

# Treatment of nonalcoholic fatty liver disease

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## Abstract

**Treatment of patients with nonalcoholic fatty liver disease (NAFLD) has typically been focused on the management of associated conditions such as obesity, diabetes mellitus and hyperlipidemia. NAFLD associated with obesity may resolve with weight reduction, although the benefits of weight loss have been inconsistent. Appropriate control of glucose and lipid levels is always recommended, but not always effective in reversing the liver condition. Results of pilot studies evaluating ursodeoxycholic acid, gemfibrozil, betaine, N-acetylcysteine, vitamin E ( $\alpha$ -tocopherol), metformin and thiazolidinedione derivatives suggest that these medications may be of potential benefit for patients with NAFLD. These medications, however, need first to be tested in well-controlled trials with clinically relevant end-points and extended follow up. A better understanding of the pathogenesis and natural history of NAFLD will help to identify the subset of patients at risk of progressing to advanced liver disease, and hence, those patients who should derive the most benefit from medical therapy.**

**Key words:** Steatohepatitis, Diabetes, Obesity, Steatosis, Ursodeoxycholic acid.

## Introduction

Nonalcoholic fatty liver disease (NAFLD) is a medical condition that may progress to end-stage liver disease.<sup>1</sup> The spectrum of NAFLD is wide and ranges from simple fat accumulation in hepatocytes (steatosis) without biochemical or histological evidence of inflammation or fibrosis, to fat accumulation plus necroinflammatory activity with or without fibrosis (steatohepatitis), to the development of advanced liver fibrosis or cirrhosis (cirrhotic stage).<sup>2</sup> All these stages are histologically indistinguishable from those produced by excessive alcohol consumption, but occur in patients who do not abuse alcohol. Nonalcoholic steatohepatitis

(NASH) represents only a stage within the spectrum of NAFLD. The clinical implications of NAFLD and NASH are mostly derived from its common occurrence in the general population as well as its progressive potential.

NAFLD is probably the most common liver disease in many countries affecting 10% to 24% of the general population.<sup>3</sup> There is a direct correlation between body mass index (BMI) and prevalence and severity of NAFLD. The prevalence of NAFLD increases by 4.6-fold in obese people. About two to three fourths of obese (BMI = 30 kg/m<sup>2</sup>) individuals have NAFLD whereas more than 90% of severely obese (BMI > 35 kg/m<sup>2</sup>) people have NAFLD. NAFLD affects 2.6% of children and this figure increases up to 53% in the obese child population. NAFLD is the cause of asymptomatic elevation of aminotransferases in 42% to 90% of cases once other causes of liver disease are excluded and represents a common explanation for abnormal liver tests in blood donors. Regardless of BMI, type 2 (non-insulin dependent) diabetes mellitus significantly increases the prevalence and severity of NAFLD. About a half (range 21% to 78%) of patients with type 2 diabetes mellitus have NAFLD.<sup>3</sup>

NAFLD and NASH should be differentiated from steatosis with or without hepatitis resulting from well-known, secondary causes of fatty liver (*Table 1*) because they have distinctly different pathogeneses and outcomes. The terms “NAFLD” and “NASH” should be reserved for those patients in whom none of the causes of fatty liver disease listed in *Table 1* are causing the liver condition. Other liver diseases that may present with a component of steatosis such as viral or autoimmune hepatitis and metabolic/hereditary liver diseases should be appropriately excluded.

Although the natural history of NAFLD and its different stages remain unknown, it is clear that some patients, particularly those with simple steatosis follow a relatively benign course. Simple steatosis usually remains stable for many years, and will probably never progress in many patients.<sup>4-9</sup> Most patients who develop problems from NAFLD have steatohepatitis or NASH, at least as we currently understand this condition. Hence, the decision to intervene with medical therapy should be aimed at arresting disease progression, and ideally, be restricted to those patients (i.e., NASH patients) at risk of developing advanced liver disease.

This paper reviews 1) existing medical therapy for patients with NAFLD and NASH; 2) the emerging data from clinical trials evaluating potentially useful medications; and 3) the potential therapeutic implications of recent studies on the pathogenesis of this liver disease.

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## Treatment of associated conditions

### Underlying metabolic conditions

A large body of clinical and epidemiological data gathered during the last three decades indicates that obesity, type 2 diabetes mellitus and hyperlipidemia are major associated conditions or predisposing factors leading to the development of NASH.<sup>1,3-11</sup> Hence, it is reasonable to believe that the prevention or appropriate management of these conditions would lead to improvement or arrest of the liver disease.

