Heterogeneous roles of Caspase8-signalling in hepatocytes and non-parenchymal cells in a model of liver stem cell activation and sclerosing cholangitis

K. Chaudhary¹, C. Liedtke², C. Trautwein², K. Streetz²

Abstract

Background and aim:
Caspase-activation plays a fundamental role in the maintenance of tissue homeostasis by clearing injured cells to maintain tissue homeostasis through receptor mediated apoptosis. An important role of caspase8 during immune-mediated liver injury and regeneration was previously demonstrated by our laboratory. We aim to examine its role during hepatic stem cell (Oval cells) activation, a process closely related to liver regeneration.

Materials and Methods:
For this approach, hepatocyte specific conditional caspase8 knockout (casp8Δhepa) animals and mice with an ubiquitous deletion of caspase8 (casp8ΔMx) were compared in the DDC (3,5-diethocarbonyl-1,4-dihydrocollidine) model of oval cell activation and secondary sclerosing cholangitis. Oval cells were isolated and characterized by flow cytometry (FACS) and real time PCR. Liver tissue was further analysed by IHC.

Results:
Higher transaminases and bilirubin levels were observed in Casp8ΔMx levels compared to wild type (WT) and control, while casp8Δhepa animals were protected after 4-weeks of DDC-feeding. Additionally, histological analysis revealed reduced liver injury and immune-cell infiltration in Casp8Δhepa mice. Thereby caspase3 and -8 activities were reduced in both knockout strains as demonstrated by caspase3 and -8 assay, suggesting other mechanisms being responsible for the phenotype. Correlating to the stronger liver injury, casp8ΔMx mice displayed more proliferation in periportal areas where LPC emerge and reside as indicated by 5-fold higher BrdU incorporation rate and significantly higher CD133, Cyclin A, D and E mRNA expression. The following analysis of hepatic progenitor cells by flow cytometry (sca-1, OC-1, -2 and -3) as well as by immunohistochemistry (CK-19) unrevealed a significant stronger (5-fold) oval cell activation in casp8ΔMx mice, while casp8Δhepa had significantly less then WT mice after 4-weeks of DDC-feeding. Deletion of caspase8 itself was not evident in isolated progenitor cells. Preliminary data now point to an additionally enhanced infiltration of immune-cells (CD45, CD4, CD11b, CD4) in casp8ΔMx mice. This finally resulted in a stronger fibrosis progression of the underlying sclerosing cholangitis induced by DDC in casp8ΔMx mice, as evidenced by an enhanced expression of collagen and α-SMA.

Conclusions:
Our data suggest a differential role of death receptor mediated liver injury through caspase8 activation in individual liver cell types. While hepatocyte specific knockout provided protection from liver damage an ubiquitous deletion of caspase8 triggered more injury and inflammation. This was finally related to a significantly stronger activation of the liver progenitor cell compartment and more tissue remodelling.