

Supplementary Materials for

DNA Deamination Is Required for Human APOBEC3A-Driven Hepatocellular Carcinoma *In Vivo*

Running title: Human APOBEC3A as a Master Driver of Cancer

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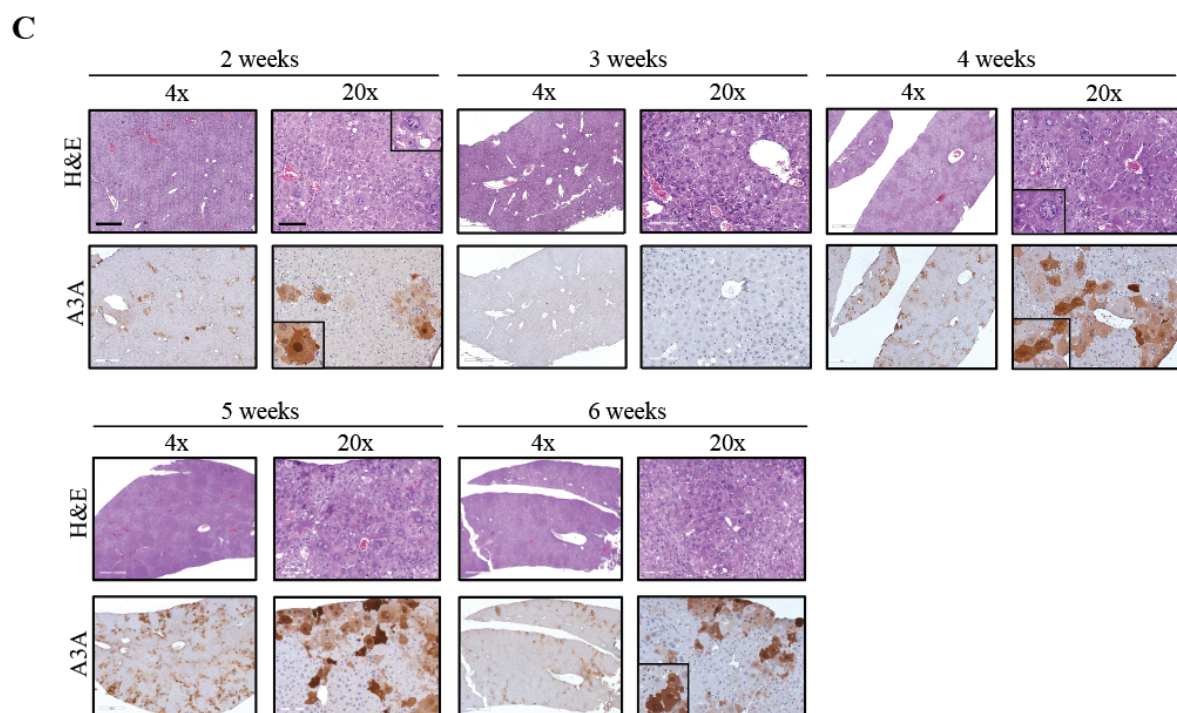
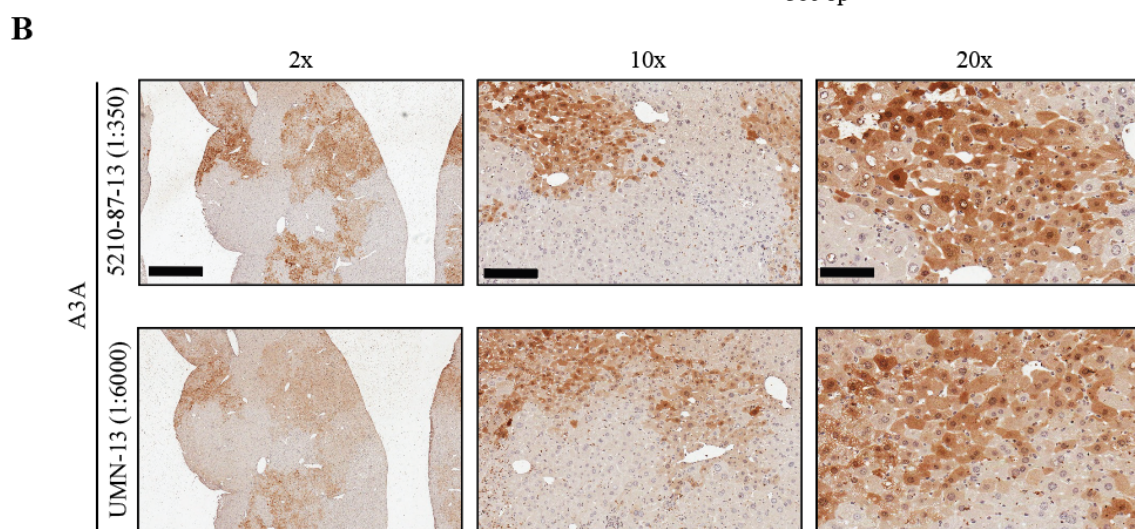
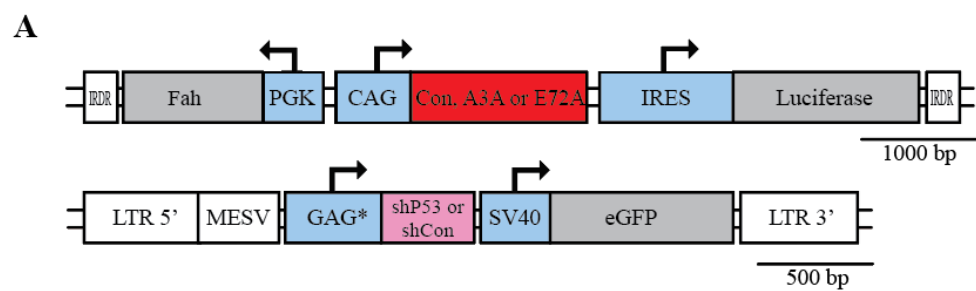
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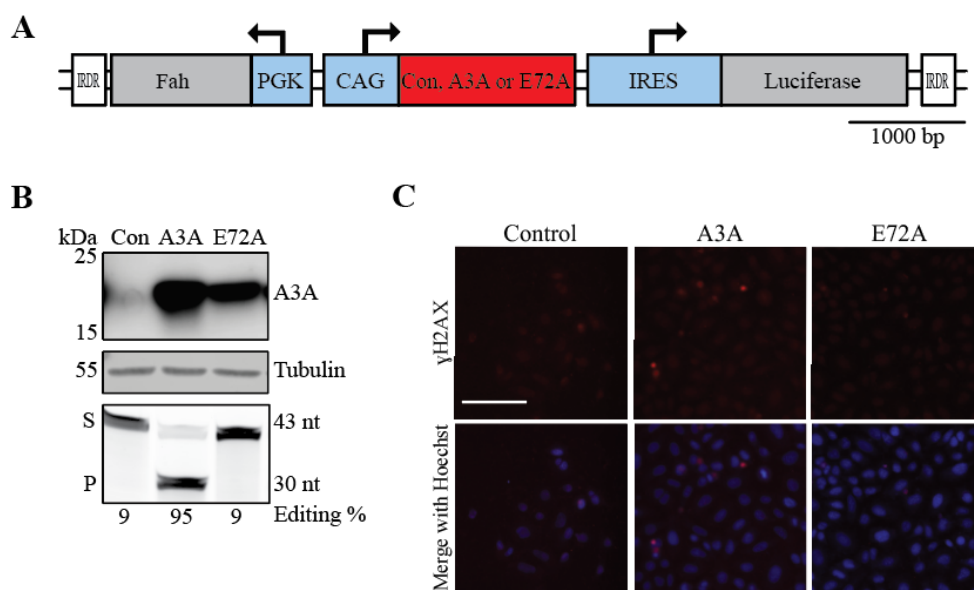
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Supplementary Information: Supplementary Figures S1-S3



