

Shifting gears: liver SR-BI drives reverse cholesterol transport in macrophages

Astrid E. van der Velde, Albert K. Groen

J Clin Invest. 2005;115(10):2699-2701. <https://doi.org/10.1172/JCI26241>.

Commentary

Cholesterol efflux from macrophages, the first step in reverse cholesterol transport (RCT), is assumed to play a critical role in the pathogenesis of atherosclerosis. However, in vivo proof supporting this hypothesis is lacking, due to difficulties in determining the activity of this first step in RCT. In this issue of the *JCI*, Zhang et al. apply their recently developed method for measuring RCT in vivo to estimate RCT in mouse models with varying levels of HDL turnover. A surprisingly efficient clearance of cholesterol to feces is observed in mice overexpressing hepatic scavenger receptor class B type I (SR-BI), whereas in *SR-BI*-knockout mice, cholesterol clearance is diminished. The study demonstrates that hepatic SR-BI is a positive regulator of macrophage RCT in vivo.

Find the latest version:

<https://jci.me/26241/pdf>





1. Saitz, R. 2005. Clinical practice. Unhealthy alcohol use. *N. Engl. J. Med.* **352**:596–607.
2. Grant, B.F. 1994. Alcohol consumption, alcohol abuse and alcohol dependence. The United States as an example. *Addiction*. **89**:1357–1365.
3. Harwood, H.J., Fountain, D., and Livermore, G. 1998. Economic costs of alcohol abuse and alcoholism. *Recent Dev. Alcohol*. **14**:307–330.
4. Rose, R. 1998. A developmental behavior-genetic perspective on alcoholism risk. *Alcohol Health Res. World*. **22**:131–143.
5. Tupala, E., and Tiihonen, J. 2004. Dopamine and alcoholism: neurobiological basis of ethanol abuse [review]. *Prog. Neuropsychopharmacol. Biol. Psychiatry*. **28**:1221–1247.
6. Oswald, L., McCaul, M., Wong, D., and Wand, G. 2005. Association of ventral striatal dopamine release with cortisol and drug liking. *Neuropsychopharmacology*. **30**:821–832.
7. Yoder, K., et al. 2005. Dopamine D(2) receptor availability is associated with subjective responses to alcohol. *Alcohol. Clin. Exp. Res.* **29**:965–970.
8. Volkow, N., Fowler, J., and Wang, G. 2004. The addicted human brain viewed in the light of imaging studies: brain circuits and treatment strategies. *Neuropharmacology*. **47**(Suppl. 1):3–13.
9. Liu, X., and Weiss, F. 2002. Reversal of ethanol-seeking behavior by D1 and D2 antagonists in an animal model of relapse: differences in antagonist potency in previously ethanol-dependent versus nondependent rats. *J. Pharmacol. Exp. Ther.* **300**:882–889.
10. Merikangas, K.R., et al. 1998. Comorbidity of substance use disorders with mood and anxiety disorders: results of the International Consortium in Psychiatric Epidemiology. *Addict. Behav.* **23**:893–907.
11. Koob, G.F. 2003. Alcoholism: allostasis and beyond. *Alcohol. Clin. Exp. Res.* **27**:232–243.
12. Willinger, U., et al. 2002. Anxiety as a predictor of relapse in detoxified alcohol-dependent patients. *Alcohol Alcohol*. **37**:609–612.
13. Pandey, S.C., Zhang, H., Roy, A., and Xu, T. 2005. Deficits in amygdaloid cAMP-responsive element-binding protein signaling may play a role in genetic predisposition to anxiety and alcoholism. *J. Clin. Invest.* **115**:2762–2773. doi:10.1172/JCI24381.
14. Koob, G. 2004. A role for GABA mechanisms in the motivational effects of alcohol. *Biochem. Pharmacol.* **68**:1515–1525.
15. Li, T.-K., Lumeng, L., and Doolittle, D.P. 1993. Selective breeding for alcohol preference and associated responses. *Behav. Genet.* **23**:163–170.
16. McClung, C.A., and Nestler, E.J. 2003. Regulation of gene expression and cocaine by CREB and ΔFosB. *Nat. Neurosci.* **6**:1208–1215.
17. Hoffman, P., and Tabakoff, B. 1990. Ethanol and guanine nucleotide binding proteins: a selective interaction. *FASEB J.* **4**:2612–2622.
18. Yang, X., Horn, K., Baraban, J.M., and Wand, G.S. 1998. Chronic ethanol administration phosphorylation of cyclic AMP-response element binding protein in granule cells of rat cerebellum. *J. Neurochem.* **70**:224–232.
19. Constantinescu, A., Diamond, I., and Gordon, A.S. 1999. Ethanol-induced translocation of cAMP-dependent protein kinase to the nucleus. Mechanism and functional consequences. *J. Biol. Chem.* **274**:26985–26991.
20. Li, J., Li, Y.-H., and Yuan, X.-R. 2003. Changes of phosphorylation of cAMP response binding protein in rat nucleus accumbens after chronic ethanol intake: naloxone reversal. *Acta Pharmacol. Sin.* **24**:930–936.
21. Pandey, S.C., Roy, A., Zhang, H., and Xu, T. 2004. Partial deletion of the CREB gene promotes alcohol-drinking behaviors. *J. Neurosci.* **24**:5022–5030.
22. Valdez, G., and Koob, G. 2004. Allostasis and dysregulation of corticotropin-releasing factor and neuropeptide Y systems: implications for the development of alcoholism. *Pharmacol. Biochem. Behav.* **79**:67–689.
23. Thiele, T.E., Marsh, D.J., Ste. Marie, L., Bersntein, I.L., and Palmiter, R.D. 1998. Ethanol consumption and resistance are inversely related to neuropeptide Y levels. *Nature*. **396**:366–369.
24. Thiele, T., and Badia-Elder, N. 2003. A role for neuropeptide Y in alcohol intake control: evidence from human and animal research. *Physiol. Behav.* **79**:9–101.
25. Suzuki, R., Lumeng, L., McBride, W.J., Li, T.-K., and Hwang, B.H. 2004. Reduced neuropeptide Y mRNA expression in the central nucleus of amygdala of alcohol preference and anxiety. *Brain Res.* **1014**:251–254.
26. Kampov-Polevoy, A.B., Matthews, D.B., Gause, L., Morrow, A.L., and Overstreet, D.H. 2000. P rats develop physical dependence on alcohol via voluntary drinking: changes in seizure thresholds, anxiety, and patterns of alcohol drinking. *Alcohol. Clin. Exp. Res.* **24**:278–284.
27. Merlo-Pich, E., and Weiss, F. 1998. Neurocircuitry targets in ethanol reward and dependence. *Alcohol. Clin. Exp. Res.* **22**:3–9.
28. Valdez, G.R., Zorrilla, E.P., Roberts, A.J., and Koob, G.F. 2003. Antagonism of corticotropin-releasing factor attenuates the enhanced responsiveness to stress observed during protracted ethanol abstinence. *Alcohol*. **29**:55–60.
29. Hyttia, P., and Koob, G. 1995. GABAA receptor antagonism in the extended amygdala decreases ethanol self-administration in rats. *Eur. J. Pharmacol.* **283**:151–159.
30. Krystal, J.H., et al. Naltrexone in the treatment of alcohol dependence. *N. Engl. J. Med.* **345**:1734–1739.

