HHS Public Access

Author manuscript

Hepatology. Author manuscript; available in PMC 2016 June 09.

Published in final edited form as:

Hepatology. 2015 April ; 61(4): 1392–1405. doi:10.1002/hep.27678.

CHALLENGES AND OPPORTUNITIES IN DRUG AND BIOMARKER DEVELOPMENT FOR NONALCOHOLIC STEATOHEPATITIS: FINDINGS AND RECOMMENDATIONS FROM AN AMERICAN ASSOCIATION FOR THE STUDY OF LIVER DISEASES (AASLD) - FOOD AND DRUG ADMINISTRATION (FDA) JOINT WORKSHOP

Arun J. Sanyal, MBBS, MD[1], **Scott L. Friedman, MD**[2], **Arthur J. McCullough, MD**[3], and **Lara Dimick, MD**[4]

[1]Div. of Gastroenterology, Hepatology and Nutrition, Dept. of Internal Medicine, Virginia Commonwealth University School of Medicine, Richmond, Virginia

[2]Div. of Liver Diseases, Icahn School of Medicine at Mount Sinai, New York, New York

[3]Dept. of Gastroenterology and Pathobiology, Cleveland Clinic Lerner College of Medicine, Cleveland, Ohio

[4]Food and Drug Administration, US Federal Government

Abstract

Nonalcoholic fatty liver disease (NAFLD) is the most common cause of chronic liver disease in North America. It is a growing contributor to the burden of chronic liver disease requiring liver transplantation. Cirrhosis is also associated with an increased risk of hepatocellular cancer which may occur even in the absence of cirrhosis in subjects with nonalcoholic steatohepatitis (NASH) the histological form of NAFLD associated with increased liver-related mortality. The diagnosis of NASH currently requires a liver biopsy. There are also no FDA-approved therapies for NASH. There is therefore a need to develop better diagnostic and therapeutic strategies for patients with NASH targeting both those with early stage disease as well as those with advanced liver fibrosis. There are unique challenges in the design of studies for these target populations. The long relatively asymptomatic time interval in the progression of NAFLD and NASH to cirrhosis and ultimately liver failure, along with gaps in knowledge regarding disease modifiers combine to present significant challenges in trial design. There is therefore an urgent need to develop methods

Arun J. Sanyal M.D., MCV Box 980341, Richmond, VA 23298-0341, asanyal@mcvh-vcu.edu
Scott L. Friedman M.D., Box 1123, Icahn School of Medicine at Mount Sinai, 1425 Madison Ave, Room 1170C, New York, NY 10029, scott.friedman@mssm.edu
Arthur McCullough M.D., Cleveland Clinic, Department of Gastroenterology/A31, 9500 Euclid Avenue, Cleveland, OH 44195, mcculla@ccf.org
Lara Dimick-Santos, M.D., Food and Drug Administration, 10903 New Hampshire Ave., Silver Spring, MD 20093, Lara.Dimick@fda.hhs.gov

Conflicts of Interest: These are provided separately and are also available from the American Association for Study of Liver Diseases.

to identify the populations at particular risk of disease progression and to validate endpoints that reflect meaningful changes in health status in this population.

This manuscript summarizes the discussion at a joint workshop held September 5th and 6th, 2013, in Silver Spring, Maryland, sponsored by the FDA and the AASLD to develop guidance on diagnostic and therapeutic modalities for NASH.

