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A lipid to treat non-alcoholic fatty liver disease – The dawn of 'lipo-rehabilitation'?

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COMMENTARY ON:

A nuclear-receptor-dependent phosphatidylcholine pathway with antidiabetic effects. Lee JM, Lee YK, Mamrosh JL, Busby SA, Griffin PR, Pathak MC, Ortlund EA, Moore DD. Nature 2011 May 25;474(7352):506–510. doi:10.1038/nature10111. Copyright (2011). Reprinted by permission from Macmillan Publishers Ltd.

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Abstract*: Nuclear hormone receptors regulate diverse metabolic pathways and the orphan nuclear receptor LRH-1 (also known as NR5A2) regulates bile acid biosynthesis. Structural studies have identified phospholipids as potential LRH-1 ligands, but their functional relevance is unclear. Here we show that an unusual phosphatidylcholine species with two saturated 12 carbon fatty acid acyl side chains (dilauroyl phosphatidylcholine (DLPC)) is an LRH-1 agonist ligand in vitro. DLPC treatment induces bile acid biosynthetic enzymes in mouse liver, increases bile acid levels, and lowers hepatic triglycerides and serum glucose. DLPC treatment also decreases hepatic steatosis and improves glucose homeostasis in two mouse models of insulin resistance. Both the antidiabetic and lipotropic effects are lost in liver-specific Lrh-1 knockouts. These findings identify an LRH-1 dependent phosphatidylcholine signalling pathway that regulates bile acid metabolism and glucose homeostasis.*

With the advent of increasingly sedentary lifestyles and changing dietary patterns, the prevalence of obesity and insulin resistance have increased and with this non-alcoholic fatty liver disease (NAFLD) has rapidly become the most common cause of abnormal liver biochemistry in many developed countries [1,2]. It is apparent that, whilst the majority of patients with NAFLD have simple steatosis, a significant number exhibit steatohepatitis and may subsequently progress to fibrosis, cirrhosis and hepatocellular carcinoma. Pooled data indicate that up to 5.4% of patients with steatohepatitis develop complications of end-stage liver disease during long-term follow-up and many experience cardiovascular morbidity [1–3]. Despite substantial clinical and basic research in this field, a targeted therapy for NAFLD has so far proved elusive [2].

It is recognised that increased hepatic free fatty acid (FFA) flux on a background of obesity and insulin resistance drives progressive NAFLD through direct hepatocyte lipotoxicity; oxidative stress secondary to free radical production during fatty acid oxidation; endoplasmic reticulum stress; endotoxin/TLR4 induced Kupffer cell activation and cytokine release [4]. Consequent cellular damage triggers immune mediated hepatocellular injury, activates caspase dependent and independent necrotic and apoptotic cell death pathways and ultimately leads to stellate cell activation and fibrogenesis [5,6]. However, in recent years, as our understanding of the pathogenesis of NAFLD has grown, the view that steatosis causes progressive NAFLD has been revised. This began with the recognition that, rather than representing an initiating 'first hit', steatosis was an early adaptive response to hepatocyte stress through which potentially lipotoxic FFAs are partitioned into relatively inert intracellular triglyceride stores [4,7]. Now, work suggesting that a phospholipid could be an orally active, direct therapy for NAFLD and perhaps the broader metabolic syndrome may offer the prospect of 'lipo-rehabilitation'.

In their recent letter to *Nature*, Lee and colleagues explore a potentially important but under-researched aspect of lipid function, the role of phospholipid species as nuclear receptor agonists [8]. Amongst its pleiotropic functions, the orphan nuclear receptor LRH-1 (NR5A2) influences embryonic development; cellular replication and differentiation; and metabolic processes including reverse cholesterol transport and hepatic bile acid metabolism [9–11]. It is accepted that modest increases in bile acids may reduce steatosis [12] and so, founded on reports that loss of LRH-1 decreases bile acid levels [10,11], Lee *et al.* pursued the hypothesis that an LRH-1 agonist would increase bile acid synthesis and thus ameliorate NAFLD. Structural studies had already identified phospholipids as potential LRH-1 ligands, although the functional relevance of this interaction was unknown [13,14], and so the authors screened a number of different phospholipids for effects on human LRH-1 transactivation *in vitro*. This led to the identification of two phosphatidylcholine species that, at relatively high concentrations, could activate LRH-1. Focusing on one, dilauroyl phosphatidylcholine (DLPC), which is structurally

