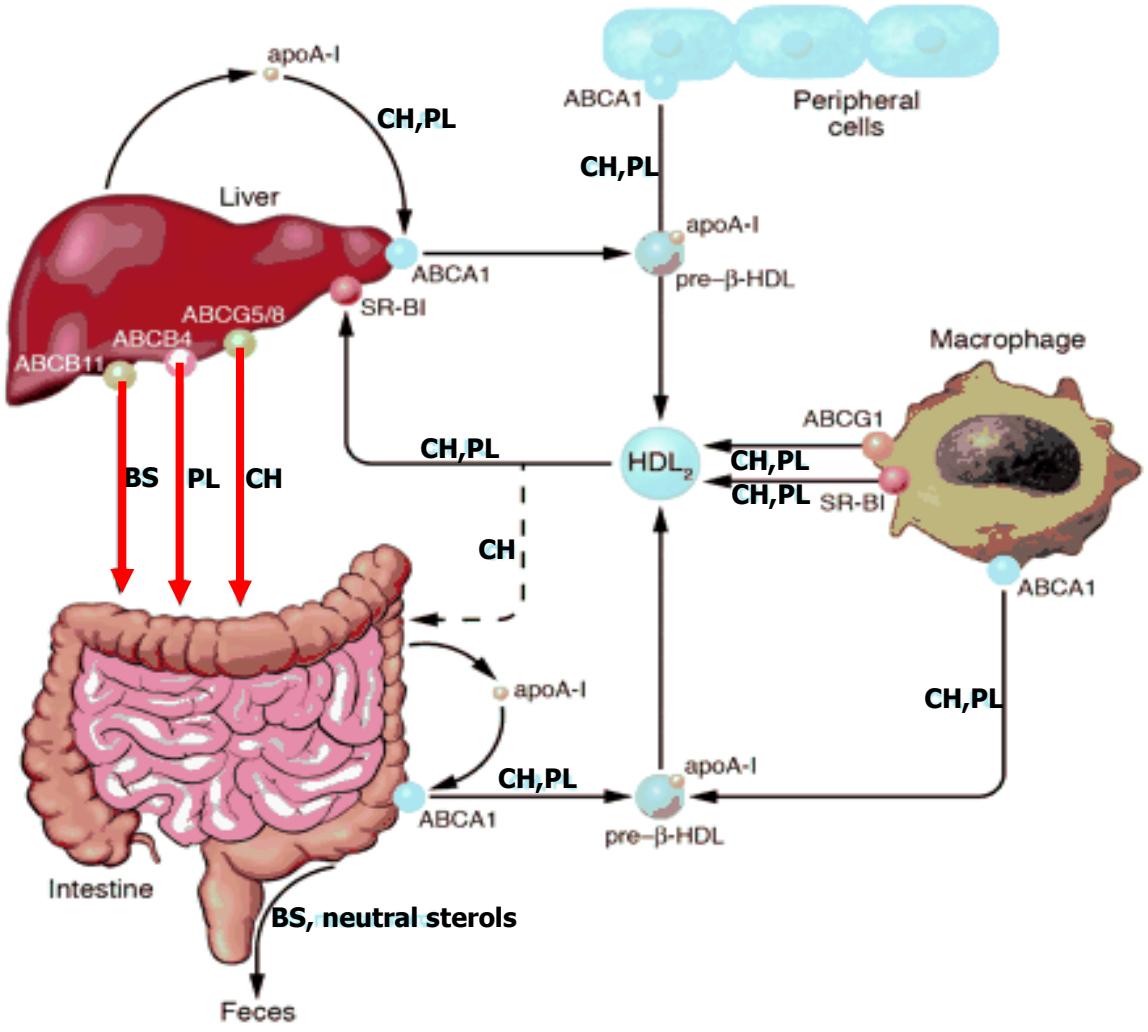
Reverse cholesterol transport



1. Plasma HDL-c determines RCT



2. Obligate role of bile in RCT

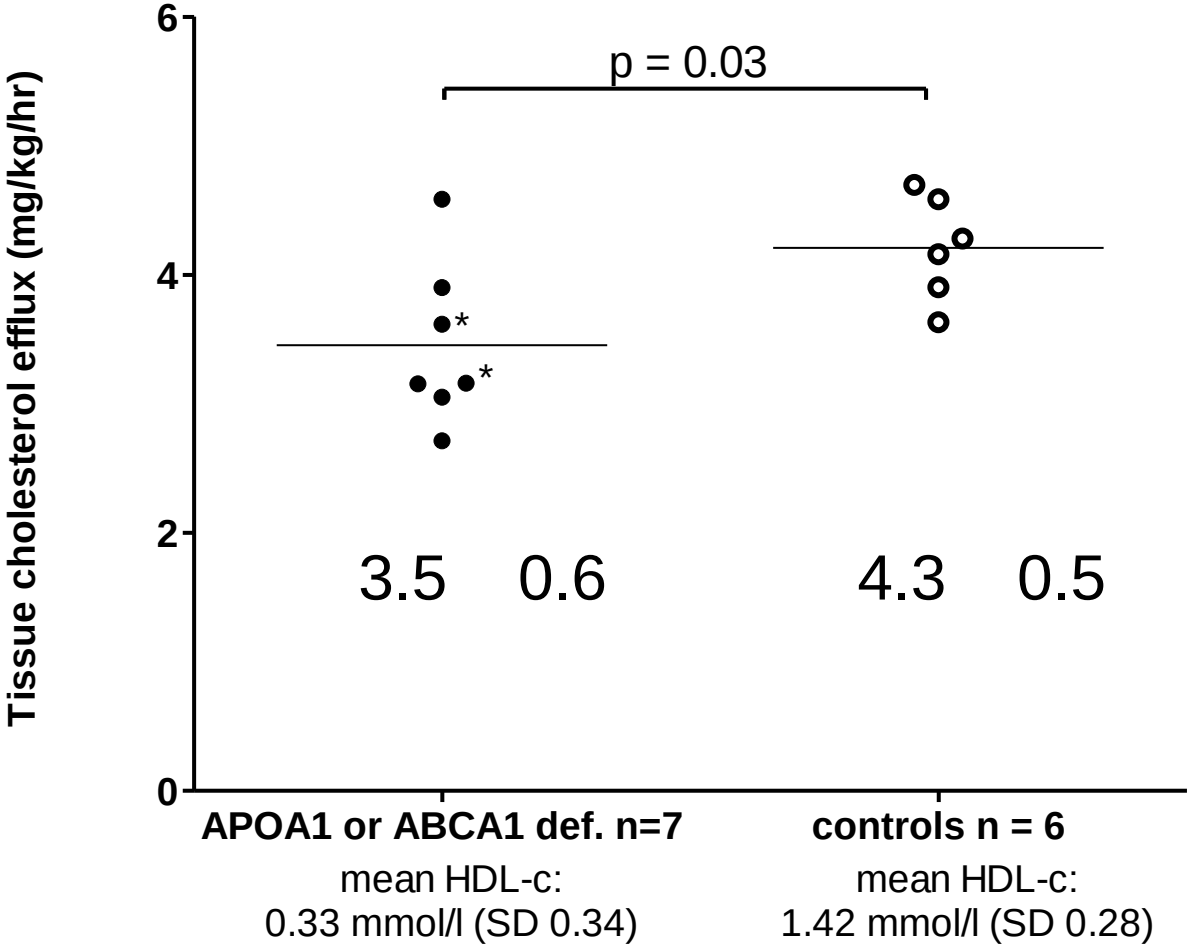


1. Plasma HDL-c and RCT

- Biliary and fecal cholesterol excretion not impaired in ABCA1-/- mice [1,2]
- rHDL increased plasma HDL-c and centripetal flux to the liver, but not FSE in ABCA1-/- mice [3]
- Upregulation of individual steps in RCT did not affect FSE in normolipidemic mice [4]
- Human FSE studies conflicting

