Diagnostic and Treatment Implications of Nonalcoholic Fatty Liver Disease and Nonalcoholic Steatohepatitis

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Abstract: Nonalcoholic fatty liver disease (NAFLD) affects 75 to 100 million adults in the United States and is the leading cause of chronic liver disease worldwide, fueled by the rising epidemic of obesity and metabolic syndrome. NAFLD is the hepatic manifestation of metabolic syndrome; thus, accurately assessing and managing comorbid metabolic syndrome components is paramount. Nonalcoholic steatohepatitis (NASH) is a subset of NAFLD that includes a more progressive and advanced form of the disease, with a greater risk of fibrosis progression. Correctly diagnosing and staging NAFLD and distinguishing the subset of NASH patients is not only critical for disease monitoring and prognostication, but also holds potential implications for therapies. Although the current therapeutic landscape for NAFLD does not offer many options, future therapies are on the horizon. Properly staging the severity of disease and fibrosis is especially important when considering the eligibility and cost-effectiveness of these therapies.

Nonalcoholic fatty liver disease (NAFLD) is the leading cause of chronic liver disease worldwide, with 75 to 100 million adults affected in the United States alone. NAFLD is the hepatic manifestation of metabolic syndrome, and although the exact pathogenesis of NAFLD is not well understood, there are likely multifactorial pathways that involve insulin resistance, oxidative injury, hepatic iron deposition, gastrointestinal hormone crosstalk, gastrointestinal bacteria, and genetic predisposition.[1] NAFLD is a general term that encompasses 2 subsets of patients: individuals with nonalcoholic fatty liver (NAFL), which is defined by the presence of at least 5% hepatic steatosis without evidence of hepatocellular injury, and individuals with nonalcoholic steatohepatitis (NASH), which is defined by the presence of at least 5% hepatic steatosis and inflammation with hepatocellular injury (eg, ballooning), with or without fibrosis. Although the natural history of NAFLD involves progression from NAFL to NASH, disease progression likely involves a continuum with intermediate stages rather than a clear, distinct line that separates NAFL from NASH. Furthermore, disease progression may not be linear and may take on

