

# Nonalcoholic Fatty Liver Disease: A Review of Current Understanding and Future Impact

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Nonalcoholic fatty liver disease (NAFLD), already the most common form of liver disease in the United States, can be expected to increase in prevalence and severity in parallel with national epidemics of obesity and type 2 diabetes. NAFLD is frequently associated with insulin resistance. While insulin resistance, and thereby hyperinsulinemia, are, in large part, metabolic consequences of obesity, the basis of diversity in severity and progression of inflammation and fibrosis is not known. Increased susceptibility to oxidative stress is likely to play a role. Several patient characteristics have been associated with more severe histological findings in patients with NAFLD, including type 2 diabetes, hypertension, age over 40 years, and higher transaminases. Liver biopsy is, however, required to accurately grade and stage NAFLD histologically. Although the natural history of NAFLD is relatively poorly defined, NAFLD is increasingly recognized as an important cause of decompensated liver disease. Weight reduction and improved insulin sensitivity are associated with improved biochemical and histological parameters of NAFLD. There are, however, no proven safe and efficacious pharmacological treatments for NAFLD.

The term *nonalcoholic fatty liver disease* (NAFLD) is used to describe a spectrum of histologic findings ranging from simple steatosis to nonalcoholic steatohepatitis (NASH) with progressive fibrosis and liver failure. On the basis of current prevalence of obesity and type 2 diabetes, NAFLD can be estimated to affect between 6–30 million people in the United States, including more than 600,000 with cirrhosis.<sup>1,2</sup> With epidemics of obesity and type 2 diabetes in North America, NAFLD has become an important emerging public health issue. As the prevalence and severity of obesity continue to increase in the United States,<sup>3–6</sup> with concomitant increases in the prevalence of type 2 diabetes and dyslipidemias, the prevalence of all grades of NAFLD can also be expected to increase.

NASH is characterized by histopathologic features similar to those associated with alcohol-induced liver injury, in the absence of excessive alcohol ingestion.<sup>7</sup> The histologic characteristics of NASH are macrovesicular steatosis, nu-

clear glycogenation, lobular and portal inflammation, and, occasionally, Mallory's hyaline.<sup>7,8</sup> NASH is almost always a chronic condition and is most frequently associated with obesity (central, as measured by waist circumference, and overall, as measured by body mass index [BMI]) and type 2 diabetes mellitus.<sup>7,9–15</sup> NASH can be a severe, progressive form of liver disease, leading to the development of cirrhosis.<sup>12,13</sup> The overall prevalence of NASH in adults in North America, based on large autopsy-based analysis, has been reported to be 18.5% in obese and 2.7% in nonobese individuals.<sup>12</sup> Of obese individuals found to have NASH at autopsy, of which most cases were not suspected ante mortem, 13.8% had bridging fibrosis or cirrhosis. The corresponding figure for lean individuals was 6.6%.<sup>12</sup> A more recent study of the clinicopathologic features of 32 patients with NASH found the prevalence of cirrhosis to be 8%.<sup>14</sup>

## Clinical Features/Diagnosis

Most patients who are ultimately diagnosed as having NASH are referred for evaluation of abnormal liver biochemistries, often detected serendipitously. In contrast to the ratio seen in alcoholic liver disease, aminotransferase levels are typically increased  $\leq 4 \times$  normal, with ALT usually greater than AST.<sup>11,13,16</sup> Alkaline phosphatase level is usually elevated  $\leq 2 \times$  normal, with bilirubin levels usually within the normal range.<sup>11,13,16</sup>

In patients with abnormal liver biochemistries, a detailed history is essential to exclude, or otherwise, the presence of excessive alcohol consumption, steatohepatitis inducing pharmacotherapy, surgical procedures, and occupational exposure to hepatotoxins. A nutritional history, particularly of rapid weight gain or loss, is also important. The great majority of the clinical conditions that are associated with the development of steatohepatitis can readily be excluded once a thorough history has

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*Abbreviations used in this paper:* BMI, body mass index; NAFLD, nonalcoholic fatty liver disease; NASH, nonalcoholic steatohepatitis; ROS, reactive oxygen species.

