PEMT as a key enzyme in metabolic reprogramming in chronic hepatitis C infection

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Introduction: Hepatitis C virus (HCV) replication is closely linked to lipid metabolism. Therefore, lipidomic analysis of HCV infected hepatic cells can offer insights into the pathogenesis of HCV infection and identify molecular targets that can serve as potential targets for new treatments.

Aim: The aim of this project was to study the effects of HCV infection on intracellular lipid homeostasis and turnover in hepatic cells. We have previously reported HCV-induced changes in neutral lipids; so now present data on phospholipids.

Methods: Huh7 cells were infected with HCV (JFH1 strain) and cultured until they were 90% infected. Cellular lipids were separated by high performance thin layer chromatography (HP-TLC) to measure changes in the major phospholipid species. The rate limiting enzymes of phosphatidylcholine metabolism were knocked down and the effects on HCV replication were measured.

Results: HP-TLC showed increased amounts of phosphatidylcholine in lipid extracts from whole cells and from ER fractions of JFH1 infected Huh-7 cells. PC was the only phospholipid species detected in purified lipid droplets, and was significantly increased in infected cells. PYTC1 (CTP: phosphocholine cytidylyltransferase) and PEMT (phosphatidylethanolamine N-methyltransferase) are the rate limiting enzymes of PC biosynthesis in hepatocytes. Silencing PYTC1 had no effect on HCV replication or infectivity. However, when PEMT was silenced, both viral replication and infectivity were decreased by more than 50%, and less lipids accumulation was observed.

Conclusion: Our previous data reveal global changes in lipid abundance, particularly in the ER, which are predicted to impact the HCV life cycle and pathogenesis. We now report increased PC content in the ER and in lipid droplets. Our data suggest that in HCV infected cells the minor PC synthesis pathway is most important, as inhibiting PEMT inhibits replication and production of infectious virus.

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