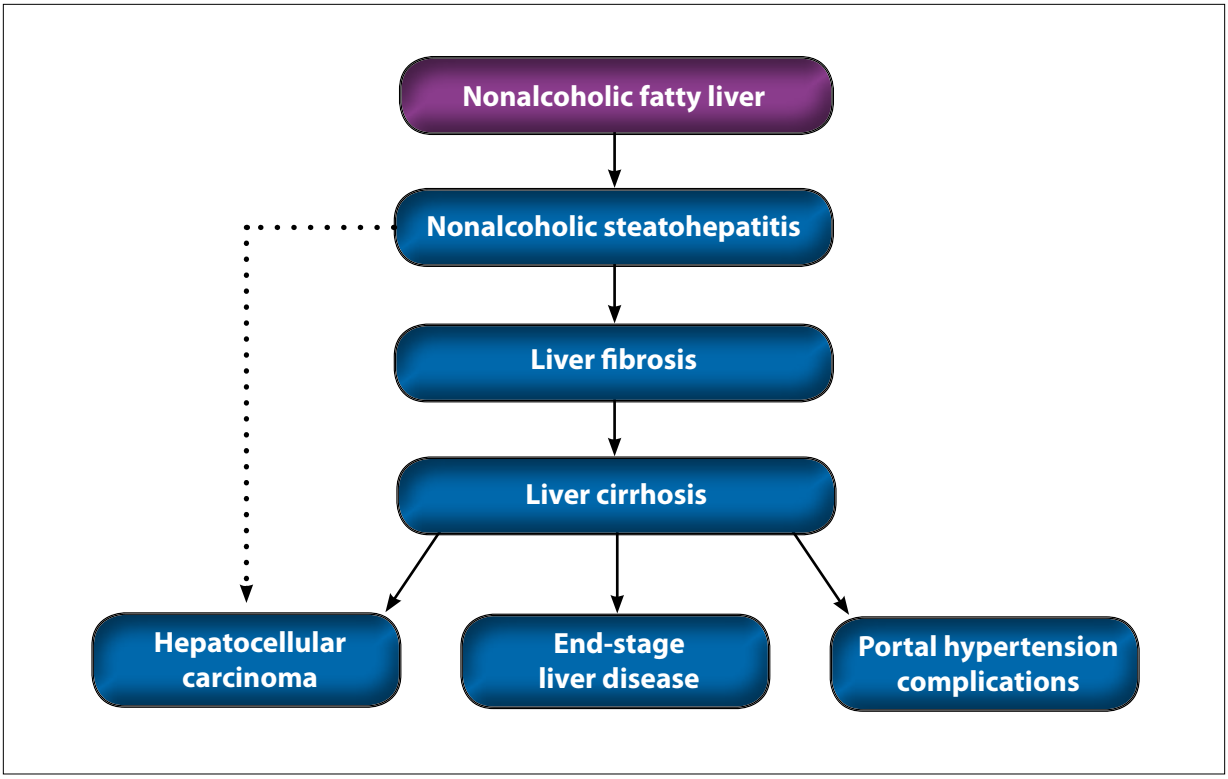

Figure. Cascade of disease progression among individuals with nonalcoholic fatty liver disease. The dotted line demonstrates the increasing evidence of hepatocellular carcinoma in noncirrhotic patients with nonalcoholic steatohepatitis.

a natural history with stages of progression and regression. Further disease progression among NASH patients involves development of fibrosis, cirrhosis, and cirrhosis-related complications such as hepatocellular carcinoma and end-stage liver disease (Figure).[2] Although accurately identifying NASH is important to guide disease monitoring, prognostication, and therapeutic considerations, no consistent biomarkers exist, and liver biopsy remains the gold standard for histologic diagnosis. This article discusses the distinguishing features of NAFL vs NASH, the diagnostic tools by which clinicians can accurately categorize these distinct subsets of disease, and potential implications that accurate staging may have on the need for NAFLD therapies on the horizon.

Epidemiology

The worldwide prevalence of NAFLD continues to rise, with an estimated 25% to 45% of US adults affected.[3] Current estimates suggest that approximately 68% of all US adults meet body mass index criteria for being overweight or obese.[4] However, many of these estimates are derived from survey- or cohort-based studies, the majority of which are biased due to underrepresentation of ethnic minorities or misclassification biases. Furthermore, it is broadly recognized that NAFLD awareness among both patients and providers is low, and, thus, existing prevalence studies likely underestimate the true burden of this disease. Nevertheless, it is important to note that trends in NAFLD prevalence parallel the rising prevalence of obesity and metabolic syndrome in the United States, with recent research demonstrating that metabolic syndrome affects nearly 35% of all US adults and 50% of individuals aged 60 years or older.[5] Given the lack of sensitive or specific biomarkers for NASH, the diagnosis relies primarily on histologic data. However, the paucity of such data at the population level makes estimating the prevalence of NASH among US adults challenging. The understanding of NAFLD progression is such that a subset of patients who have NAFL will develop NASH, among which 20% will develop fibrosis and progress to cirrhosis.[6] Because performing liver biopsies on such a large patient population is neither feasible nor pragmatic, the evolving paradigm of noninvasive tools for diagnosis and staging in order to guide future therapies will be especially important.

Diagnostic Tools

NAFL is often asymptomatic for decades prior to its transition to NASH, which can clinically manifest with nonspecific symptoms of vague right upper quadrant pain, fatigue, and malaise.[7] A physical examination does not offer clear pathognomonic findings that definitively diagnose NAFL or NASH, although 5% to 18% of NAFLD patients have evidence of hepatomegaly noted on physical examination.[8,9] Although NAFLD as a whole is generally characterized by disease progression, it is not entirely clear who actually progresses and at what rate disease progression occurs in all populations. Furthermore, disease progression may not be linear, and the concept of a seesaw pattern, with stages of disease progression and regression, has been used to describe the natural history of NAFLD. Early diagnosis of NASH is important, as the condition is associated with increased risks of disease progression compared with patients with NAFL only. Thus, NASH patients require a more comprehensive assessment of stage and severity of disease, and, in the near future, assessment of eligibility for NASH-specific therapies. As clinicians' understanding of NASH continues to evolve, further risk stratification with tiered-risk subcategories will help guide clinical management. Specifically, the noninvasive scoring systems and advanced technologies currently used for the evaluation and monitoring of other liver diseases will play an instrumental role in the clinical management of NAFLD. Natural history studies and biopsy series have highlighted stage F2 fibrosis as an important point of clinical intervention, suggesting the potential for implementing therapeutic interventions to achieve reduction of both all-cause and liver-specific mortality. Although overweight individuals with metabolic syndrome, insulin resistance, and hepatic steatosis have increased risks of NAFLD with or without fibrosis, clinicians must be aware of the limitations in relying solely on noninvasive markers for assessment of fibrosis. There are currently several noninvasive methods used to assess fibrosis, and despite their limitations, they provide valuable information when evaluating patients with NAFLD. Because the risk of liver-specific mortality increases by a factor of 50 to 80 in patients with stage F3 or F4 fibrosis related to NASH, it is important to diagnose and stage the disease effectively.[10,11]