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been elicited. Of those clinical conditions that are associated with NASH that cannot be excluded by simple history taking, Wilson's disease, viral hepatitis, and autoimmune liver disease require serologic/biochemical exclusion. The great majority of patients with NAFLD will have 1 or more features of the metabolic syndrome (increased waist circumference, hypertriglyceridemia, low high-density lipoprotein cholesterol, hypertension, and a fasting glucose of 110 mg/dL or higher).<sup>17–21</sup>

On direct questioning, a minority of patients with NASH describe excessive fatigue and/or right upper quadrant pain.<sup>22</sup> It is not clear whether these symptoms are more common among patients with NASH than among age, gender, and BMI matched individuals without NASH.

An ultrasonographic examination of the liver will detect hepatic steatosis with a sensitivity of between 66%–100%,<sup>23–25</sup> although the sensitivity is reduced for degrees of steatosis less than 30%. The same might be said of computed tomography, magnetic resonance imaging, and radionuclide techniques, all of which have findings that are characteristic of hepatic steatosis. However, none of these techniques are able to distinguish simple steatosis from steatohepatitis with progressive fibrosis. Because NASH is by definition a clinicohistologic entity, histology is required to confirm the diagnosis.

### The Role of Liver Biopsy

The role of liver biopsy in the diagnosis and management of NAFLD is still evolving. In addition to determining severity of disease, a liver biopsy can also be helpful in determining the effects of medical treatment. On the other hand, liver biopsies are associated with morbidity and cost. Limiting biopsies to patients at greatest risk for histologically progressive NAFLD is an obvious clinical goal. Several factors have been associated with a greater odds ratio of finding more severe inflammation grade and/or fibrosis stage on liver biopsy. These include age  $\geq 45$  years, BMI  $\geq 30$  kg/m<sup>2</sup>, presence of type 2 diabetes mellitus, and a ratio of AST to ALT of 1.<sup>26</sup> Among overweight patients (BMI  $\geq 25$  kg/m<sup>2</sup>) with abnormal liver biochemistries,  $\geq 2$  stage fibrosis has been reported to be independently associated with age  $\geq 50$  years (odds ratio, 14.1), BMI  $\geq 28$  kg/m<sup>2</sup> (odds ratio, 5.7), triglycerides  $\geq 1.7$  mmol/L (odds ratio, 5), and ALT  $\geq 2N$  (odds ratio, 4.6). A score combining age, BMI, triglycerides, and ALT has been reported to have a 100% negative predictive value for septal fibrosis when scoring 0 or 1 (100% sensitivity for a specificity of 47%).<sup>27</sup> Similarly, in a small study of patients with class II obesity (BMI  $\geq 35$  kg/m<sup>2</sup>), the presence of 2 factors (out

of 3 including increased index of insulin resistance, systemic hypertension, and increased ALT) had a sensitivity of 0.8 and specificity of 0.89 for NASH.<sup>28</sup>

All of the information that we have regarding identifying patients with more histologically severe NAFLD has important limitations. First, the study populations were highly selected (eg, were referred for a specialty consultation and had abnormal liver biochemistries, or were performed only in patients with class II obesity undergoing bariatric surgery). Thus, although of potential utility in population studies, none of the factors associated with more severe histologic injury are of clear utility in the management of individual patients who might have inflammation and fibrosis despite normal transaminase levels. Indeed, the entire histologic spectrum of NAFLD can be seen in individuals with normal ALT values, and the histologic spectrum is not significantly different among patients with normal ALT levels from those with elevated ALT levels.<sup>19</sup> Second, there are no prospective studies validating any of the predictive models or determining their predictivity for future histologic progression. In short, if confirming the diagnosis and/or degree of histologic severity of NAFLD is thought to be important, a biopsy should be considered. Conversely, if the results of a liver biopsy are unlikely to affect management, or if the risk outweighs any potential benefit, a presumptive diagnosis should be made. A presumptive diagnosis can be based on suggestive clinical findings (eg, features of the metabolic syndrome), abnormal transaminase levels, suggestive imaging (eg, increased echogenicity on ultrasound), and negative work-up for other etiologies (eg, viral, metabolic, and autoimmune liver diseases and pharmacotherapy).

