

Supplementary Figure Legends

Supplementary Figure 1: Microscopic evaluation of DEN induced liver tumours

Haematoxylin and eosin (H&E) images from **(a)** WT and **(b)** *Casp2*^{-/-} mice showing hepatocyte karyomegaly at 10 months age. Scale bar represents 50µm. **(c)** Representative H&E image of liver tumours in WT mice. Scale bar represents 1mm. **(d)** Hepatocellular carcinoma showing neoplastic hepatocytes with nuclear atypia (arrows). Scale bar represents 100µm. **(e)** Hepatocellular carcinoma showing bi- and multi-nucleated cells. Insets showing a large multinucleated tumour cell (left) and bizarre mitotic figure (right). Scale bar represents 50µm. **(f)** Hepatocellular carcinoma with nuclear atypia and karyomegaly. Nuclear inclusions (cytoplasmic invaginations) (ii, iii) and numerous cytoplasmic inclusions (globular bodies) (ii, iv) are evident. Scale bar represents 50µm.

Supplementary figure 2: Lipid profile of WT and *Casp2*^{-/-} mice 10 months after DEN injection

(a) Total triglycerides, **(b)** cholesterol, **(c)** HDL, **(d)** LDL and **(e)** cholesterol/HDL. While serum cholesterol levels increased demonstrating, increased biliary obstruction due to tumour growth, no significant difference was observed between the groups. Values are given as mean ± SEM (n=6, PBS and n=11, DEN). Statistics were performed using Student's t-test: * p<0.05, ** p<0.01 and *** p<0.001.

Supplementary figure 3: Cleaved caspase-3 staining of WT and *Casp2*^{-/-} livers 10 months after DEN injection

Cleaved caspase-3 staining of WT and *Casp2*^{-/-} livers with brown positive staining highlighted. Scale bar represents 50µm.

Supplementary figure 4: Densitometric analysis of immunoblots

Immunoblot quantitation showing increased **(a)** JNK pathway activation. No significant difference observed for **(b)** ERK pathway. Significant increases in **(c)** Sirt1, **(d)** p53 and **(e)** p21 was observed in DEN injected *Casp2*^{-/-} mice. Band intensity was measured using Bio-Rad Image Lab software and normalised to total protein intensity.

Supplementary figure 5: TUNEL staining in mice livers following acute DEN injection

Low magnification images showing necrotic patches at 24h, with significantly reduced TUNEL positive cells in *Casp2*^{-/-} 24h after DEN injection. Scale bar represents 1mm.

Supplementary figure 6: qPCR analysis of mice livers 10 months after DEN injection

Relative **(a)** β-catenin, **(b)** p53, **(c)** mdm2, **(d)** puma and **(e)** noxa mRNA levels in mice 10 months after DEN injection. Values are given as mean ± SEM (n=4, PBS and n=8, DEN). Statistics were performed using Student's t-test. * p<0.05 and ** p<0.01.

Supplementary figure 7: Effect of DEN treatment at 6 weeks of age in one year old mice livers

(a) H&E images of mice livers and nuclear/cytoplasmic ration. Scale bar represents 50 μ m. (b) Serum TNF α and (c) IL6 levels. Values are given as mean \pm SEM (n=7, PBS and n=10, DEN). (d) Immunoblots showing activation of ERK and JNK signaling. Statistics were performed using Student's t-test. * p<0.05, ** p<0.01 and *** p<0.001.