Keywords

nonalcoholic steatohepatitis; nonalcoholic fatty liver; nonalcoholic fatty liver disease; cirrhosis; model for end stage liver disease; hepatic venous pressure gradient; quantitative liver function tests; biomarkers; type 2 diabetes mellitus; metabolic syndrome

1. Introduction

Nonalcoholic fatty liver disease (NAFLD) is the most common cause of chronic liver disease in North America and is emerging as a leading cause of liver-related morbidity and mortality (1, 2). There are two major clinical-histological phenotypes of NAFLD: (a) nonalcoholic fatty liver (NAFL) and (b) nonalcoholic steatohepatitis (NASH) (3, 4). It is estimated that about 30% of the adult population and at least 10% of children in the Western world have NAFLD (5–8). It is further estimated that 20–25% of individuals with NAFLD have NASH (3),(9). Subjects with NASH have increased morbidity and mortality from cardiovascular, cancer and liver-related events including hepatocellular cancer compared to the general population (10–14). NAFLD has been linked to an increased risk of development of type 2 diabetes (15–20). Given the growing burden of end stage liver disease due to NASH, it is a health care priority to advance both diagnostic and therapeutic strategies for patients with NASH (21–23).

There are numerous stakeholders involved in developing strategies for NASH. They include academia, the National Institutes of Health, pharmaceutical and device manufacturers, developers of diagnostics, the U.S. Food and Drug Administration (FDA), health care providers, professional associations, insurance providers and the patients themselves.

The FDA and the American Association for Study of Liver Diseases (AASLD) jointly sponsored a workshop in September 2013 to tackle this challenge. This workshop assembled major stakeholders to discuss specific challenges and opportunities to facilitate development of therapeutics for NASH. While general consensus was obtained in several areas, there also remain areas where there is a need for ongoing dialogue. This manuscript summarizes the discussion at the workshop. It also identifies gaps in knowledge that represent important barriers towards progress. The workshop participants agreed that a critical component of the drug development process for NAFLD is the demonstration of benefit with respect to clinically meaningful outcomes, and the development of surrogates that are reasonably likely to predict irreversible morbidity or mortality.

Individual groups engaged in developing new diagnostics or treatments for NASH are encouraged to communicate directly with the FDA through the pre-IND consultation program for early advice on their development programs. The FDA also has established

mechanisms for interactions with outside stakeholders to discuss regulatory science in a non-binding setting. Examples include the Voluntary Exploratory Data Submissions (VXDSs) and Critical Path Innovation Meeting (CPIM)[1] pathways.

2. Background

NAFLD is a clinical-histological syndrome characterized histopathologically by predominantly macrovesicular steatosis with varying amounts of inflammation, cytological ballooning and fibrosis (24). By definition, it occurs despite minimal (< 2–3 units/day) or no alcohol intake (25). The histological and clinical spectrum of the disease is briefly summarized below. Interested readers are referred to several excellent review articles for more details (3, 26–30).

The histological spectrum of NAFLD extends from fatty liver to steatohepatitis (4, 31). Nonalcoholic steatohepatitis (NASH) is characterized by steatosis, inflammation and cytological ballooning with varying amounts of pericellular fibrosis (14, 32). These lesions are classically most pronounced in zone 3 but there may be other patterns of distribution as well. In children, for example, zone 1 inflammation and periportal fibrosis are typically more pronounced than in adults (33). Disease progression is characterized by increasing fibrosis and cirrhosis in a subset of patients.

The principal risk factors for NAFLD include excess body weight, insulin resistance, type II diabetes, hypertension, low high-density lipoproteins and hypertriglyceridemia (3, 20). Subjects, even those with significant histological changes, are frequently either asymptomatic or have nonspecific symptoms (e.g. fatigue). The lack of specific symptoms or signs in such individuals is a significant obstacle to diagnosis before development of cirrhosis when treatment of the underlying disease can potentially avoid cirrhosis the principal cause of liver-related mortality in this population. In addition, there are very little data available to allow differentiation of subjects who will progress from subjects who will not progress to clinically relevant hepatic disease. Therefore, the development of new diagnostics that can differentiate those with NAFLD who will progress to clinically significant disease represent a major research priority. Liver-related symptoms such as jaundice, pruritus, anasarca, ascites, hepatic encephalopathy, and gastrointestinal bleeding are late manifestations of NAFLD and reflect the presence of end-stage liver disease (i.e., cirrhosis). Occasionally, NAFLD may present with subacute liver failure, with or without a clear cut precipitating factor (34).