Shifting gears: liver SR-BI drives reverse cholesterol transport in macrophages

Astrid E. van der Velde and Albert K. Groen

Academic Medical Center Liver Center, Academic Medical Center, Amsterdam, The Netherlands.

Cholesterol efflux from macrophages, the first step in reverse cholesterol transport (RCT), is assumed to play a critical role in the pathogenesis of atherosclerosis. However, in vivo proof supporting this hypothesis is lacking, due to difficulties in determining the activity of this first step in RCT. In this issue of the JCI, Zhang et al. apply their recently developed method for measuring RCT in vivo to estimate RCT in mouse models with varying levels of HDL turnover. A surprisingly efficient clearance of cholesterol to feces is observed in mice overexpressing hepatic scavenger receptor class B type I (SR-BI), whereas in SR-BI-knockout mice, cholesterol clearance is diminished (see the related article beginning on page 2870). The study demonstrates that hepatic SR-BI is a positive regulator of macrophage RCT in vivo.

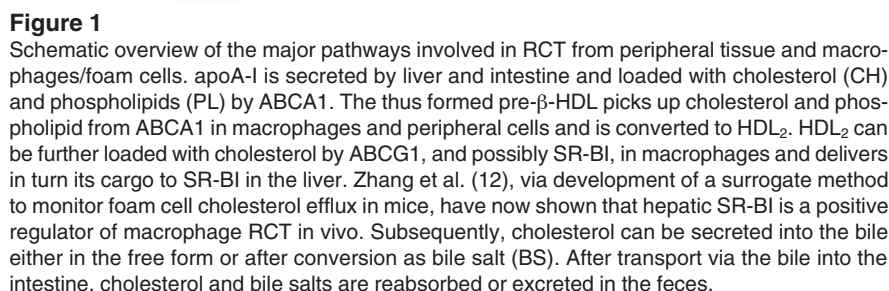
Nonstandard abbreviations used: RCT, reverse cholesterol transport; SR-BI, scavenger receptor class B type I.