### Pathogenesis of Nonalcoholic Fatty Liver Disease

Although there are many conditions that can be associated with steatosis and/or steatohepatitis, the terms *NAFLD* and *NASH* almost always refer to steatosis and steatohepatitis associated with obesity and insulin resistance/hyperinsulinemia.

It has been proposed that progression from simple steatosis to steatohepatitis and to advanced fibrosis results from 2 physiologic events ("hits").<sup>29</sup> The first event is thought to be insulin resistance, leading to the accumulation of fat within hepatocytes and associated increased lipid peroxidation. Second, oxidative stress increases, precipitating cytokine release and, ultimately, Fas ligand mediated hepatocellular injury.

### Lipid Metabolism in Nonalcoholic Fatty Liver Disease

The net accumulation of fat within hepatocytes, a prerequisite for NAFLD in general, could potentially result from alterations in the uptake, synthesis, degradation, or secretory pathways of hepatic lipid metabolism. The rate of appearance of fatty acids within hepatocytes can increase through increased (1) hepatocyte FFA synthesis, (2) uptake of circulating FFAs derived from peripheral fat stores, and (3) extraction of FFAs through hydrolysis of chylomicrons via increased lipoprotein lipase activity.

Esterification of FFAs with glycerol-3-phosphate to form triglycerides and phospholipids occurs in the presence of insulin and glucose. The rate of hydrolysis of triglycerides increases as insulin levels decrease. Hyperinsulinemia, as occurs in insulin resistance, is, in contrast, associated with increased triglyceride formation and diminished rates of hydrolysis, both peripherally and within the liver. Furthermore, apolipoprotein B-100, a rate-determining step in triglyceride and FFA export from hepatocytes, is also diminished by hyperinsulinemia and in patients with NASH.<sup>30</sup>

The role of leptin, an adipocyte-derived antiobesity hormone, in steatosis and insulin resistance in NAFLD is not clear. Relative leptin deficiency has been proposed to contribute to steatosis in NAFLD,<sup>31,32</sup> as has leptin excess.<sup>33,34</sup> Leptin-deficient mice develop obesity and steatohepatitis. Increased lipogenesis in both liver and adipose tissue of leptin-deficient mice has been suggested as a mechanism.<sup>32</sup> More recently, the transcription factor sterol regulatory element-binding protein-1 (SREBP-1), which plays an important role in the regulation of lipogenesis *in vivo*, has been shown to be important in the development of hepatic steatosis in the leptin-deficient mouse model of fatty liver disease.<sup>35</sup>

Obesity, when associated with hyperinsulinemia and insulin resistance, is associated with a number of metabolic effects relevant to the development of hepatic steatosis. These include increased absolute hepatic FFA uptake, increased esterification of hepatic FFAs to form triglycerides, increased FFA synthesis from cytosolic substrates, decreased apolipoprotein B-100 synthesis with subsequent decreased export of FFAs and triglycerides, decreased hydrolysis of triglycerides, diminished hepatic triglyceride and FFA export, and increased beta oxidation of mitochondrial long-chain fatty acids. Although the relative contribution of these effects to the net retention of fat within hepatocytes is not known, each of the described potential contributing mechanisms to he-

patic steatosis might be predicted to occur as a result of insulin resistance/hyperinsulinemia.

For an individual patient, it is impossible to exclude a genetic basis of hepatic steatosis. Increased hepatic synthesis of lipids could result from mutations that cause leptin deficiency or leptin receptor inhibition, for example. Genetic causes of decreased hepatic lipid export can also be postulated. However, the high prevalence of obesity-associated steatosis and the fact that hepatic steatosis can readily be explained on the basis of well-established physiologic effects of insulin resistance and hyperinsulinemia both point to steatosis as a nongenetic event in NAFLD.