Steatosis and steatohepatitis may resolve with weight reduction, although the benefits of weight loss have been inconsistent. An early report describes two patients whose biopsy showed steatosis, necroinflammation and fibrosis that significantly improved following 11 and 20 kg weight loss over 1 year.<sup>12</sup> In another report,<sup>13</sup> five obese patients stopped eating for some time and lost 14-30 kg within 1 month. Serum levels of liver enzymes appeared to be unaffected by starvation. The hepatic fat content decreased in three of them, but fibrosis became more prominent in four of the five patients.<sup>13</sup> In another series,<sup>14</sup> ten obese patients who were treated with prolonged fasting for a mean of 71 days and lost a mean of 41 kg had a marked reduction in fatty infiltration, but areas of focal necrosis were more numerous and some patients developed bile stasis as well. Similar effects were noted in seven obese subjects after treatment with a diet of 500 calories per day and who experienced a mean weight reduction of 60 kg during a mean period of five months. In this same series, 14 patients maintained a mean weight loss of almost 65 kg for 1.5 years and in nine patients the liver biopsy findings normalized with only rare areas of focal necrosis in the remaining five patients.<sup>14</sup>

Another case-series of 39 obese patients reported a marked biochemical improvement particularly in those patients who lost more than 10% of body weight.<sup>15</sup> Liver biopsies were not performed in any of these patients. In another series,<sup>16</sup> 41 morbidly obese patients with different stages of NAFLD had a median weight loss of 34 kg during treatment with a very-low calorie formula diet (388 kcal per day). The degree of fat infiltration improved significantly. However, a fifth of patients, particularly those patients with more pronounced reduction of fatty changes and a faster weight loss, developed slight portal inflammation or fibrosis. None of the patients losing less than 230 g per day or approximately 1.6 kg per week developed fibrosis. A significant improvement in liver biochemistries was noted regardless of the histological changes. In a more recent study,<sup>17</sup> liver biochemistries and the degree of fatty infiltration improved significantly in 15 obese patients with different stages of NAFLD who were treated with a restricted diet (25 cal/kg/d) plus exercise for 3 months. Improvement in the degree of inflammation and fibrosis also occurred in some patients.

Information regarding the effect of weight loss in obese children with NAFLD is sparse. In a case-series report,<sup>18</sup> 7 out of 9 obese children with NAFLD who adhered to treatment with hypocaloric diet and exercise lost about 500 g per week. Weight loss led to improvement in serum aminotransferase levels and degree of hepatic steatosis evaluated by ultrasonography. Post-treatment liver histology normalized in the only child who underwent liver biopsy. In a more recent series,<sup>19</sup> 33 obese children with abnormal liver tests due to NAFLD underwent 6 months of treatment with a moderately hypocaloric diet (mean 35 cal/kg/d; carbohydrates 65%, protein 12%, fat 23%) plus aerobic exercise (= 6 hours/week) to obtain a weight loss of about 500 g per week. Liver tests became normal in all

**Table I.** Causes of fatty liver disease.

Nutritional	Drugs*	Metabolic/Genetic	Other
Protein-calorie malnutrition	Glucocorticoids	Lipodystrophy	Inflammatory bowel disease
Starvation	Synthetic estrogens	Dysbetalipoproteinemia	Small bowel diverticulosis with bacterial overgrowth
Total parenteral nutrition	Aspirin	Weber-Christian disease	HIV infection
Rapid weight loss	Calcium channel blockers	Wolman's disease	Environmental hepatotoxins
Gastrointestinal surgery for obesity	Amiodarone	Cholesterol ester storage	Phosphorus poisoning
	Tamoxifen	Acute fatty liver of pregnancy	Petrochemical exposure
	Tetracycline		Toxic mushrooms
	Methotrexate		Organic solvents
	Perhexiline maleate		<i>Bacillus cereus</i> toxins
	Valproic acid		
	Cocaine		
	Antiviral agents		
	Zidovudine,		
	Didanosine,		
	Fialuridine		

\* Partial list of agents that produce fatty liver in humans. Some drugs produce inflammation as well.

children who lost weight whereas the degree of steatosis evaluated by ultrasonography improved significantly or normalized in all children who lost  $\geq 10\%$  of body weight. In another report,<sup>20</sup> six obese children with NAFLD had improvement in serum aminotransferases with weight loss after a mean follow-up of 18 months.