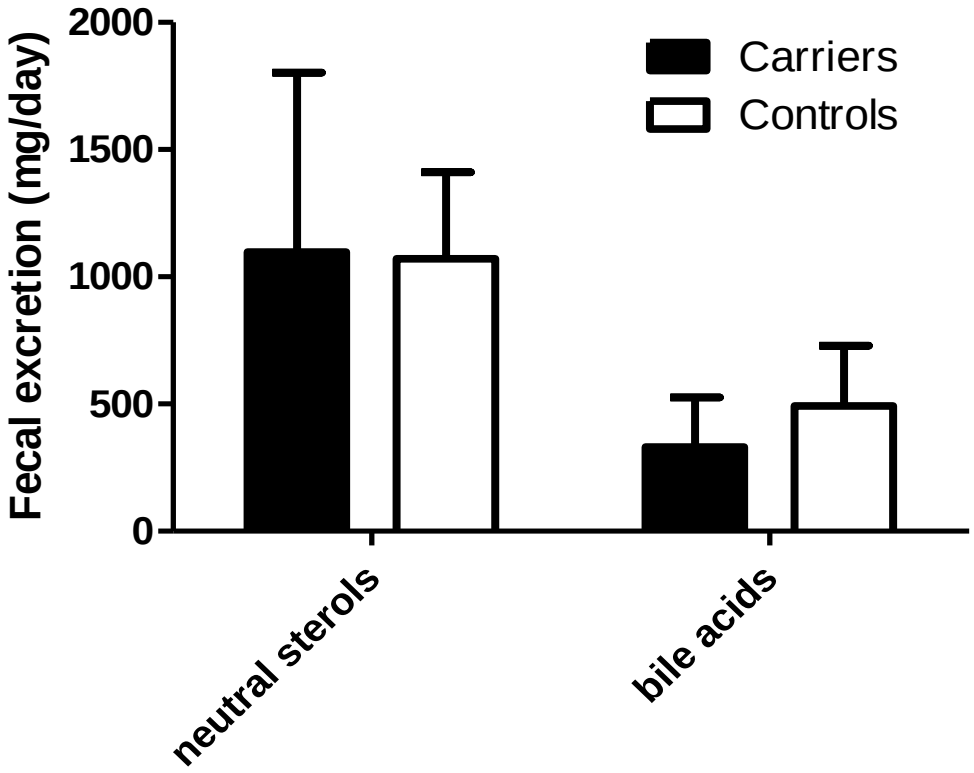
1. Groen, JCI 2001
2. Xie, JLR 2009
3. Jolley, JLR 1998
4. Alam, JBC 2001

Impaired *in vivo* tissue cholesterol efflux in subjects with genetically low HDL-c



Holleboom, Jakulj, Groen, Stroes, unpublished

FSE is equal in low HDL-carriers and controls

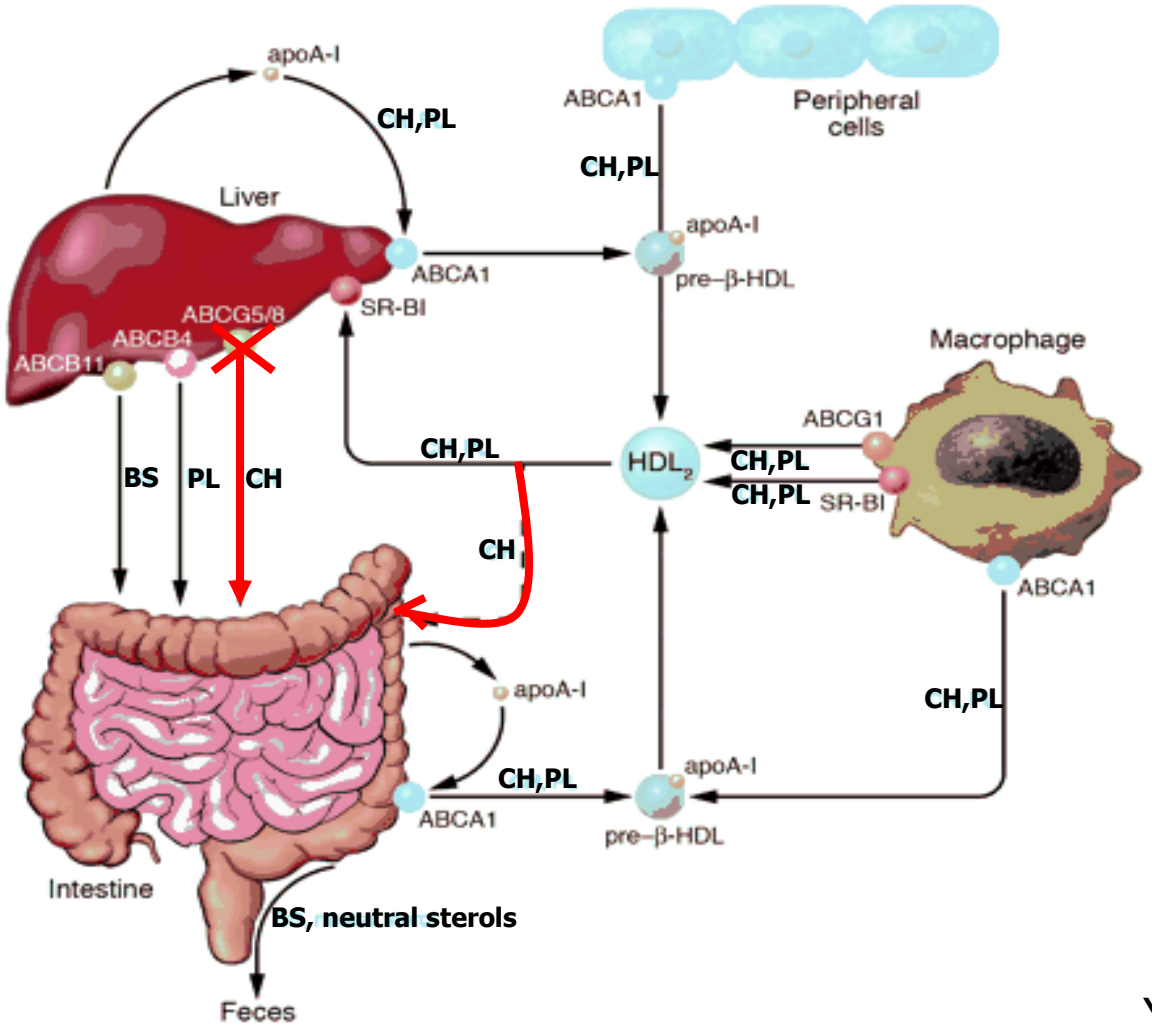


Holleboom, Jakulj, Groen, Stroes, unpublished

1. Plasma HDL-c determines RCT

- Plasma HDL-c does not adequately reflect RCT in mice and humans with isolated low HDL-c
- Despite impaired TCE in carriers of mutations in *APOA1* or *ABCA1*, compensating mechanisms exist
- Fecal sterol excretion, the obligate endpoint of RCT may depend on alternative (non-HDL?) pathways

2. Obligate role of bile in RCT

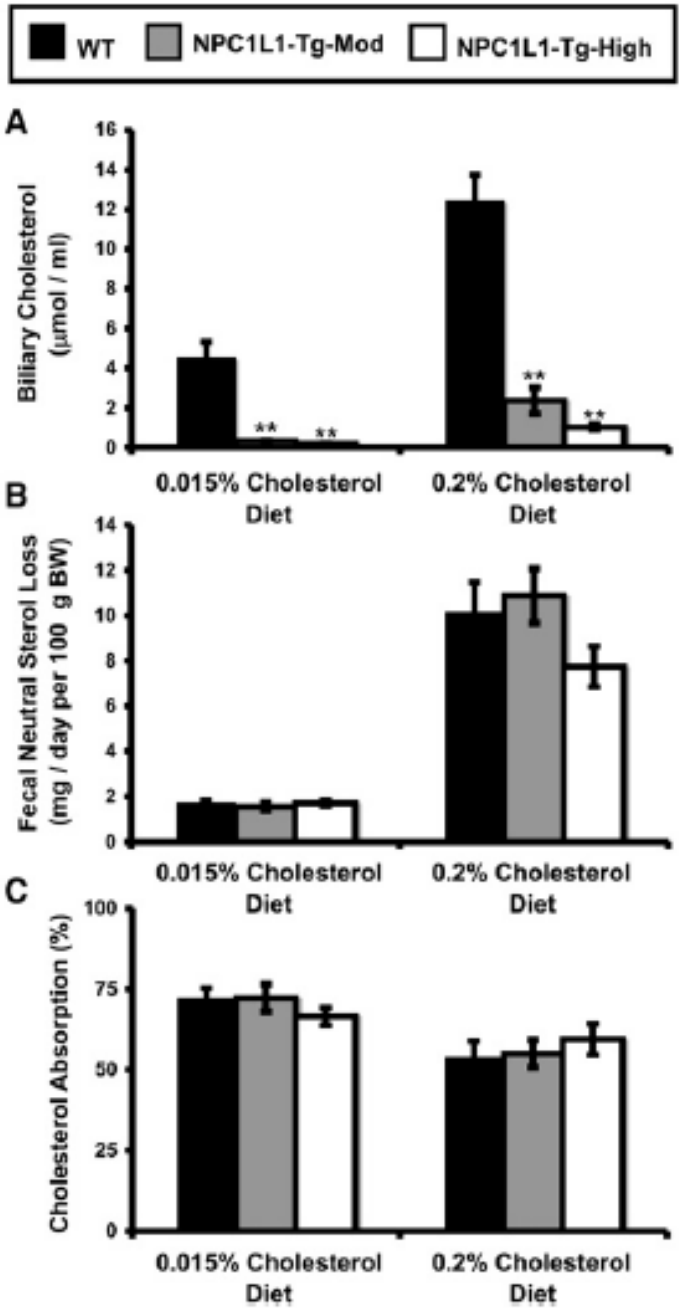


Yu, PNAS 2002

Non-biliary cholesterol excretion

- Increased FNS loss in dogs with complete biliary diversion and cholesterol free diet (Pertsemlidis, JCI 1973)

