Angiopoietin-2 Promotes Pathological Angiogenesis and Is a Novel Therapeutic Target in Non-Alcoholic Fatty Liver Disease

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Background:

Angiogenesis contributes to the development of non-alcoholic steatohepatitis (NASH) and promotes inflammation, fibrosis and progression to hepatocellular carcinoma (HCC). Angiopoietin-2 (Ang-2) is a key regulator of angiogenesis. We investigated the role of Ang-2 and its potential as therapeutic target in NASH using human samples, in vivo mouse models and in vitro assays.

Methods:

Serum Ang-2 levels were determined in 104 obese patients undergoing bariatric surgery and concomitant liver biopsy. The effect of the Ang-2/Tie2 receptor inhibiting peptibody L1-10 was evaluated in the methionine-choline deficient (MCD) and streptozotocin-western diet mouse models, and in vitro on endothelial cells and bone marrow-derived macrophages. Liver histology, immunohistochemistry, cytokine expression and flow cytometric analyses were performed. The hepatic vasculature was visualized with μCT scans and scanning electron microscopy of vascular casts. Gene expression analysis was performed on FACS-isolated liver endothelial cells and monocytes.

Results:

Serum Ang-2 levels were increased in patients with histological NASH compared to patients with simple steatosis, and correlated with hepatic CD34 immunoreactivity. In accordance, serum and hepatic Ang-2 levels were increased in mice with steatohepatitis. L1-10 treatment reduced hepatocyte ballooning and fibrosis in MCD fed mice, both in a preventive and therapeutic setting, and this was associated with reduced angiogenesis and normalization of the vascular micro-architecture. Liver-isolated endothelial cells and monocytes from MCD fed L1-10-treated mice showed reduced expression of leukocyte adhesion and inflammatory markers, respectively, compared to cells from untreated MCD fed mice. In the streptozotocin-western diet model, therapeutic Ang-2 inhibition was able to reverse NASH and attenuated HCC progression. L1-10 treatment mitigated the increased cytokine production in LPS-stimulated endothelial cells, but not macrophages.
**Conclusion:** Our data indicate significant potential for the inhibition of Ang-2 signaling as a novel strategy to target pathological angiogenesis and halt the progression of non-alcoholic fatty liver disease.