Hepatocyte-specific depletion of augmenter of liver regeneration (ALR) protein alters miRNA signature linked to lipid homeostasis leading to excessive steatosis

Sudhir Kumar1,2, Richa Ram1,2, Rebekah Karns1, Bal Krishan Sharma1, Chandrashekar R. Gandhi1,2,3, 1Department of Pediatrics, CCHMC, Cincinnati, OH, 2Cincinnati VA Medical Center, Cincinnati, OH, 3University of Cincinnati, Cincinnati, OH

Augmentor of liver regeneration (ALR) is a fundamental life protein expressed in all mammalian organs. ALR’s presence in mitochondria is essential for their function and survival. Depletion of ALR from liver (hepatocyte)-specific ALRflp/flp/Alb-Cre (ALR-L-KO) mice causes mitochondrial injury, robust lipid accumulation and apoptosis between 1 and 2 weeks post-birth. Steatosis is regressed upon reapparance of ALR by 4 weeks but continued death of Cre-expressing hepatocytes induces regeneration, inflammation and fibrosis. To delineate mechanisms of this pathological progression, we investigated whether ALR depletion alters expression of micro-RNAs (miRNAs), which play important roles in numerous biological processes including lipid homeostasis. We performed miRNA-seq analysis and profiled the hepatic miRNA expression in 1-, 2-, and 4-week old mice. Of the 765 micro-RNAs identified in WT and ALR-L-KO livers, 203 showed differential expression: 125, 106 and 141 miRNAs were up-regulated and 70, 97 and 61 miRNAs down-regulated at 1, 2 and 4 weeks respectively in ALR-L-KO compared to the WT mice. miR-708-3p, miR-540-3p and miR-541-5p, which have 3'UTR-binding sites for peroxisome proliferator-activated receptor (PPAR)α, carnitine palmitoyl transferase a (CPT1a) and mitochondrial transcription factor A (TFAM) (all down-regulated at 2 weeks in ALR-L-KO mice), were all found to be up-regulated at 2 weeks in ALR-L-KO compared to the WT mice. Increase in these miRNAs upon ALR depletion was also concurrent with increased expression of elongation of very long fatty acids 6 (ELOVL6) and steroyl CoA desaturase (SCD1) that are involved in de novo lipogenesis, and decreased expression of acyl CoA oxidase 1 (ACOX1) and CPT1 that promote β-oxidation of fatty acids. The effect of ALR depletion on these miRNAs was recapitulated in vitro in cultured hepatocytes. Likewise, hepatic expression of miRNAs implicated in inflammation and fibrosis (miR-199a-1-5p, miR-146b-5p, miR-194-1-5p, miR-1843a, miR-410-3p and miR-434-3p) was also altered in a way to promote these pathologies. These findings suggest that ALR’s regulation of miRNAs is critical to hepatic lipid homeostasis with potential implications in nonalcoholic fatty liver disease/steatohepatitis.

Support or Funding Information
This work was supported by DoD grant W81XWH-14-PRMRP-IIRA and NIH R21 AA020846 to CRG.