- Genetically modified mice with impaired biliary secretion:
  - C7a hydroxylase -/- (Schwarz, JLR 1998)
  - Abcb4-/- (Kruit, Gastroenterology 2005)
  - Hepatic NPC1L1 +++ (Temel, JCI 2007)
  - Hepatic ACAT2 -/- (Brown, JBC 2008)



NPC1L1 $^{\text{Liver-Tg}}$ mice

> 90% reduction of biliary cholesterol secretion

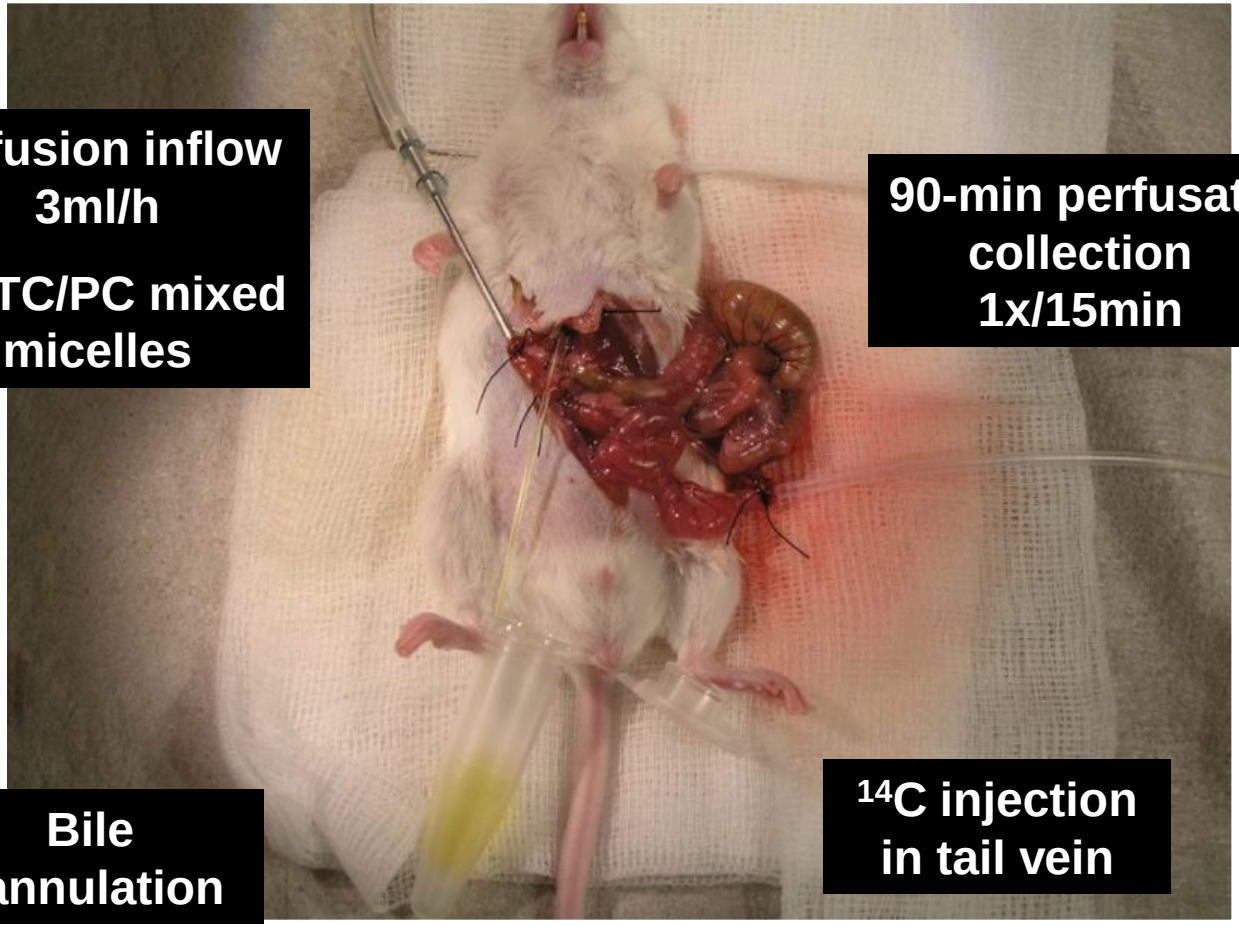
Normal FNS excretion

Normal intestinal cholesterol absorption

Similar plasma cholesterol levels as in wildtype

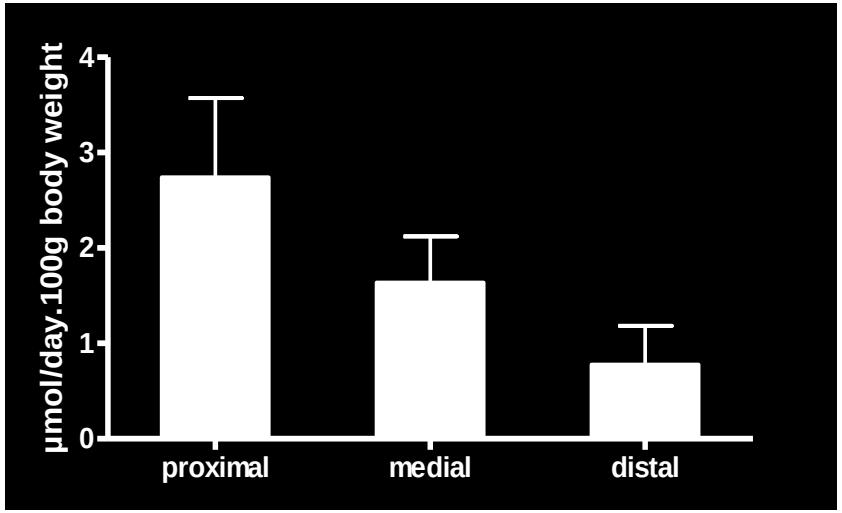