### Insulin Resistance in Nonalcoholic Fatty Liver Disease

The conditions most commonly associated with NAFLD, obesity, type 2 diabetes, and metabolic syndrome, are heterogenic, multifactorial diseases. On the basis of the third National Health and Nutrition Examination Survey, the overall prevalence of metabolic syndrome in adults has been estimated to range from 24% (for individuals older than 20 years) to 40% for people older than 60 years.<sup>36</sup>

Both genetic and environmental factors are probably important in the pathogenesis of insulin resistance in NAFLD. Studies of high-risk populations suggest that insulin resistance, including patients with type 2 diabetes, has a genetic component that contributes to susceptibility to insulin resistance.<sup>37,38</sup> The contribution of genetic factors to the risk for insulin resistance and type 2 diabetes appears to be small, however.<sup>39,40</sup> Although genetic mutations in the insulin receptor occur, they are rare.<sup>41,42</sup> Although many genes might contribute to an insulin-resistant phenotype, no genetic defect has been found as the basis for insulin resistance in type 2 diabetes.<sup>43</sup>

Obesity is strongly correlated with insulin resistance,<sup>44–46</sup> particularly when central or truncal.<sup>47,48</sup> Several observations support a causal effect of obesity for insulin resistance. These include (1) obese patients with type 2 diabetes who lose sufficient weight to generate a BMI less than 30 to normalize insulin sensitivity,<sup>49,50</sup> with associated increased glycogen synthase activity<sup>49</sup> and restoration of insulin receptor kinase activity to normal levels,<sup>51</sup> and (2) weight loss can prevent progression of glucose intolerance to type 2 diabetes.<sup>52–55</sup>

Obesity is generally associated with multiple acquired factors predisposing to insulin resistance including sedentary lifestyle,<sup>56</sup> high-fat diets,<sup>57</sup> medications (eg, thiazide diuretics),<sup>58</sup> and glucose toxicity.<sup>59</sup> Although the precise mechanism of truncal obesity-associated insulin resistance is not known, release of FFAs from abdominal adipocytes into the portal circulation with subsequent

induction of hepatic insulin resistance and stimulation of glucose<sup>45</sup> is likely to contribute.

In addition to the metabolic effects of obesity described above, increased abundance of several proteins, the regulation of which is unclear, has been associated with an inhibition of insulin action. These include Rad (ras associated with diabetes)<sup>60</sup> and PC-1 (a membrane glycoprotein that has a role in insulin resistance),<sup>61</sup> which reduces insulin-stimulated tyrosine kinase activity. Tumor necrosis factor- $\alpha$ ,<sup>62</sup> which down-regulates insulin-induced phosphorylation of insulin-receptor substrate-1 and reduces the expression of the insulin-dependent glucose-transport molecule Glut4, might also be involved in NAFLD-associated insulin resistance.

It has been proposed that leptin, which has been reported to be increased in patients with NAFLD,<sup>34,63,64</sup> is a source of hepatic insulin resistance in NAFLD.<sup>35</sup> Another potentially important factor in insulin resistance in NAFLD is adiponectin (formerly called adipocyte complement-related protein of 30 kd).<sup>65</sup> Adiponectin is a 30-kd collagen-like protein related to C1qA, B, and C components of the complement system. Expression of adiponectin is reduced in obese mice and humans, particularly in obese individuals with type 2 diabetes. Plasma triglycerides, postprandial plasma glucose levels, and insulin sensitivity have all been shown to inversely correlate with adiponectin levels.<sup>66,67</sup> Weight loss and treatment with thiazolidinediones increase plasma adiponectin in animals<sup>68</sup> and humans.<sup>69</sup> Administration of adiponectin to obese or diabetic mice reduces food intake, tissue triglycerides, and plasma glucose levels, and it increases insulin sensitivity and muscle FFA oxidation.<sup>68,70,71</sup> The recently identified peptide hormone "resistin" (named in recognition of its association with insulin resistance) might represent a link between obesity and insulin resistance.<sup>72</sup> Administration of anti-resistin antibody reduces blood glucose levels and increases insulin sensitivity in obese mice, whereas administration of recombinant resistin to normal mice affects both factors negatively.<sup>72</sup>

For the great majority of patients with NAFLD, insulin resistance seems likely to be a metabolic consequence of obesity.