Based on the analysis of these studies,<sup>12-20</sup> it is clear that weight loss, particularly if gradual may lead to improvement in liver histology. However, rate and degree of weight loss required for normalization of liver histology have not been established. It seems that the means by which, or how fast, weight loss is achieved is important and may play a critical role in determining whether more severe liver damage will develop. In patients with a high degree of fatty infiltration, pronounced reduction of fatty changes and fast weight loss may promote portal inflammation and fibrosis. Similarly, starvation or total fasting may lead to development of pericellular and portal fibrosis, bile stasis and focal necrosis.<sup>13,16</sup> This paradoxical effect seen in some patients may be due to increased circulating free fatty acid levels derived from fat mobilization and thus, a greater rate of exposure of the liver to an unusual high amount of fatty acids. Furthermore, liver tests, in particular serum aminotransferase levels, almost always improve or normalize with weight loss and they are poor predictors of worsening of liver histology due to weight loss.

Different caloric restrictions have been evaluated. A formula providing 600-800 cal per day with 45-100 g of high-quality animal proteins, less than 100 g carbohydrate and less than 10 g fat in adults can be tried, particularly for those patients who are  $\geq 30\%$  overweight.<sup>21</sup> This low-calorie diet has been recommended by some authors only after a more conventional 1200 calorie diet has been tried and proven unsuccessful. Diet to produce weight loss should always be prescribed on an individual basis and considering the patient's overall health. Patients who have obesity-related disease such as diabetes mellitus, hyperlipidemia or cardiovascular disease will require close medical supervision during weight loss to adjust the medication dosage as needed. Further studies are necessary to determine the most appropriate content of the formula to be recommended for obese patients with NAFLD and NASH.

Medications used to reduce appetite will result in weight reduction in many patients.<sup>22</sup> These medications are associated with rare, but potentially serious side effects, including pulmonary hypertension.<sup>23</sup> It remains to be proven whether the risk-to-benefit ratio of these (or other) medications justifies their use in patients with NAFLD or NASH.

Obese patients with type II diabetes mellitus should be enrolled in a low-calorie diet and exercise program. In patients with diabetes and hyperlipidemia, good laboratory control is always recommended, but not always effective in reversing the liver disease. In obese ob/ob mice, an animal model of steatosis,<sup>24</sup> metformin, an oral hypoglycemic medication led to improvement in liver tests and degree of steatosis. Based on these findings, metformin and other in-

sulin-sensitizing medications are being evaluated in humans with NAFLD and NASH (see pharmacological therapy).

Gastric and intestinal bypass, popular weight-reducing surgical procedures in the 1960's and 1970's, have been almost abandoned, mainly because of the high frequency of severe NAFLD followed by liver failure, particularly in patients undergoing jejunioileal bypass.<sup>25,26</sup> The development of NAFLD in obese patients undergoing intestinal or gastric bypass may be due to a combination of additive factors including protein/calorie malnutrition, increased fluxes and liver exposure to free fatty acids, and bacterial overgrowth in the defunctionalized intestinal segment. In this regard, enteral and parenteral supplementation of amino acids and proteins may be of benefit.<sup>27</sup> Also, in a series of 33 obese patients undergoing intestinal bypass,<sup>28</sup> metronidazole given at random intervals after surgery led to a significant improvement or normalization in the degree of steatosis.

Patients receiving long-term total parenteral nutrition may develop NAFLD partially because of choline deficiency. Choline supplementation has been reported to improve or revert hepatic steatosis.<sup>29,30</sup> Similarly, bacterial overgrowth in the resting intestine along with the lack of enteral stimulation has been implicated in the genesis of liver damage, including NAFLD, in patients on long-term total parenteral nutrition. Polymyxin B, a nonabsorbable antibiotic that specifically binds to the lipid A-core region of lipopolysaccharide<sup>31</sup> and metronidazole,<sup>32</sup> have been shown to significantly improve the degree of fatty infiltration and reduce the production of tumor necrosis factor in rats receiving total parenteral nutrition.