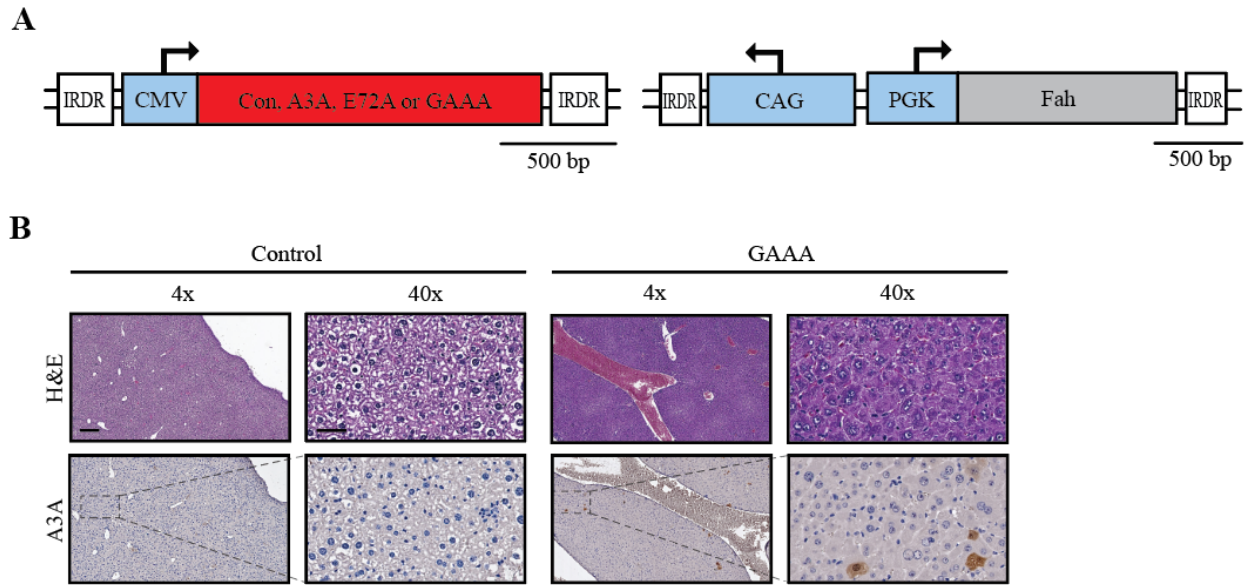
Supplementary Figure S1. Human A3A expression in murine hepatocytes.