Temel, JCI 2007

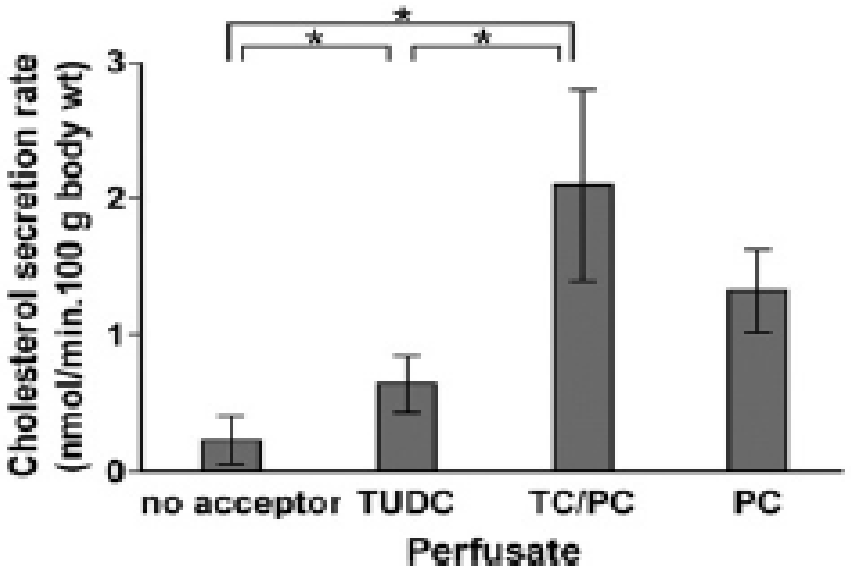
Intestinal perfusion experiments

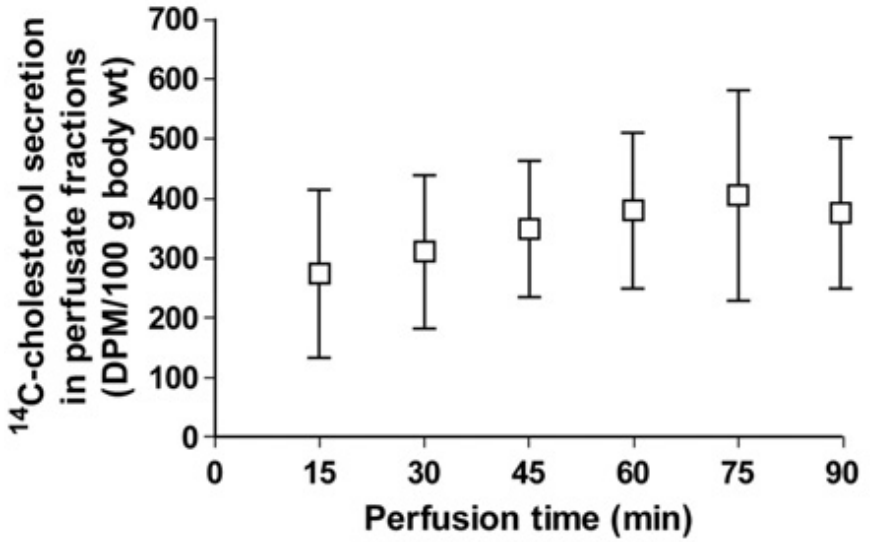


Van der Velde, Groen, Gastroenterology 2007

Trans-intestinal cholesterol excretion



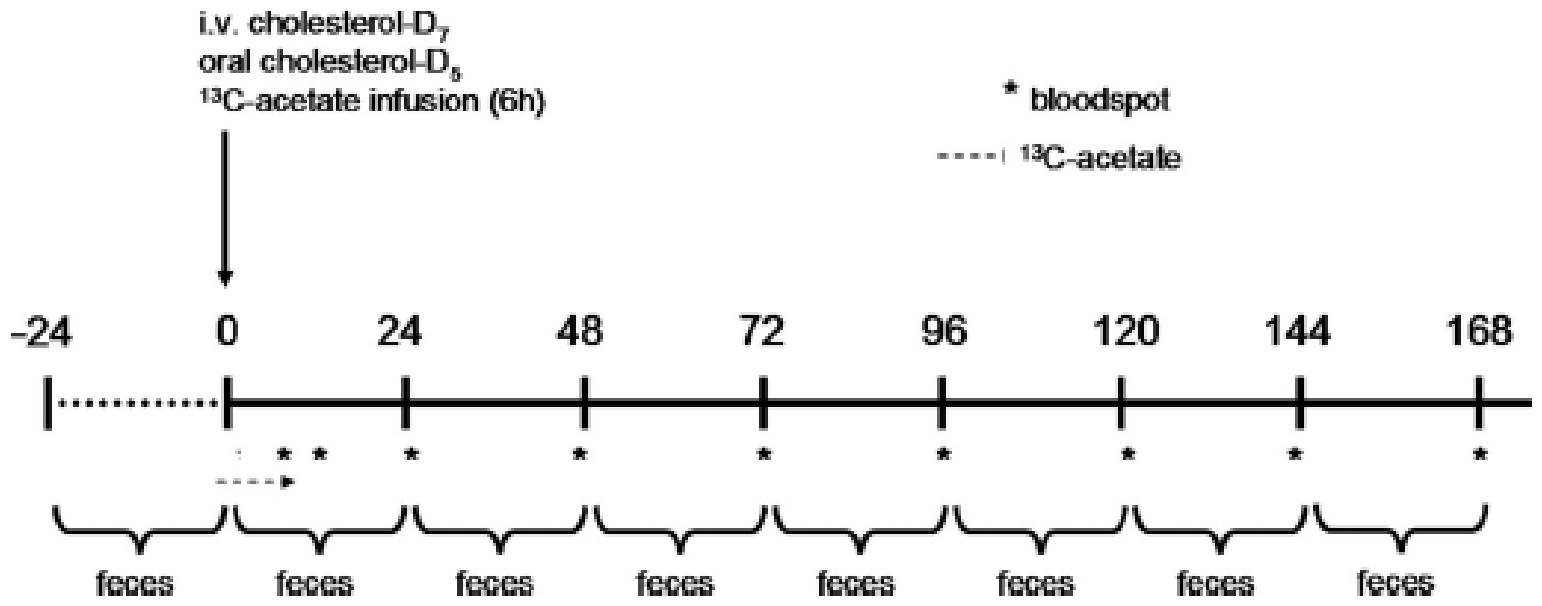




Van der Velde & Groen, Gastroenterology 2007

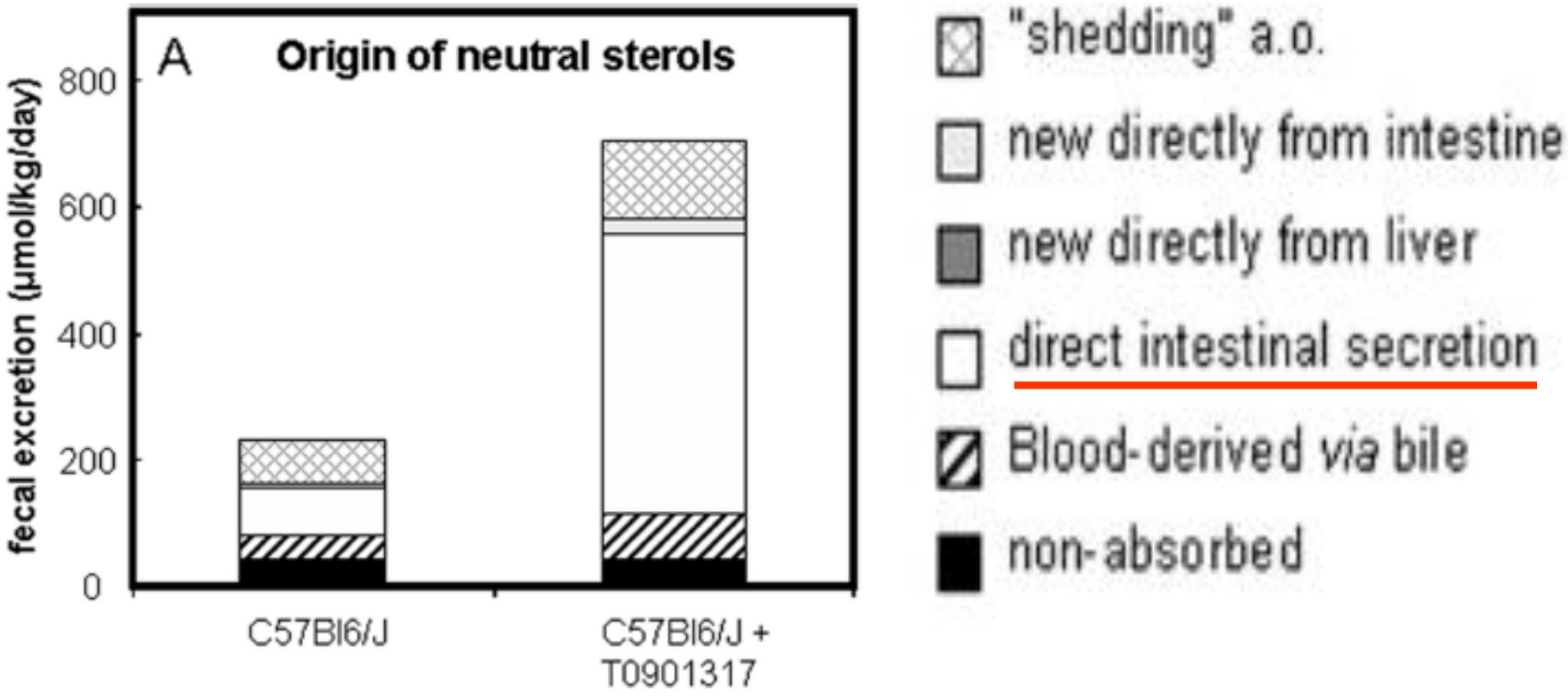
In vivo stable isotope study

Quantification of fractional and absolute contributions to FNS loss in vivo in mice +/– LXR agonist