Conflict of interest: The authors have declared that no conflict of interest exists.

Citation for this article: *J. Clin. Invest.* **115**:2699–2701 (2005). doi:10.1172/JCI26241.

The transport of excess cholesterol from the periphery into the liver and bile, followed by excretion in the feces, is defined as reverse cholesterol transport (RCT) (Figure 1). Since the original definition of RCT by Glomset and Norum in 1973 (1), this pathway has become increasingly

popular as a target for therapeutic strategies aimed at achieving the regression of atherosclerosis. Theoretically, a lipid-laden macrophage can release its contents by activation of efflux pathways. HDL is considered to be the primary cholesterol carrier in RCT, but conclusive evidence for this contention has been lacking until now. It is not an easy task to estimate net cholesterol flux from peripheral tissues to feces. Neutral sterols found in feces are derived from several sources. The major source is the liver, where the bulk of body cholesterol and bile salts are synthesized. After their secretion from the liver in bile, both components are secreted into the intestine, where up to 95% of bile salts and 30–60% of cholesterol is reabsorbed. The nonabsorbed cholesterol partly undergoes bacte-



Regulation of cholesterol efflux from macrophages

An imbalance in the pathways responsible for cellular cholesterol influx and efflux causes the conversion of a macrophage into a foam cell. Influx of cholesterol into macrophages may occur via a number of

affinity for free apoA-I and pre- β -HDL, whereas ABCG1 and SR-BI probably interact primarily with more mature HDL (4) (Figure 1). In all of these steps, the different forms of HDL play a pivotal role. Consequently, it has long been thought that plasma HDL levels accurately reflect the rate of RCT. Since many epidemiological studies have shown a strong inverse relationship between cardiovascular disease risk and HDL levels, this seemed a plausible paradigm. Particularly elegant studies by Dietschy and colleagues (5–7) have challenged this concept. Jolley, Dietschy, et al. (6) could not discern any effect on cholesterol homeostasis in *Apoa1*-null mice with very low HDL levels. Similar results were reported by our group in experiments with *Abca1*-null mice, in which HDL is almost absent (8). Alam et al. (9) upregulated the expression of proteins that mediate individual steps believed to be involved in HDL trafficking pathways in normolipidemic mice and did not find any effect on RCT. In humans, 2 studies have demonstrated a significant effect of apoA-I or reconstituted HDL infusions on neutral sterol output (10, 11). The major pitfall in all of these studies is the lack of differentiation between the different sources contributing to fecal neutral sterol output. As we have discussed above, cholesterol efflux from foam cells – the most relevant step in RCT with respect to atherosclerosis – may be only a minor contributor to total RCT. All efforts to visualize HDL-mediated regulation of macrophage cholesterol efflux may have failed because of the inability to measure this minor flux

In this issue of the *JCI*, Zhang et al. attempt to circumvent this problem by using a surrogate method to monitor cholesterol efflux from macrophages (12). For this purpose, mouse J774 cells were labeled in vitro with [³H]cholesterol and loaded with lipid by incubation with acetylated LDL. Subsequently, the cells were injected into the peritoneum of mice, and RCT was measured by studying the appearance of the label in plasma, liver, and feces. In an earlier study, the authors showed that most of the tritiated cholesterol was esterified in J774 cells, and after 24 hours, a significant amount appeared in the feces, in the form of both bile salts and neutral sterols (13). Clearly, J774 cells are not equivalent to macrophages/foam cells present in the



vessel wall or atherosclerotic lesions, and the results of the study have to be considered in light of this difference. Zhang et al. did not measure specific activities of the different cholesterol pools. This impedes an estimation of cholesterol mass transfer in the different experiments. However, the different mouse models employed in the study do allow some speculation with respect to the preferred metabolic routes involved in the handling of effluxed cholesterol. How cholesterol is transported from the peritoneal cavity to the blood is not clear, but once in plasma, the tritiated cholesterol equilibrates with plasma cholesterol. In wild-type mice, about 2% of the injected label appeared in the feces within the first 24 hours. This is a substantial amount in view of the fact that only 4% of the label was present in plasma. Overexpression of SR-BI in the liver was observed to significantly increase the clearance rate, particularly in the presence of apoA-I overexpression. These findings are very suggestive of an important role for HDL in macrophage cholesterol efflux. Changes in specific activity of plasma cholesterol in the different mouse models may, however, have confounded these apparently clear results. For instance, overexpression of SR-BI induced a 6-fold decrease in total serum cholesterol, whereas the percentage of [^3H]cholesterol decreased less than 3-fold and thus invoked an increase in plasma-specific activity (12). This may explain at least part of the observed increase in fecal output of tritiated cholesterol without an increase in net fecal output. Alam et al. (9) used similar mouse models to inves-

