Biliary senescence drives hepatocyte VCAM-1 expression and liver neutrophil infiltration

Background: Biliary epithelial cell (BEC) senescence and associated secretory phenotype (SASP) are present in biliary diseases, but their effects on hepatocyte biology and associated liver inflammation are underexplored. This is relevant as cellular senescence and associated inflammation contribute to the pathology of biliary disease. Methods: Liver tissue from patients with biliary diseases (primary biliary / sclerosing cholangitis, biliary atresia), and from a murine model of BEC senescence and cholestasis (CK19CreERTMdm2flox/floxR26RtdTomatoLSL, DDC diet), was analysed with fluorescence microscopy. Murine BECs were treated with etoposide to induce senescence and co-cultured with naïve primary hepatocytes. The resultant conditioned medium was incubated with bone marrow-derived neutrophils in a migration assay. Results: Biliary disease patients showed: 1) increased VCAM-1+ hepatocytes (median %: 5.67 vs 0.59; P = 0.014); 2) increased p21+ BECs (median %: 17.02 vs 2.52; P < 0.001); and 4) %VCAM-1+ section correlation with %MPO+ neutrophils (rs = 0.438; P= 0.029). Neutrophils were closer to p21+ BECs than p21- BECs in human livers (median nearest distance: 20.0 vs 29.9 μm; P < 0.001); and to VCAM-1+ hepatocytes than VCAM1-hepatocytes in human (mean nearest distance: 18.0 vs 25.2 μm; P< 0.001) and murine (median nearest distance: 33.8 vs 48.2 μm; P < 0.001). Senescent murine BECs had SASP features of human biliary diseases. Hepatocytes exposed to senescent BECs displayed transcriptional overexpression of Vcam1 and increased neutrophil activity on functional enrichment analysis. Neutrophil migration was increased following treatment with conditioned medium from murine hepatocytes co-cultured for 72 hours with senescent BECs vs non-senescent BECs (P< 0.001); there was no difference in monocultures of senescent BECs vs non-senescent BECs (P =0.999). Conclusion: Neutrophils preferentially migrate towards senescent BECs in biliary diseases. Hepatocytes promote neutrophil migration in biliary senescence, potentially mediated by VCAM-1 expression. Targeting neutrophil recruitment directly, or via VCAM-1, may alter the course of biliary disease progression. Disclosures: Marisa Ferreira: Nothing to Disclose, Alastair Kilpatrick: Nothing to Disclose, Daniel Rodrigo-Torres: Nothing to Disclose, Sofia FerreiraGonzalez: Nothing to Disclose, Victoria Gadd: Nothing to Disclose, Tak Yung Man: Nothing to Disclose, Rhona Aird: Nothing to Disclose, Candice AshmoreHarris: Nothing to Disclose, Gareth Hardisty: Nothing to Disclose, Wei-Yu Lu: Nothing to Disclose, JuanCarlos Acosta: Nothing to Disclose, Luke Boulter: Nothing to Disclose, Stuart Forbes: Resolution Therapeutics: Founder