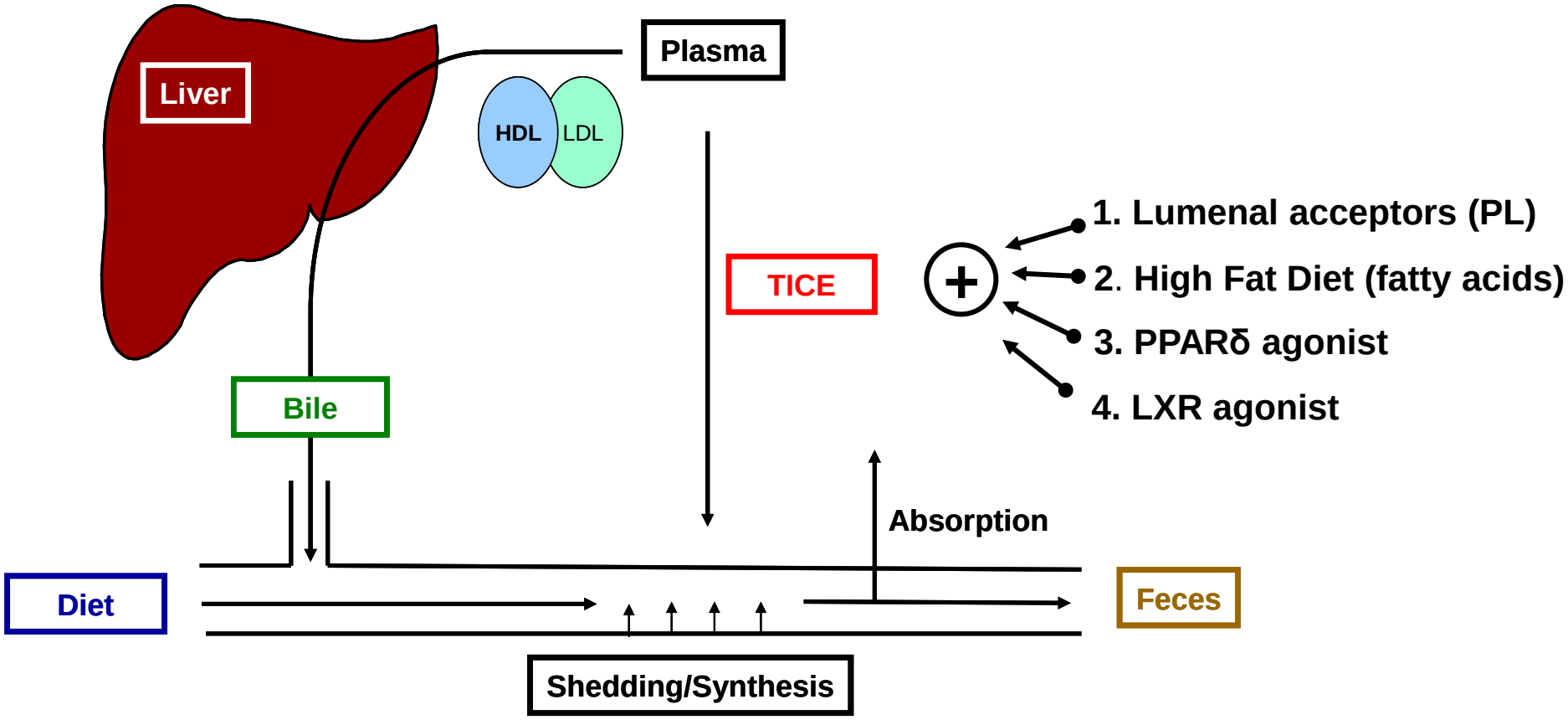
van der Veen, Groen, JBC 2009

LXR-induced increase in FNS loss is largely due to TICE stimulation



van der Veen, Groen, JBC 2009

Stimulation of TICE in mouse models

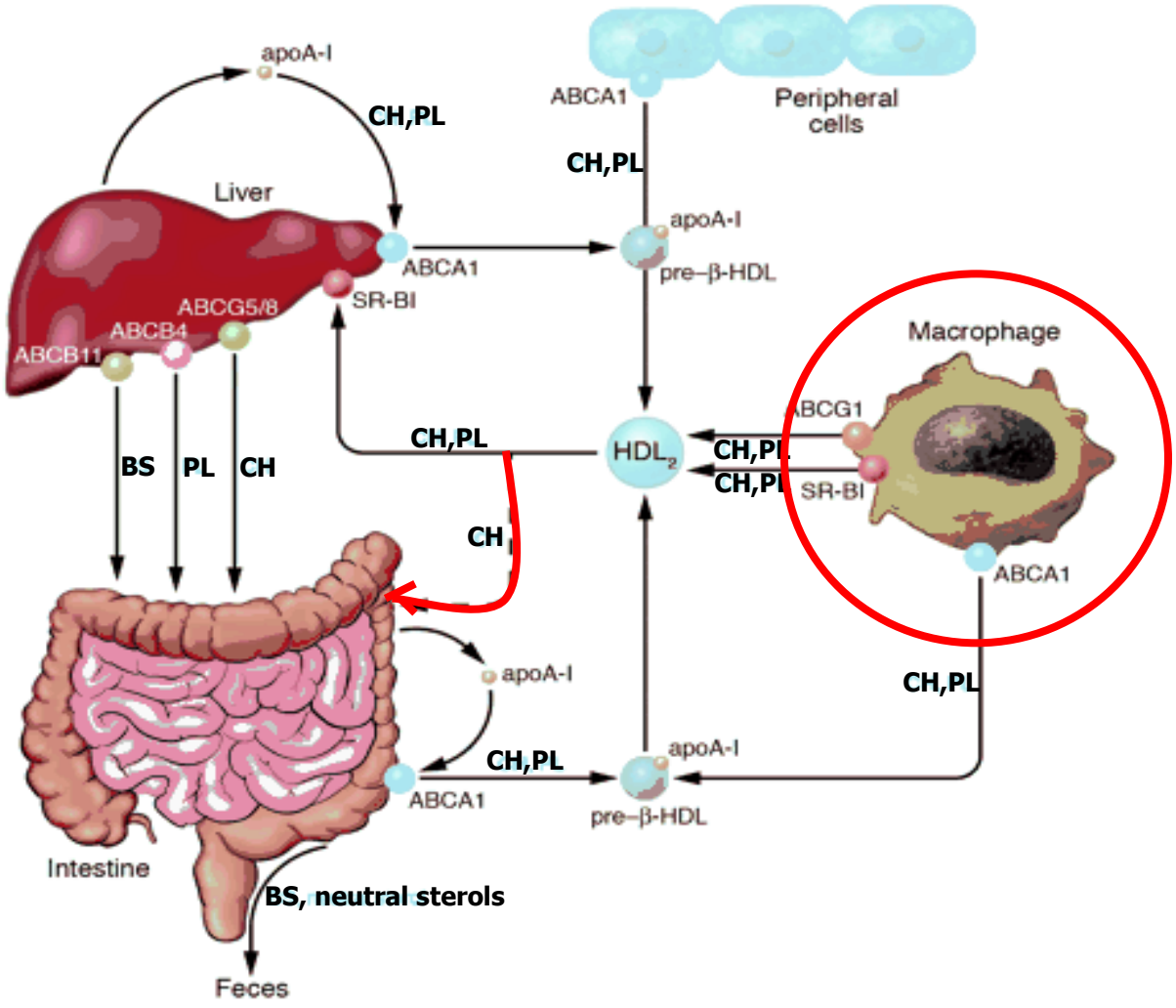


1. Van der Velde, Groen, Gastroenterology 2007

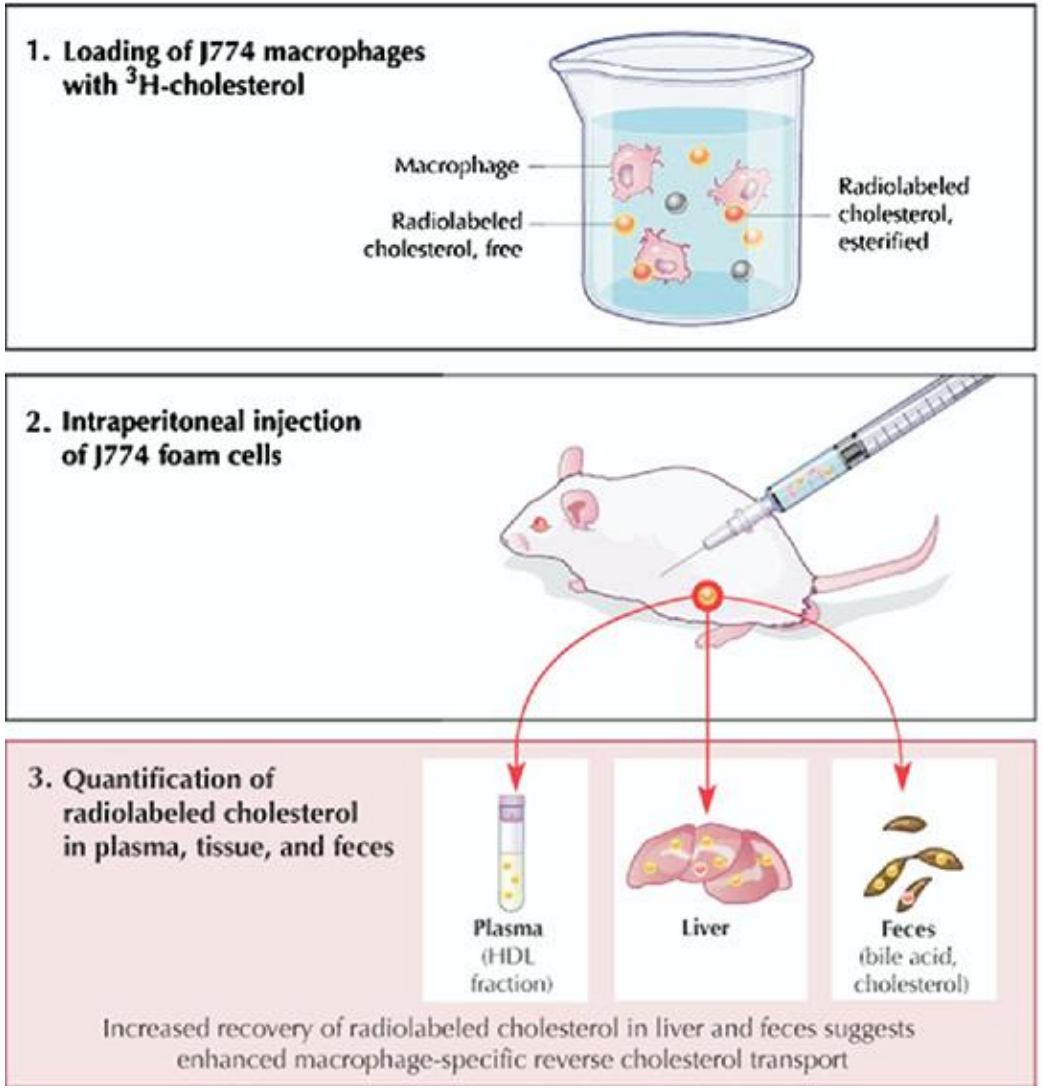
2. Van der Velde, Groen, Am J Physiol Gastrointest Liver Physiol 2008