### Drugs and hepatotoxins

Several drugs and environmental exposure to some hepatotoxins (*Table I*) have been recognized as potential causes of fatty liver, steatosis, steatohepatitis and even cirrhosis. Although the liver condition resulting from these secondary causes differs strikingly from NAFLD in pathogenesis and outcomes, they should always be sought in patients with NAFLD since their withdrawal, when possible, usually leads to resolution of the liver disease.

### Promising pharmacological therapy

Based on the fact that rapid weight loss may worsen NAFLD, use of medications that can directly reduce the severity of liver damage independent of weight loss is a reasonable alternative. However, pharmacological therapy directed specifically at the liver disease has only recently been evaluated in patients with NAFLD or NASH. Most of these studies have been uncontrolled, open-label and lasting one year or less and only few of them have evaluated the effect of treatment on liver histology (*Table II*).

The decision to intervene with pharmacological therapy aimed at the underlying liver disease is based on the

anticipated risk of progression to severe liver disease. Pharmacological therapy may be of particular benefit for those patients who do not lose weight or cannot maintain long-term weight reduction as usually happens in most obese patients. Pharmacological therapy may also benefit those patients who lack risk factors or associated conditions such as non-obese, non-diabetic patients and those with a normal lipid profile.

Clofibrate is a lipid-lowering drug that decreases the content of hepatic triglyceride in rats with ethanol-induced hepatic steatosis. Based on this, a pilot study was performed to evaluate the usefulness of clofibrate (2 g a day) in the treatment of patients with NASH.<sup>33</sup> After a year of treatment, no significant changes in liver tests or degree of steatosis, inflammation or fibrosis was found.

In a recent report,<sup>34</sup> 46 patients with NASH were randomized to treatment with gemfibrozil 600 mg per day for 4 weeks or no treatment. A significant improvement in aminotransferase levels was noted with gemfibrozil compared to baseline values, which did not occur in the untreated patients.

Ursodeoxycholic acid (UDCA) is the epimer of chenodeoxycholic acid and appears to replace endogenous bile acids, some of which may be hepatotoxic, with the non-hepatotoxic ursodeoxycholic acid. UDCA has also membrane stabilizing or cytoprotective effects as well as antiapoptotic actions. Four, open-label, pilot studies have evaluated the therapeutic benefit of UDCA in patients with NASH. In one of these studies, 24 patients,<sup>33</sup> received UDCA at a dose of 13-15 mg/kg/d for 12 months which led to a significant improvement in liver tests and the degree of steatosis compared to baseline. In another study,<sup>35</sup> liver tests normalized or significantly improved after 6 months of treatment with UDCA (10/mg/kg/d) in 13 patients with NASH. In another study,<sup>36</sup> 31 patients with

NASH were randomized to UDCA (10 mg/kg/d) plus low-fat diet or low-fat diet alone for 6 months. Normalization of liver tests was significantly more common among patients treated with UDCA plus diet than diet alone. In the latest study,<sup>37</sup> UDCA (250 mg three times a day) given for 6-12 months improved aminotransferase levels in 24 patients with NASH; UDCA therapy also improved several serum markers of fibrogenesis. Based on these promising results, a large scale, placebo-controlled trial of UDCA in NASH patients is now underway in the United States.

Betaine, a normal component of the metabolic cycle of methionine increases S-adenosylmethionine levels which in turn protect the liver from ethanol-induced triglyceride deposition in rats.<sup>38</sup> In a recent study, betaine 20 g day was given to 8 patients with NASH. After a year of treatment, a significant improvement in serum aminotransferase levels was noted whereas the degree of steatosis, necroinflammatory activity and fibrosis improved or remained unchanged in all patients.<sup>39</sup>

N-acetylcysteine is a glutathione prodrug that increases glutathione levels in hepatocytes, which in turn constrains hepatocyte production of reactive oxygen species, and hence, protects against oxidative stress in the liver. Recently,<sup>40</sup> 11 patients with NASH were treated with N-acetylcysteine (1 g a day) for 3 months. A significant improvement in aminotransferase levels occurred at the end of treatment.