Patients at high risk for NASH with subsequent fibrosis and liver cancer should receive advanced testing to confirm the diagnosis, evaluate the level of hepatocyte damage, and stage the fibrosis. Although liver biopsy is the traditional and most widely accepted method of diagnosing NASH and staging fibrosis, its associated limitations and potential complications, the vast cohort of patients who require disease staging, and the increasing availability and accuracy of noninvasive methods have made liver biopsy less common. The application of noninvasive approaches, including surrogate serum biomarkers, validated predictive scoring systems, and imaging modalities, has come to the clinical forefront. Although the use of genetic screening for polymorphisms such as patatin-like phospholipase domain-containing protein 3 and transmembrane 6 superfamily member 2 may contribute a more substantial role in the future, these tests are rarely used clinically today.

Aminotransferase Biomarker

The persistent elevation of alanine aminotransferase (ALT) levels may suggest the presence of hepatic steatosis and/or steatohepatitis, but advanced disease can present with normal aminotransferase levels, thus reflecting the poor sensitivity and specificity of using ALT biomarker alone in NAFLD-related diagnosis and disease staging.[12,13] Studies have suggested that normal ALT levels are found in 30% to 60% of patients with biopsy-confirmed hepatic steatosis. Furthermore, low-grade liver injury can be missed due to inappropriately high laboratory reference ranges for ALT, and lower cutoffs for defining normal ALT levels may improve the sensitivity.[13,14] However, the level of elevation does not correlate well with disease severity, emphasizing the importance of developing improved biomarkers for more accurate disease monitoring and prognostication.[7] Furthermore, elevation of ALT itself may also not be a strong predictor of risk of disease progression, and, thus, a comprehensive assessment of metabolic risk factors and other comorbidities that may lead to a more aggressive natural history in NAFLD should be conducted. Precise thresholds of ALT elevation for NAFLD diagnosis and disease staging have not been clearly established and reflect the need to incorporate multiple tools to accurately assess disease severity.[15,16]

Other biomarkers have shown even lower prognostic value in the detection of underlying hepatic steatosis. In some patients, alkaline phosphatase can be elevated 2 to 3 times above the upper limit of normal, whereas total bilirubin and albumin levels are often normal. Serum ferritin, antinuclear antibody, and antismooth muscle antibody may also be elevated in some patients, but these laboratory-based tests are neither sensitive nor specific enough to be incorporated into a NAFLD-related diagnostic and staging algorithm.

Noninvasive Assessment of Nonalcoholic Fatty Liver Disease

Certain risk factors, such as insulin resistance, obesity, and metabolic syndrome, are associated with an increased

risk of NAFLD, but it is not yet clear which patient populations would benefit from screening for NAFLD and which diagnostic modality would be the most cost-effective. The American Association for the Study of Liver Diseases guidelines state that routine screening for NAFLD, even in high-risk patients, is not currently recommended given the uncertainty of diagnostic tests, availability of treatments, and lack of clarity on long-term benefits and cost-effectiveness of screening.[15] Elevations in laboratory enzymes or liver function tests may trigger a diagnostic cascade that leads to a diagnosis of NAFLD, whereas hepatic steatosis is often incidentally observed on imaging tests performed for other reasons.

The early diagnosis and quantification of the severity of hepatic steatosis may prompt further evaluation and more aggressive management of metabolic comorbidities. Multiple radiologic techniques are available, but ultrasonography is often the initial radiologic assessment performed given its low cost and ease of accessibility. However, it is important to note that ultrasonography is up to 93% sensitive when hepatic steatosis exceeds 33% of the hepatic parenchyma affected; thus, in patients with lower levels of hepatic steatosis involvement, ultrasonography may be suboptimal. Therefore, this modality is useful with moderate to severe hepatic steatosis but poorly detects early-stage NAFL.[17] Controlled attenuation parameter using the FibroScan (Echosens) probe is an emerging technology that has shown correlation with hepatic steatosis grade, but is limited by increasing abdominal and visceral adiposity found with increasing body mass index.[18-20] Magnetic resonance imaging (MRI) approaches 100% sensitivity in the detection of hepatic steatosis, even with steatosis levels as low as 5.56%.[21] Computed tomography has not been shown to have improved sensitivities in mild steatosis, but carries additional radiation exposure and increased cost.[22] Thus, its role in the routine assessment of NAFLD is currently unclear.