3. Vrins, Groen, JLR 2009 *4. van der Veen, Groen, JBC 2009*

Is TICE an anti-atherogenic mechanism?

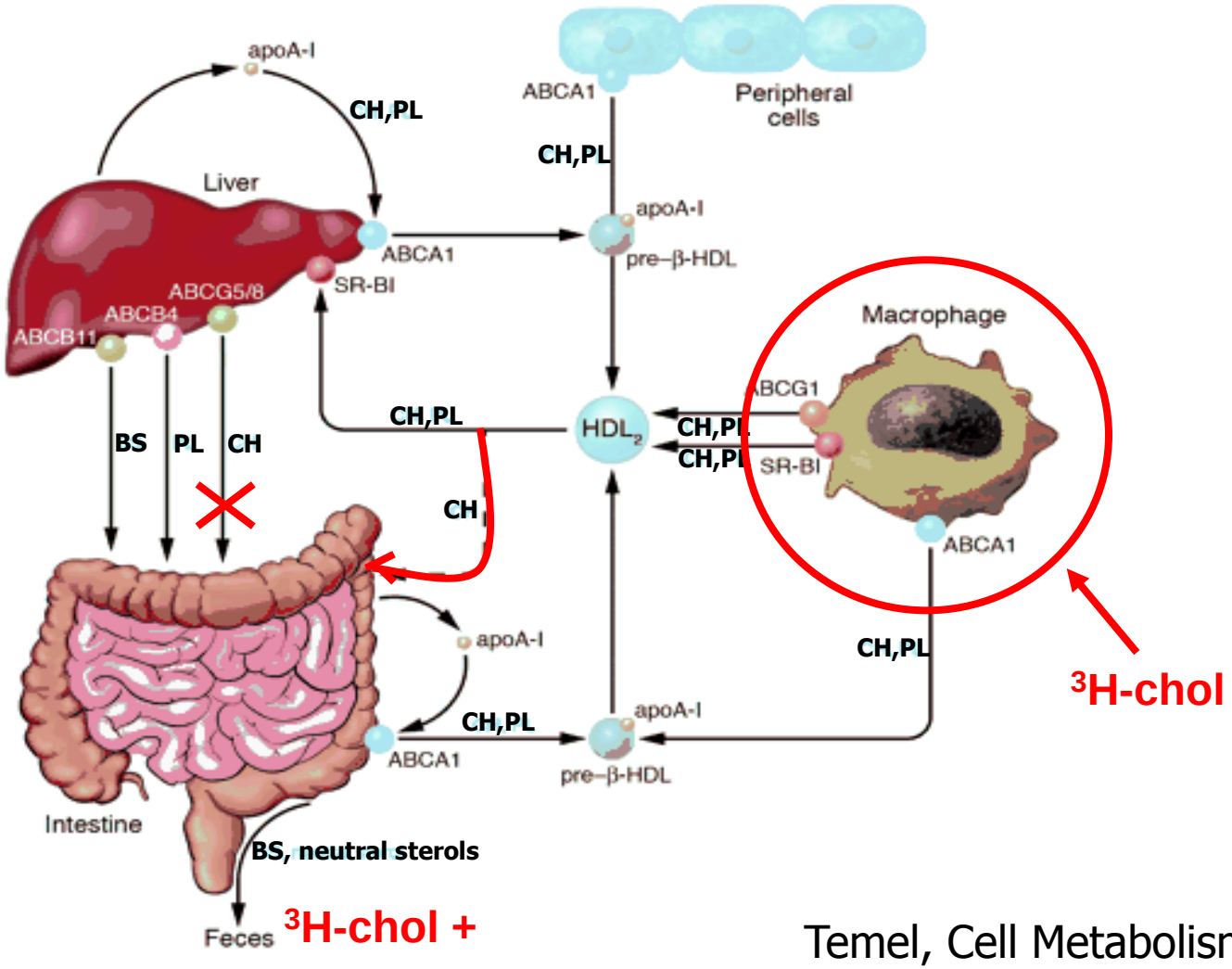


Macrophage-specific RCT assay



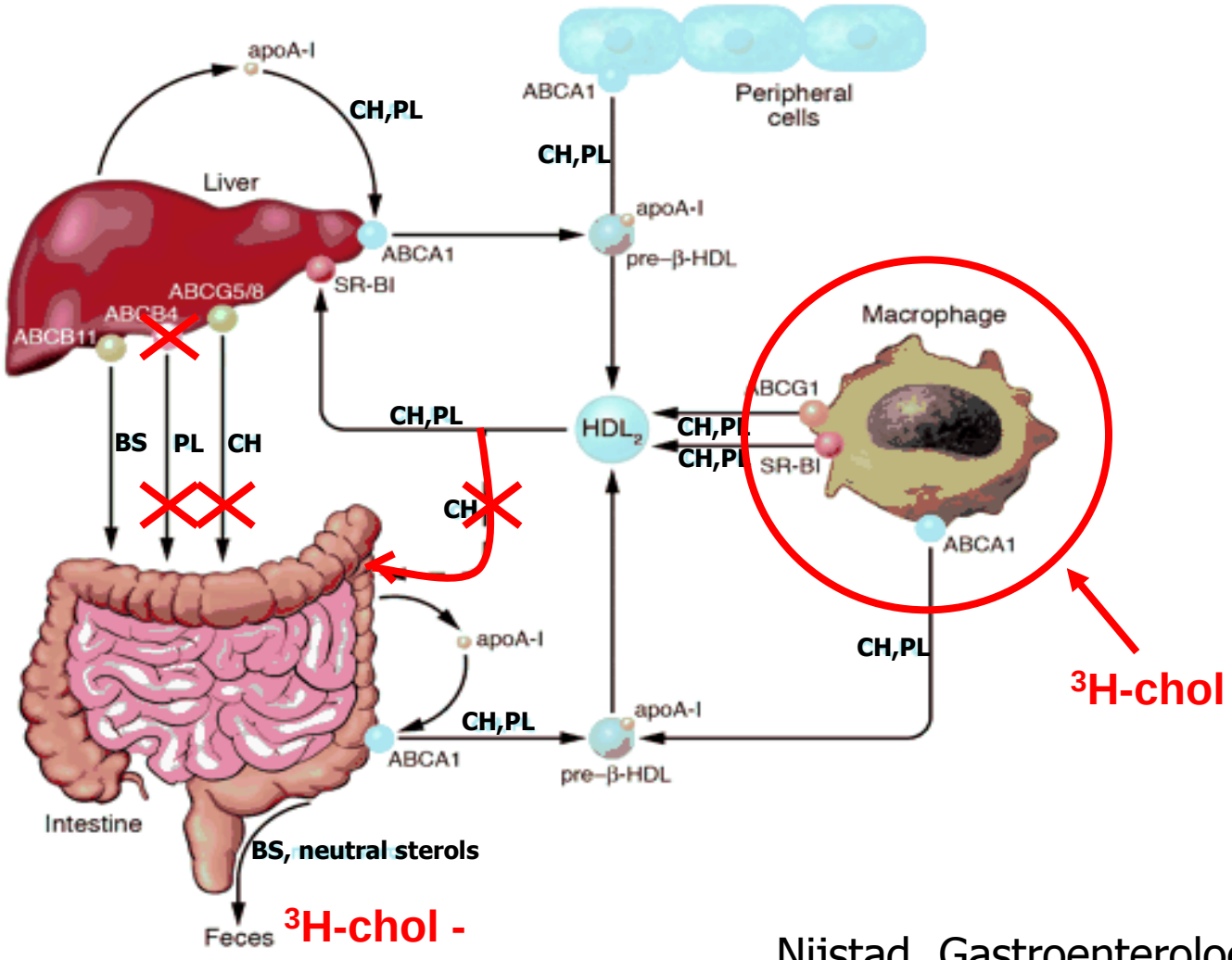
deGoma, JACC 2008

Non-biliary Φ-RCT in liver-NPC1L1++ mice



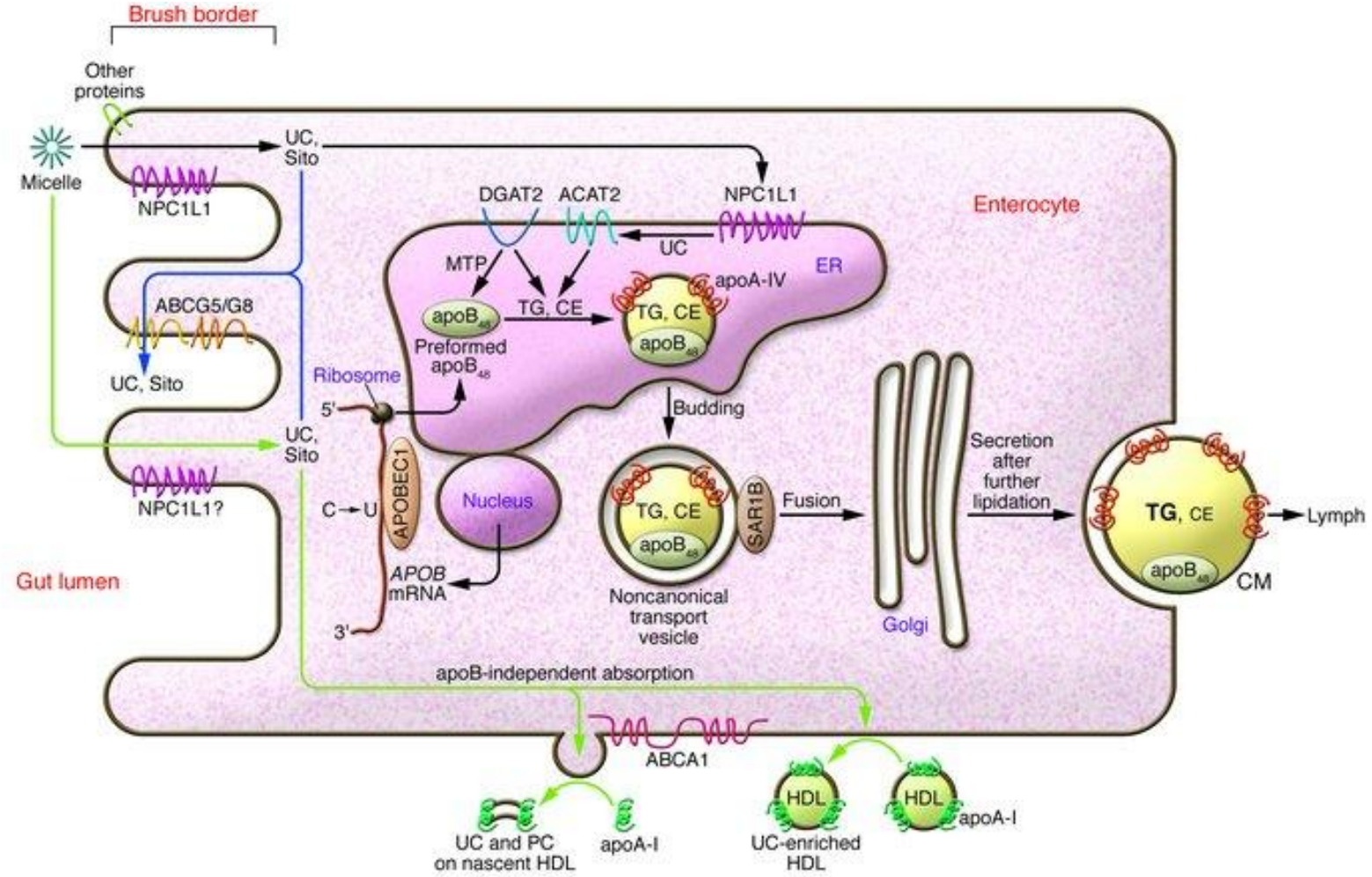
Temel, Cell Metabolism 2010

Obligate biliary Φ-RCT in Abcb4 -/- mice



Nijstad, Gastroenterology 2011

Underlying mechanisms?



Van der Velde, World J Gastroenterol 2010

Van der Velde, Brufau, Groen, Curr Opin Lipidol 2008

TICE in humans?