Vitamin E ( $\alpha$ -tocopherol), a potent antioxidant particularly effective against membrane lipid peroxidation, suppresses TNF- $\alpha$ , IL-1, IL-6 and IL-8 expression by monocytes and/or Kupffer cells and inhibits liver collagen  $\alpha$ 1(I) gene expression.<sup>41</sup> A recent case-series study reports the results of treatment with vitamin E in 11 children with NAFLD. Vitamin E 400-1200 IU orally per day was given for 4 to 10 months and led to a significant improvement in liver tests.<sup>42</sup> In another study,<sup>43</sup>  $\alpha$ -tocopherol at a dose of

**Table II.** Drugs evaluated in the treatment of nonalcoholic fatty liver disease.

Author	Drug	No. of patients	Type of study	Compared with	Duration of treatment	Aminotrasferases	Histology
Laurin et al. (1996) <sup>33</sup>	UDCA	24	Open-label	Baseline	12 months	Improved	Improved
Laurin et al. (1996) <sup>33</sup>	Clofibrate	16	Open-label	Baseline	12 months	No improvement	No improvement
Guma et al. (1997) <sup>35</sup>	UDCA + diet	24	Open-label Randomized	Baseline Diet alone	6 months	Improved†	ND
Ceriani et al. (1998) <sup>36</sup>	UDCA + diet	31	Open-label	Baseline Diet alone	6 months	Improved†	ND
Holoman et al. (2000) <sup>37</sup>	UDCA	24	Open-label	Baseline	6-12 months	Improved	ND
Basaranoglu et al. (1999) <sup>34</sup>	Gemfibrozil	46	Open-label Randomized	No treatment Baseline	1 month	Improved	ND
Abdelmalek et al. (2000) <sup>39</sup>	Betaine	8	Open-label	Baseline	12 months	Improved	Improved
Gulbahar et al. (2000) <sup>40</sup>	N-Acetylcysteine	11	Open-label	Baseline	3 months	Improved	ND
Lavine (2000) <sup>42</sup>	Vitamin E	11*	Open-label	Baseline	4-10 months	Improved	ND
Caldwell et al. (2001) <sup>46</sup>	Troglitazone	10	Open-label	Baseline	3-6 months	Improved	Improved
Hasegawa et al (2001) <sup>43</sup>	Vitamin E	22	Open-label	Baseline Diet	12 months	Improved	Improved†
Marchesini et al (2001) <sup>48</sup>	Metformin	14	Open-label	Baseline	4 months	Improved	ND

UDCA, ursodeoxycholic acid; \* Study performed in children; † Liver biopsy performed in 9 patients post-treatment; ND = not done.



300 mg/d was given for 1 year to 12 patients with liver biopsy-proven NASH and 10 non-biopsied patients with NAFLD. Liver tests improved significantly compared to baseline whereas the degree of steatosis, inflammation and fibrosis improved or remained unchanged in the nine patients with NASH who had post-treatment liver biopsy performed. Plasma levels of transforming growth factor- $\beta$ 1 in patients with NASH were reduced significantly with  $\alpha$ -tocopherol.<sup>43</sup>

Type 2 diabetes mellitus and truncal obesity are well-known conditions associated with resistance to normal peripheral actions of insulin. Indeed, insulin resistance represents the most reproducible predisposing factor for NAFLD and NASH.<sup>44</sup> Hence, it is reasonable to speculate that the use of medications that improve insulin sensitivity may benefit the liver disease of patients with associated insulin-resistance conditions. Thiazolidinediones are a new class of antidiabetic drugs that selectively enhance or partially mimic certain actions of insulin, causing an anti-hyperglycemic effect frequently accompanied by a reduction in circulating concentrations of insulin, triglycerides and nonsterified fatty acids.<sup>45</sup> Troglitazone, a thiazolidinedione derivative, was given to 10 patients with NASH for 3-6 months.<sup>46</sup> ALT levels normalized in 7 patients and although features of NASH remained in the post-treatment liver biopsy, the grade of necroinflammation improved in 4/7 patients. Troglitazone may produce hepatocellular liver damage<sup>47</sup> and is now no longer available in the United States and Europe. Other safer thiazolidinedione agents currently available may warrant further evaluation in patients with NASH.

Metformin is an antidiabetic medication that improves insulin resistance. In the ob/ob mice, an animal model of fatty liver, metformin reversed hepatomegaly as well as steatosis and aminotransferases abnormalities possibly through inhibiting hepatic expression of TNF- $\alpha$ .<sup>24</sup> Based on these results, metformin at a dose of 500 mg three times a day was given for 4 months to 14 patients with NASH.<sup>48</sup> Metformin therapy was associated with a significant improvement in liver tests and glucose disposal - as an index of insulin sensitivity, as well as a significant decrease in hepatic volume and body weight. Unfortunately post-treatment liver biopsies were not performed in any case. Metformin can produce lactic acidosis in patients with liver disease; thus, caution should be exercised when using this medication in patients with NAFLD or NASH.