### **Oxidative Stress in Nonalcoholic Fatty Liver Disease**

Although the link(s) between hepatic steatosis, inflammation, and fibrosis are not well established, increased oxidative stress, a feature of both animal models of steatohepatitis<sup>73</sup> and humans with NAFLD,<sup>15,74</sup> is likely to play an important role. A proportion of intrahepatic lipid excess occurs in the form of unsaturated

FFAs. The presence of unsaturated FFAs will result in increased lipid peroxidation by inducible hepatic microsomal cytochromes CYP2E1 and CYP4A,<sup>75</sup> a highly pro-oxidant process. Extensive lipid peroxidation also occurs in cytochrome P-450 2E1 knockout mice, suggesting that cytochrome P-450 4A enzymes might be the major contributor to microsomal lipid peroxidation.<sup>75</sup> The observation that, in a genomic analysis of histologically progressive NASH, mRNA for P-450 4A is underexpressed when compared to controls with other forms of liver disease suggests that up-regulation of microsomal cytochromes might be pretranscriptionally impaired, further contributing to hepatic steatosis.<sup>76</sup>

When pro-oxidant pathways generate more reactive species than can be consumed by antioxidant pathways (eg, via protein disulfide isomerase or reduced glutathione peroxidase), oxidative stress occurs, with resulting accumulation of reactive oxygen species (ROS, chiefly superoxide and hydroxyl radicals plus hydrogen peroxide). ROS can produce hepatocellular injury through several mechanisms, including direct inhibition of mitochondrial respiratory chain enzymes, inactivation of glyceraldehyde-3-phosphate dehydrogenase, inhibition of membrane Na/K adenosine triphosphatase activity, inactivation of membrane sodium channels, and other oxidative protein modifications. ROS are potent triggers of DNA strand breakage, with subsequent activation of the nuclear enzyme poly-adenosine 5'-diphosphate ribosyl synthetase, and eventual severe energy depletion of the cells. Mitochondrial injury (as manifested by megamitochondrion) is a hallmark of NAFLD.<sup>77-79</sup> In addition, ROS further induce lipid peroxidation, cytokine production, and induce Fas ligand, all of which might contribute to the hepatocellular injury and fibrosis.<sup>80,81</sup> Lipid peroxidation also has the potentially important effect of resulting in the production of malondialdehyde and 4-hydroxynonenal, which serve as chemoattractants for neutrophils (necroinflammation), stimulate hepatic stellate cells (fibrosis), and up-regulate transforming growth factor- $\beta$ 1 expression in macrophages (fibrosis).<sup>82</sup> Finally, ROS mediate release of tumor necrosis factor- $\alpha$  by Kupffer cells, adipose tissue, and hepatocytes.<sup>83</sup> Tumor necrosis factor- $\alpha$  increases mitochondrial permeability, impairs mitochondrial respiration, and causes depletion of mitochondrial cytochrome *c*.<sup>84,85</sup>

Both tumor necrosis factor- $\alpha$  induced caspase activation and hepatocyte death (apoptosis) are increased in NAFLD.<sup>86</sup> ROS-induced Fas ligand expression by hepatocytes is thought to contribute to hepatocyte death in NAFLD.<sup>82</sup> Oxidative stress might be exacerbated by increased mitochondrial production of ROS as a result of impaired electron flow, as occurs in obesity.<sup>80</sup>

**Table 1.** Published Experience of Treatment of NASH in Humans

Treatment (reference)	No. of patients	Duration of treatment (mo)	Biochemistry	Histology
Diet + exercise <sup>91</sup>	31	15	Improved	n/a
UDCA <sup>97</sup>	24	12	Improved	Improved
UDCA + diet × 3 <sup>103</sup>	126	24	No change	No change
Probucol <sup>98</sup>	27	6	Improved	n/a
Clofibrate <sup>97</sup>	16	12	No change	No change
Gemfibrozil <sup>99</sup>	46	1	Improved	n/a
Betaine <sup>102</sup>	8	12	Improved	Improved
Vitamins E & C <sup>114</sup>	45	6	No change	Improved
Vitamin E <sup>100</sup>	11	4–10	Improved	n/a
Metformin <sup>115</sup>	20	4	Improved	n/a
Troglitazone <sup>116</sup>	10	3–6	Improved	Improved
Rosiglitazone <sup>117</sup>	30	12	Improved	Improved
Pioglitazone + vitamin E <sup>105</sup>	20	12	Improved	Improved

n/a, not available.