- Fecal sterols of non-dietary origin present in patients with complete biliary obstruction[1]
- Bile diversion in hoFH patients produced a 6-8-fold increase in GI sterol output[2]
- Human intestinal perfusion studies: TICE estimated as ~44% of total FNS loss[3]

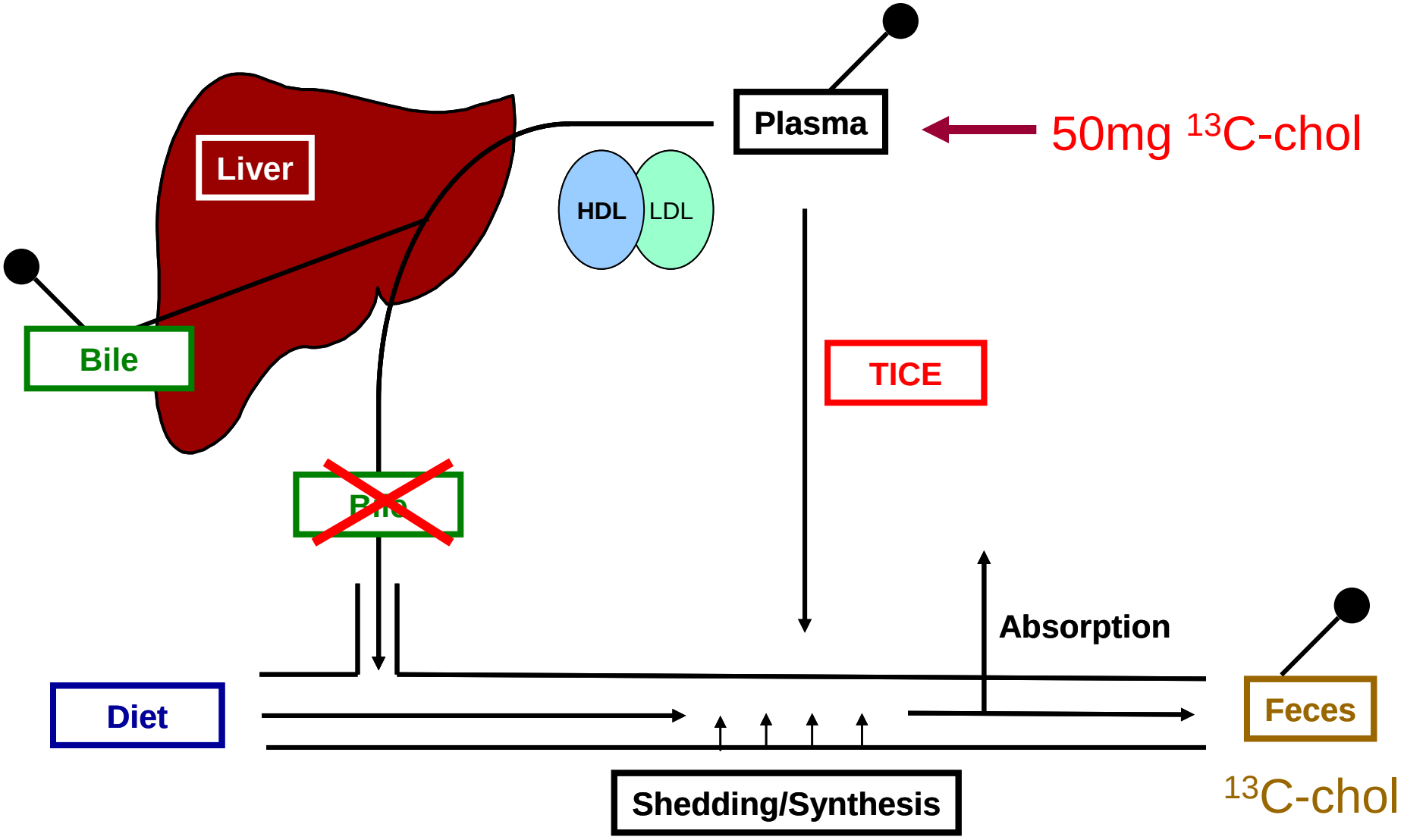
[1] Cheng, Proc Soc Exp Biol Med 1959

[2] Deckelbaum, NEJM 1977 [3] Simmonds, JCI 1967

Human TICE studies

- Proof-of-concept in patients with total biliary obstruction
- In vivo stable isotope study in subjects with intact enterohepatic cycle