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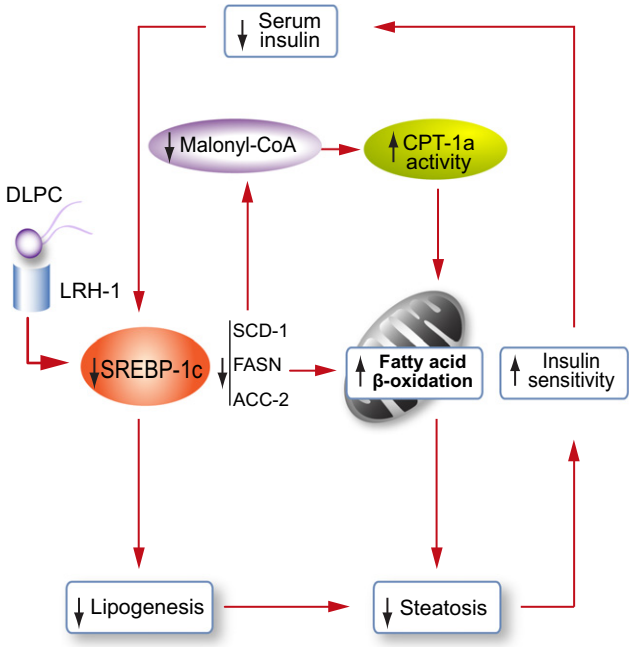

Fig. 1. The 'Virtuous Cycle' of insulin sensitizing effects and reduced NAFLD induced by DLPC.

characterized by two saturated 12-carbon fatty acid acyl side chains (C12:0/C12:0), Lee *et al.* first show that oral administration strongly induced Cyp7a1 and Cyp8b1 expression coupled with reduced serum glucose and increased serum bile acid levels in normal mice.

Following these encouraging data, the authors next examined the effect of orally administered DLPC on genetically obese, insulin-resistant *db/db* mice and in mice rendered obese and diabetic following prolonged consumption of high fat diet. The results of these studies were striking. DLPC induced significant reductions in hepatic lipid content, severity of histological NAFLD and insulin resistance/glucose intolerance. Importantly, in contrast to studies that have controlled steatosis by inhibiting triglyceride biosynthesis through targeting expression of diacylglycerol acyltransferase 2 (DGAT2) [7], no significant rise in biochemical markers of cellular injury (ALT and AST) was observed [8].

It appears that DLPC acts in an LRH-1 dependent manner to significantly suppress expression of the transcription factor SREBP-1c and its downstream target genes *ACC-2*, *SCD-1* and *FASN*. Interestingly, little or no direct effect of DLPC on expression of genes involved in fatty acid oxidation or glucose homeostasis was observed and DLPC did not appear to affect the nuclear receptors PPARα or PPARγ [8]. The authors postulate that a reduction in hepatic *de novo* lipogenesis due to suppression of SREBP-1c, likely combined with an increase in mitochondrial fatty acid β-oxidation (reduced synthesis of malonyl-CoA by ACC-2 will reduce inhibition of the rate limiting enzyme controlling mitochondrial fatty acid uptake, CPT-1a) and improved insulin sensitivity, produce a virtuous cycle ameliorating NAFLD and the metabolic syndrome (Fig. 1) [8]. Previous studies have also shown that LRH-1 negatively regulates the hepatic acute phase response and so agonists may offer additional anti-inflammatory effects [9]. Much work remains to be done to elucidate the mechanisms of DLPC activity and to confirm that these effects do not clear steatosis at the expense of increased oxidative stress. It remains unclear how DLPC enters the cell and, given the high concentrations of DLPC required to produce an effect, it may be that DLPC mediates its effects indirectly though a signaling cascade or by serving as a metabolic precursor for the true LRH-1 ligand [15].

These new data are certainly encouraging and we await with interest the results of the preliminary clinical trials that the authors have initiated however, a word of caution. Like other nuclear receptors, LRH-1 exhibits a wide spectrum of activity. This includes influencing cellular replication and immunomodulation [9]. LRH-1 has been shown to bind and activate hepatitis B virus enhancer II and so increase viral replication within hepatocytes but perhaps of greatest concern, if LRH-1 agonists were to be used as long-term therapy in NAFLD, are its potentially protumorigenic effects. LRH-1 activity regulates intestinal cell-cycle control and estradiol synthesis and has been shown to promote intestinal and breast tumor formation in murine models [9]. In humans, the LRH-1 locus has also been genetically associated with exocrine pancreatic neoplasia [9].

In conclusion, the work by Lee *et al.* provides new insights into the role of nuclear receptors in sensing variations in intracellular metabolite concentrations and the subsequent adaptive responses they can illicit through modulation of downstream gene expression. The effects of DLPC signaling via LRH-1 in murine models appear to have important therapeutic implications for NAFLD, however, there remains uncertainty about the exact mechanisms through which the effect is exerted and, for the moment at least, there is reason to be cautious regarding the long-term implications of heightened LRH-1 activity.

Conflict of interest

The authors declared that they do not have anything to disclose regarding funding or conflict of interest with respect to this manuscript.

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