Noninvasive Assessment of Nonalcoholic Steatohepatitis

While the early detection of hepatic steatosis is important, more significant is the distinction between isolated hepatic steatosis and NASH, which is characterized by a far worse prognosis given the higher risks of progression to fibrosis. The use of the ALT and aspartate aminotransferase (AST) biomarkers alone has poor correlation with NASH.[23] Of note, patients with metabolic syndrome with ultrasound-confirmed hepatic steatosis are at elevated risk of NASH regardless of AST or ALT levels.[15] Multiple pilot analyses in heterogeneous groups of patient populations have been studied to understand the combinations of clinical and laboratory parameters to noninvasively diagnose NASH. The measurement of plasma cytokeratin 18 with other biomarkers has shown some improvement in diagnostic value, but only marginal enhancement. The sensitivity and specificity of plasma cytokeratin 18 for the detection of NASH is 58% and 68%, respectively.[24] Another potential biomarker currently being studied, NIS4, is a proprietary test developed by a collaboration between Genfit and LabCorp. Although multiple biomarkers are under active investigation in the research space, there are currently no candidates to replace or even substitute liver biopsy for accurate diagnosis of NASH.

Imaging studies are limited primarily due to their inability to distinguish between the 2 similar but clinically different diseases. Evolving MRI techniques are showing promising data that support the correlation between hepatic steatosis and overall NAFLD Activity Score (NAS) response. MRI–estimated proton density fat fraction (MRI-PDFF) has demonstrated the ability to discriminate between longitudinal changes in fibrosis in response to certain treatments. This modality has a sensitivity of 89%, specificity of 47%, positive predictive value of 39%, and negative predictive value of 92% in the assessment of fibrosis in NAFLD.[25] In a retrospective analysis of patients with NAFLD or NASH, MRI-PDFF correlated with steatosis grade and NAS when the NAS was 3 or less.[26]

Noninvasive Assessment of Hepatic Fibrosis

The investigation of noninvasive tools that predict the progression of NAFLD is a major research focus due to the significant clinical impact that these tools will have on accurate prognostication. The development of fibrosis is the most important characteristic of NAFLD owing to its correlation with clinical outcomes. The presence and extent of fibrosis was found to be the major factor in predicting clinical decompensation and death.[11]

There are multiple surrogate predictor models that utilize a combination of clinical parameters with serum biomarkers that quantify the severity of fibrosis with high accuracy (Table). The NAFLD Fibrosis Score (NFS) is a well-studied and validated tool that has significant data to support its use in the prediction of liver-related outcomes.[27] The NFS uses multiple commonly ordered laboratory tests to identify the severity of fibrosis. Overall, the NFS has a sensitivity and specificity of 75% and 58%, respectively, for excluding advanced fibrosis when the score is less than -1.455. The NFS has a sensitivity of 33% and a specificity of 98% for identifying advanced fibrosis when the score is greater than 0.676.[28,29] Other commonly used noninvasive scoring systems include the AST to Platelet Ratio Index and Fibrosis-4 scores.[30]

Table. Components of the Common Noninvasive Serologic Biomarkers for Assessment of Hepatic Fibrosis in NAFLD

Noninvasive Scoring System	AST	ALT	Platelet Count	Age	Diabetes	Albumin	BMI
NAFLD Fibrosis Score[27]	x	x	x	x	x	x	x
AST to Platelet Ratio Index[51]	x		x				
Fibrosis-4 Score[52]	x	x	x	x			
BARD Score[53]	x	x			x		x

ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; NAFLD, nonalcoholic fatty liver disease.