Proof-of-concept: bile-diverted subjects

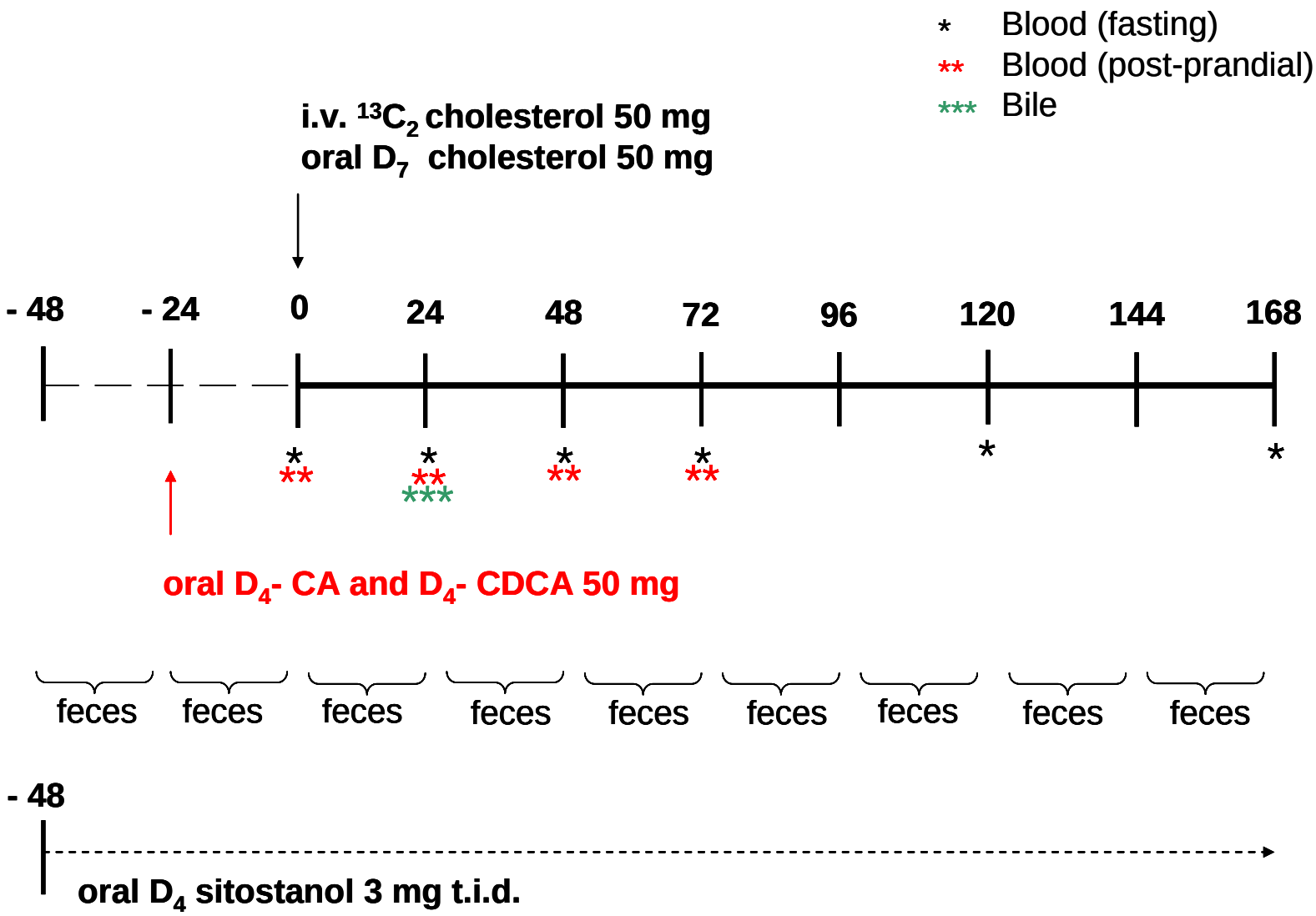


Human in vivo stable isotope study

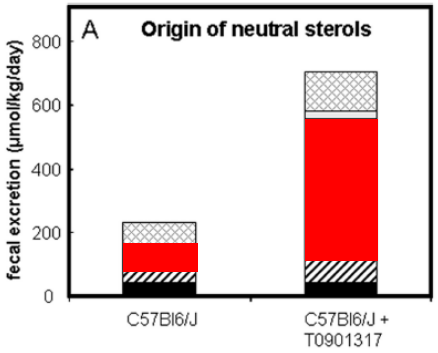
Male subjects, n	15
Age, years	61.7 ± 3.4
BMI, kg/m^2	25.7 ± 2.5
Total cholesterol, mmol/l	5.59 ± 0.65
LDL-cholesterol, mmol/l	3.74 ± 0.50
HDL-cholesterol, mmol/l	1.32 ± 0.27
Triglycerides, mmol/l	1.01 [0.66 – 2.61]

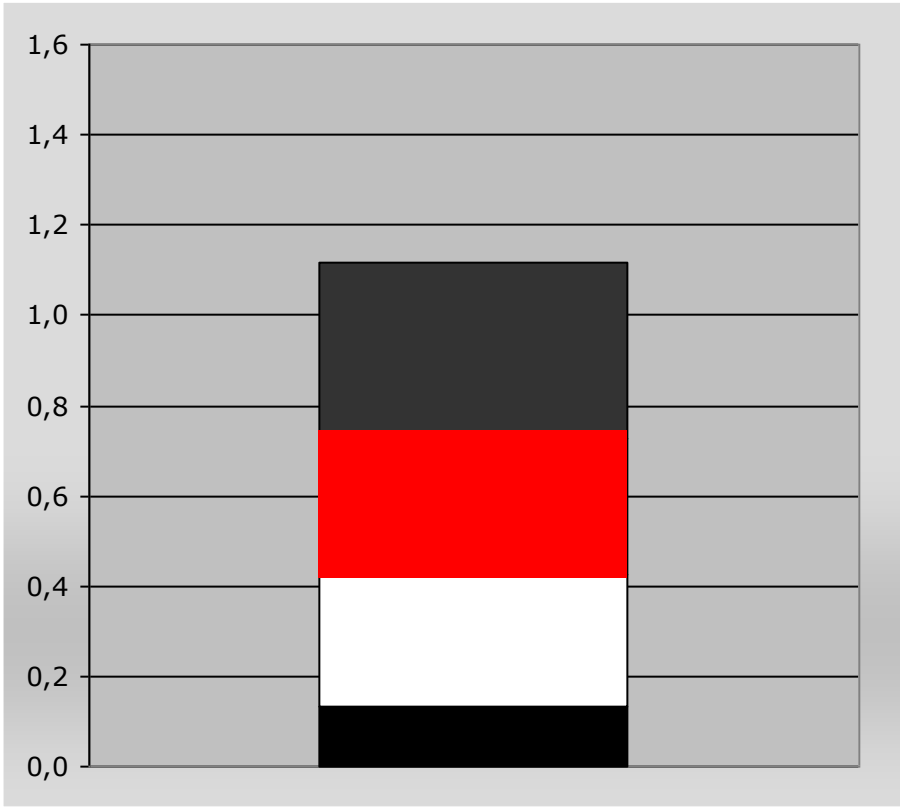
Jakulj, Stroes, Groen, unpublished

Human in vivo flux study



TICE ± 30% of FNS loss in humans





Jakulj, Stroes, Groen, unpublished

2. Obligate role of bile in RCT

- Non-biliary cholesterol excretion contributes to plasma cholesterol elimination in mice and men
- TICE might serve as an attractive target to improve RCT
- Focus on underlying molecular mechanisms and possibilities to stimulate TICE in humans

Acknowledgements

- A.K. Groen
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Nederlandse Hartstichting

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