The encouraging results of pilot studies with gemfibrozil, ursodeoxycholic acid, betaine, N-acetylcysteine,  $\alpha$ -tocopherol, and insulin sensitizing drugs (metformin, thiazolidinedione) in the treatment of NAFLD and NASH (Table II) suggest that these agents deserve further evaluation in clinical trials.

However, in order to make solid recommendations of routine administration of any of these (or other) medications in the treatment of patients with NAFLD or NASH, further, well-controlled clinical trials, are clearly necessary.

These studies must have sufficient power, adequate duration of follow up, be analyzed on an intention-to-treat basis and must also include clinically relevant end-points. The simple improvement or normalization of liver tests and/or the degree of steatosis on imaging studies such as ultrasonography or CT scan in most of the pilot studies reported to date (Table II), do not necessarily imply that these agents will have a real effect on the natural history of this liver disease. Improvement of liver histology may be a more accurate surrogate marker of a better long-term prognosis than liver test results or imaging studies. A beneficial medication for NAFLD or NASH patients should be safe and well tolerated and ideally prove beneficial in improving quality of life, delaying the development of liver-related complications and improving long-term survival.

Given the slowly progressive nature of NAFLD and NASH, hundreds of patients with this condition would need to be enrolled in prospective, clinical trials and be followed-up for a number of years, perhaps decades, in order to see a real effect of a medication on long-term survival. It may be unrealistic to believe that such a study is feasible and that will be appropriately funded. The identification of patients with NAFLD or NASH on risk of progressing to end-stage liver disease may lead to enroll in therapeutic trials those "high-risk" patients who, in theory, are expected to derive the most benefit from medical therapy.

## Future directions

In order to develop effective medical therapy for patients with NAFLD or NASH, further work is clearly needed to enhance our understanding of the pathogenesis and natural history of this condition. Some lines of evidence, albeit still inconclusive and some derived from studies in animal models of fatty liver suggest that oxidative stress/lipid peroxidation, bacterial toxins, overproduction of TNF- $\alpha$ , alteration of hepatocyte ATP stores and Cyp2E1/Cyp4A enzyme activity may play a role in the genesis and progression of NAFLD and NASH.

Acute or chronic hepatic steatosis regardless of the cause is associated with lipid peroxidation which seemed to increase with the severity of steatosis.<sup>49</sup> Malondialdehyde, an end-product of lipid peroxidation, activates hepatic stellate cells, stimulating collagen production and fibrogenesis. Malondialdehyde may also contribute to inflammation by activating NF- $\kappa$ B which regulates the expression of proinflammatory cytokines such as TNF- $\alpha$  and interleukin-8.<sup>50</sup> Another end-product of lipid peroxidation, 4-hydroxynonenal, is a strong chemoattractant for neutrophils. Furthermore, the risk factors for development of NAFLD, namely obesity with rapid weight loss and type 2 diabetes mellitus, lead to increase circulating levels of free fatty acids with consequent enhanced concentration in the liver. Free fatty acids *per se* are potentially cytotoxic<sup>51</sup> and may also increase cytochrome P450 Cyp2E1/Cyp4A activity as shown in a rat model of NAFLD using a

diet deficient in methionine-choline and in humans with NASH.<sup>52-54</sup>

This evidence suggests that oxidative stress and lipid peroxidation may, in part, be one of the critical factors involved in the genesis and probably in the progression of NAFLD and NASH. Thus, if this concept is valid, further therapeutic strategies may be directed to its inhibition. In support of the role of oxidative stress and lipid peroxidation in the pathogenesis of NAFLD and NASH is the demonstration that  $\alpha$ -tocopherol, a potent antioxidant improved liver tests in children<sup>42</sup> and adults<sup>43</sup> with NAFLD and NASH. Both  $\alpha$ -tocopherol and silymarin, a flavanoid isolated from milk thistle with antioxidant, antifibrotic and membrane-stabilizing effects<sup>55</sup> deserve to be evaluated in larger controlled trials of patients with NASH.

