Reduction of hepatic Z-alpha1 antitrypsin by RNA interference prevents and reverses liver disease including hepatic mitochondrial injury in the PiZ mouse model

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INTRODUCTION
Autosomal co- dominant genetic disorder alpha 1-antitrypsin deficiency (AATD) causes pulmonary and liver disease. Individuals homozygous for the mutant Z allele accumulate polymers of Z- AAT protein in hepatocytes, where AAT is primarily produced, resulting in hepatic injury and fibrosis that may over time lead to cirrhosis and hepatocellular carcinoma. The accumulated polymers cause oxidative stress, mitochondrial damage, and increased autophagic activity and apoptosis. Injury to mitochondria, dilated endoplasmic reticulum (ER), microvesicular fat and other ultrastructural changes can be visualized by electron microscopy (EM).

AIM
Adult PiZ mice were treated with RNAi-based therapeutic ARC-AAT to reverse the AATD-associated liver disease phenotype

METHODS
ARC-AAT, consisting of RNA trigger (the RNAi-AAT API) plus an endosome-release agent (EX1), was evaluated in the PiZ mouse model that harbors the human Z-AAT gene and recapitulates AATD liver disease. Male PiZ mice were 11-17 weeks old at baseline. They were treated for 32-33 weeks with biweekly (Q2W) intravenous injections of 8 mg/kg (mpk) ARC-AAT (8 mpk RNAi-AAT API + 4 mpk EX1) or with saline as a control. Additional control groups were treated with 12 mpk RNAi-AAT API alone or 12 mpk control RNAi trigger (RNAi-Luc) delivered with 6 mpk EX1.

- Plasma Z-AAT was measured by ELISA (Albcam).
- Soluble (monомерic) and insoluble (polymeric) Z-AAT in the liver were measured by semi-quantitative Western blotting.
- Globules of Z-AAT polymer aggregates were measured by PAS-D staining and ImageJ quantification, 3 fields of view per liver specimen.
- Liver inflammation was assessed by histological evaluation with H&E staining and morphometric analysis.
- Expression of genes previously implicated in liver injury and development of fibrosis was measured by RT-qPCR.

RESULTS
Z-AAT measurement
- 95-96% plasma Z-AAT reduction relative to baseline for duration of Q2W ARC-AAT.