Insulin but not Insulin Signaling modulates Nonalcoholic Fatty Liver Disease

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Non-Alcoholic Fatty Liver Disease (NAFLD) has been shown to associate with poorly-controlled diabetes type 1 and diabetes mellitus type 2 and if untreated, can progress to Non-Alcoholic Steatohepatitis (NASH), liver cirrhosis, and liver failure. Given insulin's roles in diabetes mellitus pathophysiology, we examined insulin and hepatic triglyceride accumulation in diabetes models *in vitro*, measuring IRS-1 and IRS-2 signal transduction, insulin signaling inhibition, and free fatty acid flux into hepatocytes via fatty-acid transport proteins 2 and 5. Using a diabetes mellitus type 1 murine model, we measured hepatic triglyceride accumulation in the presence and absence of exogenous insulin administration. **Methods:** *In vitro* model: AML-12 cells under varying insulin concentration and insulin signaling inhibitor were fed free fatty acids and harvested for triglyceride quantification and qPCR for IRS-1/2 and FATP-2/5. *In vivo* model: C57/Bl6 male mice were placed on high fat diet and diabetes type 1 was pharmacologically induced. Mice were injected with varying insulin concentrations and serum triglyceride, liver triglyceride, and liver histology were examined at sacrifice. **Results:** *In vitro* cell culture: Hepatic triglyceride accumulation was elevated at 0mU/mL ($p<0.01$) and 100 mU/mL ($p<0.05$) insulin. Under insulin signaling inhibition, hepatic triglyceride accumulation was significantly decreased at all insulin concentrations ($p<0.001$) compared to no inhibition. Insulin signaling inhibition showed dose-dependent IRS-1/2 expression, with IRS-2 elevated at 0 mU/mL insulin ($p<0.001$) and IRS-1 elevated at 100 mU/mL insulin ($p<0.01$). FATP-2/5 expression was not decreased by inhibition. *In vivo* murine model: As compared to HFD and 200 mU/mL injections, 0 mU/mL insulin injection showed marked increases in serum and liver triglyceride levels ($p<0.01$) and appreciable steatosis. **Conclusions:** Hepatocyte triglyceride accumulation shows a U shaped insulin dose-effect curve, and inhibition decreases triglyceride accumulation but does not down-regulate IRS or FATP expression *in vitro*. In addition, insulin administration reverses hepatic steatosis in a murine model of DMT1 on HFD. **Acknowledgments:** Rohit Kohli MD, Samir Softic MD, and Michelle Kirby and the Deptartment of Gastroenterology, Heptatology, and Nutrition at Cincinnati Children's Hospital Medical Center, Cincinnati, OH and the UCCOM MSSRP Program, T35 DK 60444.