tigate the influence of variation of processes involved in RCT on fecal sterol output. In mouse models of both SR-BI and apoA-I overexpression, no effect on fecal sterol output was observed, despite similar variations observed in plasma cholesterol levels. Alam et al. (9) measured total fecal excretion and could not discriminate between cholesterol derived from the periphery and that derived from macrophages. Unfortunately, Zhang et al. did not determine total neutral sterol secretion in their experiments, impeding a direct comparison with the study by Alam et al. (9).

Conclusions

A method to assess the rate of cholesterol efflux from foam cells is desperately needed to be able to define the importance of this step in the progression and possibly the regression of atherosclerosis. An initial step toward this goal has been made by Zhang et al. (12). Clearly this method requires further evaluation, but it may be a first step in the development of a surrogate assay to determine RCT in humans.

Address correspondence to: Albert K. Groen, Academic Medical Center Liver Center, Meibergdreef 69-71, 1105 BK Amsterdam, The Netherlands. Phone: 31-20-5664174; Fax: 31-20-5669190; E-mail: A.K.groen@amc.uva.nl.

1. Glomset, J.A., and Norum, K.R. 1973. The metabolic role of lecithin: cholesterol acyltransferase: perspectives form pathology [review]. *Adv. Lipid Res.* **11**:1-65.
2. Kruit, J.K., et al. 2005. Increased fecal neutral sterol loss upon liver X receptor activation is independent of biliary sterol secretion in mice. *Gastroenterology*.

128:147-156.

3. Kunjathoor, V.V., et al. 2002. Scavenger receptors class A-I/II and CD36 are the principal receptors responsible for the uptake of modified low density lipoprotein leading to lipid loading in macrophages. *J. Biol. Chem.* **277**:49982-49988.
4. Linsel-Nitschke, P., and Tall, A.R. 2005. HDL as a target in the treatment of atherosclerotic cardiovascular disease. *Nat. Rev. Drug Discov.* **4**:193-205.
5. Dietschy, J.M. 1966. Recent developments in solute and water transport across the gallbladder epithelium. *Gastroenterology*. **50**:692-707.
6. Jolley, C.D., Woollett, L.A., Turley, S.D., and Dietschy, J.M. 1998. Centripetal cholesterol flux to the liver is dictated by events in the peripheral organs and not by the plasma high density lipoprotein or apolipoprotein A-I concentration. *J. Lipid Res.* **39**:2143-2149.
7. Osono, Y., Woollett, L.A., Marotti, K.R., Melchior, G.W., and Dietschy, J.M. 1996. Centripetal cholesterol flux from extrahepatic organs to the liver is independent of the concentration of high density lipoprotein-cholesterol in plasma. *Proc. Natl. Acad. Sci. U. S. A.* **93**:4114-4119.
8. Groen, A.K., et al. 2001. Hepatobiliary cholesterol transport is not impaired in Abca1-null mice lacking HDL. *J. Clin. Invest.* **108**:843-850. doi:10.1172/JCI200112473.
9. Alam, K., Meidell, R.S., and Spady, D.K. 2001. Effect of up-regulating individual steps in the reverse cholesterol transport pathway on reverse cholesterol transport in normolipidemic mice. *J. Biol. Chem.* **276**:15641-15649.
10. Eriksson, M., Carlson, L.A., Miettinen, T.A., and Angelin, B. 1999. Stimulation of fecal steroid excretion after infusion of recombinant proapolipoprotein A-I. Potential reverse cholesterol transport in humans. *Circulation*. **100**:594-598.
11. Nanjee, M.N., et al. 2001. Intravenous apoA-I/lecithin discs increase pre-beta-HDL concentration in tissue fluid and stimulate reverse cholesterol transport in humans. *J. Lipid Res.* **42**:1586-1593.
12. Zhang, Y., et al. 2005. Hepatic expression of scavenger receptor class B type I (SR-BI) is a positive regulator of macrophage reverse cholesterol transport in vivo. *J. Clin. Invest.* **115**:2870-2874. doi:10.1172/JCI25327.
13. Zhang, Y., et al. 2003. Overexpression of apolipoprotein A-I promotes reverse transport of cholesterol from macrophages to feces in vivo. *Circulation*. **108**:661-663.