**EDITORIAL**

# Hepatic fibrosis developing in morbid obesity independent of steatohepatitis: new mechanism or the Rube Goldberg machine?

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Obesity represents a major health burden. Nonalcoholic fatty liver disease (NAFLD) is strongly associated with visceral obesity. NAFLD encompasses a broad spectrum of pathology from steatosis alone, to nonalcoholic steatohepatitis (NASH), with or without fibrosis. As the prevalence of steatosis and NASH was recently estimated to be 46 and 12 % [1], respectively, understanding the mechanism of fibrosis that leads to hepatic complications is crucial. In the current issue of *Hepatology International*, Ciupinska-Kajor et al. [2] present data suggesting the mechanism of fibrosis in NAFLD might differ based on the degree of adiposity, and that early angiogenesis could be the distinguishing feature.

Prognosis in NAFLD is variable, dependent on histologic findings. Bland steatosis alone has a benign prognosis, whereas NASH leads to increased fibrosis and mortality. A recent review of natural history data spanning a mean follow-up of 15 years found a 0.7 % prevalence of cirrhosis in cohorts with steatosis alone, compared to a 10.8 % prevalence of cirrhosis in cohorts with NASH [3]. Steatohepatitis was also shown to be associated with increased mortality. When 118 persons with NAFLD were followed up for a mean period of over 20 years, persons with NASH had a significant increase in mortality, whereas those with steatosis alone did not [4].

With different prognostic ramifications, it is important to understand the pathophysiologic mechanisms that distinguish bland steatosis from NASH. Unfortunately, events that differentiate NASH from steatosis are not well defined.

Theories do exist. The "two-hit" hypothesis involves a sequential process where first steatosis develops and a second hit is caused by oxidative stress resulting in lipid peroxidation and steatohepatitis [5]. A second theory involves "multiple parallel hits" [6]. These multiple hits include gut-derived signals such as endotoxin, adipose-derived signals such as adipocytokines, innate immunity, endoplasmic reticulum stress, and genetic differences. Important to this hypothesis is the potential for inflammation to precede steatosis in some cases. Different theories highlight the concept that NAFLD and NASH represent a heterogenous group of disorders where a histologic endpoint can be represented by numerous contributing pathogenic mechanisms.

One pathophysiologic mechanism of progressive liver disease in NAFLD is angiogenesis. Angiogenesis is associated with hepatic inflammation [7], hepatic fibrosis [8], the formation of portosystemic collateral vessels [9], and hepatic carcinogenesis [10]. In animal models of steatohepatitis, the expression of vascular endothelial growth factor (VEGF) and angiogenesis occurred in parallel to fibrosis and carcinogenesis [11]. In human tissue, angiogenesis, measured by CD 34-positive staining cells, occurred in NASH, but not normal liver or bland steatosis [12, 13]. Angiogenesis in NASH was associated with both hepatocellular apoptosis and insulin resistance [13]. The association between hepatic inflammation and angiogenesis is not unique to NAFLD. In persons with chronic hepatitis C, serum levels of proangiogenic markers, VEGF and angiopoietin-2, were elevated at baseline and decreased after interferon-based therapy [14].

The study by Ciupinska-Kajor et al. [2] mirrors the potential heterogeneity seen in the pathogenesis of NAFLD. The authors examined angiogenic markers, including VEGF A and CD 34 staining, in a population of morbidly

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obese (BMI >40 kg/m$^2$) and nonobese (BMI <30 kg/m$^2$) persons. NASH was diagnosed at an equivalent rate in the morbidly obese and the nonobese groups. Despite a similar prevalence of NASH, morbidly obese persons had a higher rate of fibrosis (82.5 vs. 43.5 %, $P = 0.003$) and were more likely to have advanced fibrosis (15 vs. 6.7 %, $P < 0.005$). All markers of angiogenesis (VEGF A, Flk-1, and CD 34) were more common in obese than nonobese subjects; however, this effect originated from the cohorts with bland steatosis and "borderline" NASH. There was no difference in expression of angiogenic markers comparing obese and nonobese subjects in the group defined as definite NASH.