- (a) Schematic representation of Fah-restorative and shRNA constructs used for hydrodynamic injections in Figure 1. Scale bar, 1000 bp and 500 bp, respectively.
- (b) Comparative IHC analysis of consecutive sections of A3A expressing livers stained with α -A3A monoclonal antibodies, 5210-87-13 or UMN-13, at 1:350 or 1:6,000 dilutions, respectively. Scale bar, 1mm, 200 μ m, and 100 μ m for 2x, 10x, and 20x magnification, respectively.
- (c) Representative H&E and A3A IHC (UMN-13) photomicrographs of A3A livers harvested 2-6 weeks p.i. Scale bar, 1 mm and 100 μ m for 4x and 20x magnification, respectively, and inset images are magnified 4-fold.



Supplementary Figure S2. The catalytic activity of A3A induces genomic damage.

- (a) Schematic representation of E72A construct used for hydrodynamic injections. Scale bar, 1000 basepairs.
- (b) Immunoblot (upper panels; 5210-87-13) and DNA deaminase activity assay (lower panel) of whole cell lysates of HeLa cells transfected with constructs containing A3A, E72A or control. Control (Con); substrate (S); product (P); nucleotides (nt). DNA deaminase activity quantification of band volume intensities (ratio of substrate to product) by densitometry reported below as Editing %.
- (c) Immunofluorescence analysis of transfected HeLa cells stained for γ -H2AX and counterstained for DNA with Hoescht. Scale bar, 100 μ m.



Supplementary Figure S3. A3A-GAAA mutant expression in hepatocytes.

- (a) Schematic representations of constructs used for all experiments in Figure 4. Scale bars, 500 bp.
- (b) H&E and IHC (UMN-13) of representative livers from the Control and GAAA cohorts harvested 72 hours p.i. Scale bar, 500 μ m or 50 μ m for 4x and 40x magnification, respectively.