Emerging radiologic technologies with high accuracy for assessing liver fibrosis continue to play increasingly important clinical roles in NASH patients. Vibration-controlled transient elastography (FibroScan) is the most-studied and commonly accessible outpatient technology for identifying the severity of hepatic fibrosis, as well as one of the most reliable and accurate modalities in the noninvasive assessment of hepatic fibrosis.[30] However, as previously mentioned, the accuracy of this tool is limited by increasing abdominal and visceral adiposity as well as other factors that may affect potential assessment of liver stiffness (eg, abdominal ascites and severe active hepatitis).[31,32]

Magnetic resonance elastography (MRE) may be superior to vibration-controlled transient elastography, but is limited by cost and availability, and is typically offered only at major academic medical centers. Two-dimensional MRE has a sensitivity of 86% and a specificity of 91% when identifying advanced fibrosis with the area under the receiver operating characteristic curve of 0.92 when distinguishing between early and advanced fibrosis.[33] MRE is a highly accurate noninvasive diagnostic tool for quantifying fibrosis that is capable of being used in clinical trial outcomes and in disease monitoring in clinical practice.[34]

Liver Biopsy

Liver biopsy is the standard diagnostic and staging tool. However, this modality is limited by its invasiveness, risk of complications, and sampling error.[35] The replacement of liver biopsy with an equally accurate noninvasive modality will provide much-needed improvements and advancements to the diagnostic field of NAFLD. A meta-analysis found that the presence of NASH on an initial liver biopsy is the strongest predictor of subsequent liver fibrosis.[36] Furthermore, a study that evaluated patients for a follow-up period of up to 33 years found that the progression of liver fibrosis is the strongest predictor of poor liver-related outcomes.[10] Consideration for biopsy is indicated in any patient with high clinical suspicion of steatosis, metabolic syndrome, diabetes mellitus, and high risk for NASH.[9]

Treatment

Lifestyle interventions such as diet and exercise are the crux of treatment for the vast majority of patients with NAFLD. Weight loss of at least 10% has been shown to correlate with significant improvements in the histologic grade as well as NAS reduction, NASH resolution, and fibrosis regression.[37] In a clinical trial with 154 patients randomized to either a dietitian-reinforced lifestyle intervention or general standard-of-care recommendations to lose weight for a time period of 12 months, resolution of NAFLD was observed in 64% of the intervention group compared to 20% in the control arm.[38] This study highlights the importance of diet in the management of NAFLD patients. The optimal diet for patients with NAFLD remains unclear, but the composition of diet appears to play a critical role.[39,40] In a study that compared the Mediterranean diet with an isocaloric low-fat and high-carbohydrate diet during a 6-week period to understand the effect on liver adipose tissue, the Mediterranean diet was associated with reduced liver fat and improved insulin sensitivity, but no difference in weight loss was observed.[41]

The US Food and Drug Administration has not yet approved a NAFLD-specific therapy for the management of NASH, but multiple clinical trials have shown some therapeutic benefit. The first randomized clinical trial to show clinical benefit, defined as histologic improvement, was the PIVENS trial.[42] Two hundred and forty-seven nondiabetic patients who had biopsy-confirmed NASH were randomized to 3 arms: (1) 30 mg of pioglitazone daily, (2) 800 IU of vitamin E daily, or (3) placebo. Vitamin E therapy was associated with significant improvements in hepatic steatosis, ballooning degeneration, insulin resistance, aminotransferase levels, and hepatic inflammation. The results in the pioglitazone arm did not meet the predefined measures of statistical significance, but additional studies have demonstrated the beneficial

impact of pioglitazone in patients with diabetes and insulin resistance.[43] Two subsequent meta-analysis studies demonstrated that the histologic improvement from pioglitazone therapy also included a reduction in hepatic fibrosis.[44,45] Although a complete discussion of potential therapeutics on the horizon is beyond the scope of this article, there are currently 4 main molecules that are in phase 3 clinical trials: obeticholic acid[46] (Ocaliva, Intercept Pharmaceuticals; REGENERATE trial), elafibranor[47] (GFT505, Genfit; RESOLVE-IT trial), cenicriviroc[48] (AURORA trial), and selonsertib[49] (STELLAR 3 and 4 trials). There is also a role for bariatric surgery in carefully selected patients with NAFLD, with a recent study demonstrating that the weight loss achieved with bariatric surgery translates into improvements in steatosis, reduction in hepatic inflammation, and improvements in insulin resistance.[50]

Summary

NAFLD is the leading cause of chronic liver disease and affects as many as 100 million US adults. However, the vast majority of these patients do not progress to NASH, and even fewer progress to fibrosis- and cirrhosis-related complications. Accurate diagnosis and disease severity staging is critical for providing prognostication as well as ensuring that appropriate monitoring is implemented. Furthermore, although there is currently a paucity of effective therapies for NAFL and NASH specifically, several therapeutic regimens on the horizon offer promising results. The current landscape of diagnostic testing modalities offers a wealth of important tools that should be used in combination, including serology- and imaging-based tests, to provide the most accurate disease staging to help with continued disease monitoring and future assessment of treatment eligibility.

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