Increased oxidative stress usually results in increased synthesis of protective antioxidant pathways and ROS scavengers. Recently published data might be important in this regard. In a genomic analysis of histologically progressive NASH, 3 genes involved in the dismutation of ROS (catalase, glutathione peroxidase, and Cu/Zn superoxide dismutase) were diminished in subjects with cirrhosis caused by NASH.<sup>87</sup> This suggests a possible pretranscriptional basis of increased oxidative stress in patients with histologically progressive NASH. Decreased mRNA levels for all 3 ROS scavengers in patients with histologically progressive NASH suggest that the basis is likely to be at the level of transcription factor activation or synthesis.

Attenuated expression of ROS scavengers is of potential importance in NASH because of the proximity of the electron transport system, where ROS are produced as a by-product of ATP production, to the mitochondrial genome. Diminished expression of genes involved in DNA repair/metabolism will enhance oxidative stress with subsequent mutation and deletion of mitochondrial DNA. Impaired mitochondrial function will promote increased lipid peroxidation,<sup>88</sup> inducing further generation of ROS. Dysregulation of ROS scavenger synthesis would account for many of the observed changes that occur in NAFLD/NASH and might play a role in the differential histologic effects of hepatic steatosis that are seen between patients.

## Treatment

Table 1 summarizes the published experience of treatment of NASH in humans.

In lieu of a proven efficacious and safe pharmacotherapy for NASH, treatment of NASH should focus on the associated conditions. In obese patients, who make up the majority of patients with NASH, treatment should

be centered around weight loss and exercise programs. Although only limited data are available, weight reduction has been shown to be efficacious in the treatment of NASH in both adults and children as measured by aminotransferase levels and ultrasonographic evidence of steatosis.<sup>22,89,90</sup> Attainment of an ideal body weight for height is not a prerequisite for improvement in aminotransferase levels and ultrasonographic evidence of steatosis.<sup>22,89–91</sup> Rapid weight loss can exacerbate steatohepatitis and should be avoided (eg, as a result of starvation diets or bariatric surgeries).

For many obese patients sustained weight loss and exercise are, unfortunately, difficult to achieve. This has led to a proliferation of empirical and semiempirical studies of pharmacotherapy of NASH. Most studies of pharmacotherapy of NASH have been small, with only a few being randomized with placebo controls. Histologic follow-up is also lacking in many studies of potential treatments of NASH.

Improved glycemic control will lower lipid levels in patients with NASH who have type 2 diabetes mellitus (approximately one third of NASH patients). Glycemic control in the absence of weight loss will not, however, improve aminotransferase levels in this patient population.<sup>92</sup> A recent study demonstrated that metformin administration was associated with reversal of histologic changes of steatohepatitis in a mouse model.<sup>93</sup>

Patients with TPN-associated steatohepatitis who are choline deficient (choline is often absent from TPN formulas) have been shown to resolve steatosis with choline supplementation.<sup>94,95</sup> In an animal model of TPN-associated NASH, polymyxin B has been reported to decrease steatohepatitis, presumably through attenuating tumor necrosis factor- $\alpha$  production by reducing hepatic exposure to bac-

terial endotoxins.<sup>96</sup> A beneficial effect of polymyxin B has not been demonstrated in humans.

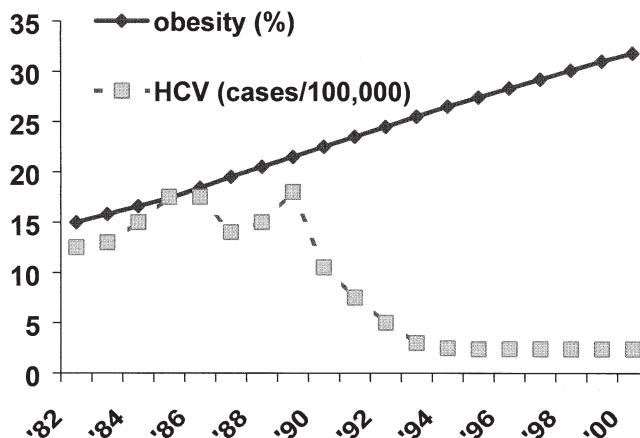
Metronidazole has been reported to be effective in improving steatosis in patients who develop NASH after jejunoileal bypass surgery in a small uncontrolled study.<sup>90</sup> Clearly, more and better studies are needed before this can be recommended as routine therapy in this setting.