A novel finding in the current manuscript is angiogenesis and development of fibrosis in morbidly obese persons with steatosis alone. This contrasted with nonobese persons where angiogenesis and fibrosis occurred mostly in the setting of NASH. There are ample data to support that adipose tissue can adversely affect the liver, and visceral fat is key. A study with labeled fat showed that the majority of free fatty acids encountered by the liver originated from the adipose tissue [15]. The visceral adipose tissue also functions as an endocrine organ releasing adipocytokines into the portal circulation. There are data to support the visceral adipose production of interleukin-6 (IL-6) adversely affecting the liver. In patients undergoing bariatric surgery, IL-6 concentrations were almost 50 % higher in the portal vein compared to the radial artery [16]. The majority of circulating IL-6 has been shown to be taken up by the liver [17] where it can result in hepatic insulin resistance [18] and was found to increase hepatic synthesis of acute phase reactants such as C-reactive protein, serum amyloid A and fibrinogen [19]. Other adipocytokines linked to visceral adipose tissue include visfatin, interleukin-8, adipsin, angiotensinogen, and resistin [20].

The finding that angiogenic mechanisms are activated in bland steatosis contrasts with other data on NAFLD [12, 13]. The current manuscript differs from other studies with the finding observed in those with morbid obesity, a population not included in the contrasting studies. This novel finding requires further validation and limitations must be recognized. The NAFLD Activity Scale (NAS) was used for the diagnosis of NASH. Although this scoring system has been used in multiple studies to diagnose NASH, the authors who defined this scoring system pointed out that the histopathologic diagnosis of NASH and a NAS score $\geq 5$ are not equivalent [21]. One might argue that the presence of fibrosis and steatosis is suggestive of NASH, regardless of the degree of lobular inflammation or hepatocyte balloon degeneration. A second observation important in NAFLD is the presence of inflammatory features have been shown to fluctuate over time. This has been demonstrated in clinical trials with serial biopsies performed in placebo arms. A recent placebo-controlled trial with 96 weeks of follow-up found resolution of NASH in 21 % of the placebo arm [22]. Sampling variability also remains an issue in NAFLD trials. In the earlier mentioned study [22], an additional relevant finding was sampling variability on different "cuts" of the same core biopsy. Logically, a morbidly obese person with steatosis and stage 2 fibrosis might have had steatohepatitis at some point in the past, or even in a different sample of a biopsy.

Another conclusion deserves further comment. The authors commented that the similar rates of steatosis and NASH in the nonobese and obese cohorts as disagreeing with the opinion that NASH is more common in individuals with morbid obesity. As the authors later pointed out, a significant selection bias of the study population exists. The persons with morbid obesity were consecutive persons prior to bariatric surgery, whereas the nonobese cohort was selected based on indication (increased aminotransferases and presumed steatosis on ultrasound). Especially notable is any biopsy without steatosis was excluded. This selection bias negates the ability to make prevalence comparisons between the two populations.

The current manuscript raises the interesting possibility of hepatic fibrosis resulting from a morbid degree of adiposity and mediated through angiogenic signals. The endocrine function of visceral adipose tissue is supported by ample evidence. A hypothesis could be generated that in persons with morbid obesity expression of cytokines and proangiogenic signals from visceral adipose tissue result in hepatic fibrosis without universally causing steatohepatitis first. The implications for these data are quite interesting. Might this be a mechanism for the high rates of fibrosis observed in persons undergoing bariatric surgery? Could early angiogenesis account for hepatocellular carcinoma in obese persons with NAFLD and no cirrhosis? Or is this an overcomplicated observation further proving that steatohepatitis is a histologically elusive. For these questions, further validation and mechanistic studies will be the arbitrator.

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