Based on the fact that metronidazole and polymixin B may prevent the development of NAFLD in obese patients undergoing intestinal bypass<sup>28</sup> and in rats receiving total parenteral nutrition,<sup>31,32</sup> a role of endotoxin/cytokine mediated injury has been suggested as a contributing factor for the development of NAFLD. More recently, it has been shown that genetically obese mice are very sensitive to the effect of lipopolysaccharide in developing NAFLD.<sup>56</sup> The mRNA of interferon gamma, which sensitizes hepatocytes to TNF- $\alpha$  toxicity, was over-expressed whereas mRNA of IL-10, which is inhibitory to TNF- $\alpha$  effects was reduced. In this model, the phagocytic activity of Kupffer cells was reduced, presumably favoring the development of systemic endotoxemia and release of proinflammatory cytokines. If this concept is valid, the potential benefit of intestinal decontamination, administration of soluble cytokine receptors and neutralizing anti-cytokine antibodies as well as drugs with anti TNF- $\alpha$  activity (i.e., pentoxifylline, anti-TNF- $\alpha$  antibodies) may warrant further evaluation as therapies for patients with NAFLD and NASH.

ATP stores in hepatocytes of mice<sup>57</sup> and humans<sup>58</sup> with steatosis seem to be potentially vulnerable to depletion compared to non-obese controls. Hence, treatment efforts primarily directed toward protecting hepatocyte ATP stores might be potentially beneficial in NAFL patients. Similarly, Cyp2E1/Cyp4A activity may contribute to hepatotoxicity in mice and humans with NAFL.<sup>52-54</sup> Treatment strategies to limit its activity such as dietary modifications (fat-reduced diet) may be beneficial.

The role of iron in the pathogenesis of NAFLD remains uncertain. Anecdotal reports suggested that increased iron stores might lead to more severe liver disease in NASH patients.<sup>59,60</sup> Studies with larger patient populations of NASH, however, have shown that this is not always the case.<sup>10,61</sup> Nevertheless, serum iron studies should always be performed in patients with NAFLD and NASH, and an iron stain with quantification of hepatic iron is also recommended in those patients with abnormal serum iron studies. If excessive iron, and perhaps also the HFE gene mutation are found serial phlebotomies, which may improve liver biochemistries,<sup>62,63</sup> may be a therapeutic option.

## When patients with NAFLD or NASH should be treated?

The decision about whether to treat an individual patient with NAFLD or NASH should be primarily dictated by knowing the potential risk of progressing to end-stage liver disease. However, since no prospective, longitudinal clinical studies have been performed, the clinical course of this condition remains unknown, and hence, treatment recommendations remain speculative.

An attempt at gradual weight loss is a useful first step in the management of patients with simple uncomplicated steatosis as well as making a concerted effort to maintain appropriate control of serum glucose and lipids levels. Perhaps this along with appropriate exclusion of other liver disease may be the only treatment recommendation for those patients with pure steatosis and no evidence of inflammation or fibrosis who seem to have the best prognosis within the spectrum of NAFLD. NAFLD patients with steatohepatitis, particularly those with fibrosis on liver biopsy may have a worse prognosis and they should be monitored closely, make a greater effort for adequate metabolic control and be offered enrollment in well-controlled clinical trials evaluating the potential benefit of promising medications. For those patients with cirrhotic stage NAFLD and decompensated disease, liver transplantation is a potential life-extending therapeutic alternative. However, NAFLD may recur in the allograft<sup>64</sup> or develop after liver transplantation for cryptogenic cirrhosis.<sup>65</sup>

## Conclusions

NAFLD and its different stages affect a high proportion of the world population. Insulin resistance and oxidative stress play a critical role in the pathogenesis of NAFLD. Liver biopsy remains the most sensitive and specific means of providing important prognostic information. Simple steatosis may have the best prognosis within the spectrum of NAFLD, but it has the potential to progress to NASH, fibrosis and even cirrhosis. No effective medical therapy is currently available for all patients with NAFLD or NASH. A useful first step in the treatment of patients with NAFLD and NASH is the management of associated conditions such as obesity, diabetes mellitus and hyperlipidemia. Gradual weight loss may improve the liver condition, but total starvation or very low calorie diets should be avoided. Appropriate control of glucose and lipid levels should always be sought. Pharmacological therapy aimed at the underlying liver disease holds promise. Liver transplantation is a therapeutic alternative for some patients with decompensated, end-stage liver disease, but NAFLD may recur, or develop after liver transplantation.

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