Very limited data are available for clofibrate,<sup>97</sup> probucol (a lipid lowering agent with antioxidant properties),<sup>98</sup> gemfibrozil,<sup>99</sup> vitamin E,<sup>100</sup> n-acetylcysteine,<sup>101</sup> and betaine<sup>102</sup> in NASH. There is no conclusive evidence for a beneficial effect for any of these agents to date. Ursodeoxycholic acid treatment was associated with biochemical improvement in NASH in a pilot study.<sup>97</sup> In a subsequent randomized, placebo-controlled study, however, ursodeoxycholic acid treatment for 2 years was associated with improvements in liver biochemistries and histology but at a similar frequency to that of the placebo group.<sup>103</sup> Whether higher doses of ursodeoxycholic acid, as used in primary biliary cirrhosis, are effective in NASH is still the subject of study.

On the basis of our understanding of the pathogenesis of NAFLD/NASH, insulin sensitization is an appealing approach to treatment. An early report suggested histologic and biochemical improvement in patients with NASH after therapy with a thiazolidenedione.<sup>104</sup> Unfortunately, although pioglitazone produces histologic and biochemical improvement in patients with NASH, it is associated with a significant increase in BMI as well as possible idiosyncratic hepatotoxicity.<sup>105</sup> Because PPAR- $\gamma$  agonists are adipogenic by nature, weight gain is likely to be a class effect of thiazolidenediones and might well negate any histologic benefit. Combined PPAR- $\gamma$ /- $\alpha$  agonists seem beneficial in animal models of steatohepatitis<sup>106</sup> but might be limited by the excess morbidity and mortality associated with PPAR- $\alpha$  agonists in large cohort studies.<sup>107,108</sup> Preliminary results of selective PPAR- $\alpha$  agonism in an animal model of NASH have been encouraging, with histologic and biochemical improvement after short courses of PPAR- $\alpha$  agonism in methionine- and choline-deficient mice.<sup>109</sup>

### Liver Transplantation for Nonalcoholic Steatohepatitis: Clues to the Future Burden of Fatty Liver Disease

Given the relentless increase in the prevalence and severity of obesity in North America, combined with younger age of onset, the frequency of NAFLD as an indication for liver transplantation would be expected to increase over time. Unfortunately, the United Network

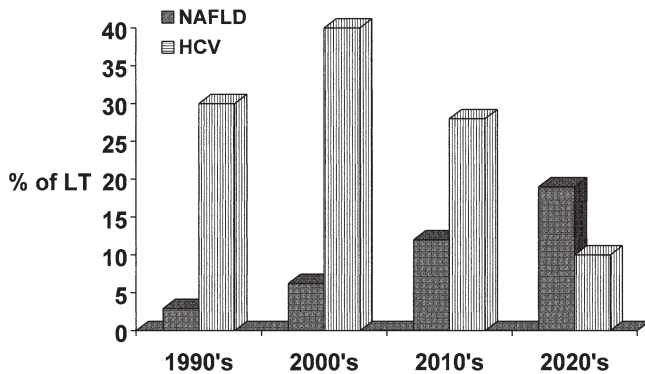


**Figure 1.** Centers of Disease Control estimates of incidence of HCV infection and obesity in the United States, 1982–2000. See <http://www.cdc.gov/>

for Organ Sharing database has not recorded the frequency of NAFLD/NASH as an indication for liver transplantation until recently, and data capture has been sporadic. Similarly the Liver Transplant Database of the National Institute of Diabetes and Digestive and Kidney Diseases did not list NASH or fatty liver disease as a specific indication or primary cause of liver disease. There are thus no national figures to indicate the frequency of liver disease associated with NASH as an indication for liver transplantation. We have previously reported that, during a 6-year period, 2.7% of liver transplants at our institution were carried out for decompensated cirrhosis as a result of NASH. In the 5 years since our initial report (between January 1, 1998, and December 30, 2003) 28 of 439 patients (6.8%) who have undergone liver transplantation at our center had histologic and clinical evidence of NASH as the primary cause of liver disease. The frequency of NASH as an indication for liver transplantation, at our center at least, has thus more than doubled in 5 years.

Because our data are based on histologic examination of explanted livers (all explanted livers undergo histologic examination), an ascertainment bias is unlikely to have accounted for the observed increase in prevalence of NASH as an indication for liver transplantation. The introduction of the MELD (model for endstage liver disease) scoring system for allocation of donor livers has not favored patients with NASH and is thus also unlikely to have affected our findings.

What, if anything, do the national increase in prevalence of obesity and our single center experience of NASH as an indication for liver transplant portend for the medical community as a whole? Data from the Centers for Disease Control provide worrying clues. Figure 1 shows Centers for Disease Control data regarding changes in the frequency of



**Figure 2.** Projected relative frequencies of NASH and HCV as indications for liver transplantation (LT).

new HCV infections and the prevalence of obesity between 1982 and 2000. Between January 1, 1998, and December 30, 2003, there were a total of 22,676 liver transplants carried out in adults in the United States according to the United Network for Organ Sharing ([www.unos.org](http://www.unos.org).data). If we extrapolate our experience nationally, the number of people undergoing liver transplantation for NASH is on the order of 1.0 per 1 million US residents/year (based on 6.4% of 22,676 adult liver transplants being carried out for NASH and assuming a mean US population in 1998–2003 of 280 million—<http://www.census.gov/population/www/projections>). On the basis of the known increases in the prevalence of obesity in the United States<sup>110</sup> (Figure 1), the frequency of liver transplantation for NASH will increase to 2.2–4.0 cases/1 million US residents/year in 10–15 years. The higher estimate reflects known increases in severity of NAFLD with degree of obesity. Steatohepatitis is found in 3% of lean, ~20% of obese, and almost half of morbidly obese people.<sup>10,12</sup> Of severely obese patients with diabetes, 100% have at least mild steatosis, one half have steatohepatitis, and ~20% have cirrhosis.<sup>111</sup>

Figure 2 shows the potential impact of these changing demographics on the relative frequency of NASH as an indication for liver transplantation. Without a safe, effective, and widely prescribed therapy for NAFLD and NASH, somewhere between 2015 and 2030, liver failure as a result of NASH will overtake HCV as the most common indication for liver transplantation in the United States. Unfortunately, in contrast to the incidence of HCV, which has decreased by more than 80% from its peak, increases in the incidence and severity of obesity and NAFLD only show signs of accelerating. Because NASH recurs frequently after liver transplantation and can result in graft loss,<sup>112,113</sup> the burden on an already stretched organ supply will be substantial. NAFLD looks set to increasingly dominate the practice of hepatology, particularly in the arena of liver transplantation.

Many of the numbers used in these projections can be debated. The wave of liver failure as a result of NASH that has washed up on the shore of our center is, however, likely to be the leading edge of a gathering storm that will soon affect all of us. We should use the time we have before the impact is fully felt to optimize management of NASH before and after liver transplantation. The current National Institutes of Health sponsored clinical trial network is a good start but is not enough. If the adipogenic and possible hepatotoxic qualities of pioglitazone and other PPAR- $\gamma$  agonists negate their beneficial effects, we will be alarmingly short of treatment options for NASH.<sup>105</sup> Multicenter studies aimed at identifying patients at risk for and defining the mechanism of progressive liver injury as well as studies aimed at optimizing the management of NASH and insulin resistance after liver transplantation are needed. The epidemic of hepatitis C infection and its impact on liver transplantation were a surprise. Liver failure as a result of NASH is a storm we can see a mile off.

## Summary

NAFLD, already the most common form of liver disease in the United States, can be expected to increase in prevalence and severity in parallel with national epidemics of obesity and type 2 diabetes. NAFLD is frequently associated with insulin resistance. Although insulin resistance and, thereby, hyperinsulinemia are, in large part, metabolic consequences of obesity, the basis of diversity in severity and progression of inflammation and fibrosis is not known. Increased susceptibility to oxidative stress is likely to play a role. Several patient characteristics have been associated with more severe histologic findings in patients with NAFLD, including type 2 diabetes, hypertension, age older than 40 years, and higher transaminase levels. Liver biopsy is, however, required to accurately grade and stage NAFLD histologically. Although the natural history of NAFLD is relatively poorly defined, NAFLD is increasingly recognized as an important cause of decompensated liver disease. Weight reduction and improved insulin sensitivity are associated with improved biochemical and histologic parameters of NAFLD. There are, however, no proven safe and efficacious pharmacologic treatments for NAFLD.

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