3. Food and Drug Administration (FDA) Regulatory Pathways

The FDA has two main pathways for drug development. The first pathway is the “regular” or “traditional” pathway for drug approval, in which a drug is approved based on either a clinical benefit endpoint or a surrogate endpoint that is known to predict clinical benefit on irreversible morbidity or mortality. A clinical endpoint is one that affects how a patient feels, functions or survives.

[1]Send inquiries about the CPIM to CPIMInquiries@fda.hhs.gov.

The second option is the Accelerated Approval Pathway (21 CFR 314.510 and 601.41, Subpart H and E). Accelerated approval applies to drugs and biologics that are intended to treat serious and life threatening illness AND that provide meaningful therapeutic benefit over existing treatments.

Accelerated approval is based on effects of a surrogate endpoint that is "reasonably likely, based on epidemiologic, therapeutic, pathophysiologic, or other evidence to predict benefit" on irreversible morbidity or mortality. The Agency may grant accelerated approval to a product for a serious or life-threatening condition upon a determination that the product 1) has an effect on a surrogate endpoint that is reasonably likely to predict clinical benefit or, 2) an effect on a clinical endpoint that can be measured earlier than an effect on irreversible morbidity or mortality and is reasonably likely to predict an effect on irreversible morbidity or mortality or other clinical benefit, taking into account the severity, rarity, or prevalence of the condition and the availability or lack of alternative treatments.

The term "reasonable likely" implies that some uncertainty remains about the relationship of the surrogate to the clinical benefit to the patient. Therefore, accelerated approval is usually contingent on a sponsor's agreement to conduct additional post-approval studies to verify and describe the drug's clinical benefit. This regulatory pathway requires that when marketing approval is granted based on the surrogate, or a clinical endpoint other than survival or irreversible morbidity, clinical trials must be carried out after marketing approval to verify and describe the drug's clinical benefit.

The preamble to the Accelerated Approval Rule, (Federal Register/Vol.57, No.239/Friday. December 11, 1992/Rules and Regulations), acknowledged that surrogate endpoints can be used for regular or traditional approval when these surrogates are validated by definitive studies. It states that "Ordinarily, products used to treat serious or life-threatening illness, for which approval is based on a surrogate endpoint that is recognized as validated by definitive studies, will be considered for approval under the traditional process rather that under accelerated approval." The Agency's draft Guidance for Industry - Expedited Programs for Serious Conditions—Drugs and Biologics (http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM358301.pdf) states: "For purposes of accelerated approval, a surrogate endpoint is a marker, such as a laboratory measurement, radiographic image, physical sign, or other measure that is thought to predict clinical benefit, but is not itself a measure of clinical benefit. Depending on the strength of the evidence supporting the ability of a marker to predict clinical benefit, the marker may be a surrogate endpoint that is known to predict clinical benefit (a validated surrogate endpoint, which could be used for traditional approval), a surrogate endpoint that is reasonably likely to predict a drug's intended clinical benefit (which could be used for accelerated approval), or a marker for which there is insufficient evidence to support reliance on the marker as either kind of surrogate endpoint (and thus cannot be used to support traditional or accelerated approval of a marketing application). Change in the rate of decline in renal function as measured by creatinine clearance, and changes in blood pressure and Hg A1C have been accepted as validated surrogates to support traditional drug approvals.

However, the preamble does not specifically define the criteria for "validated by definitive studies" that allows a surrogate to be used for traditional approval. Therefore the level of evidence necessary to determine if a surrogate is validated (i.e., acceptable for traditional or regular approval pathways), or if a surrogate is reasonably likely to predict clinical benefit is made on a case-by-case basis by the Agency. Whether an endpoint is reasonably likely to predict clinical benefit is a function of the biological plausibility of the relationship between the disease, endpoint, and the desired effect, and the empirical evidence to support that relationship. The empirical evidence may include "epidemiological, pathophysiological, therapeutic, pharmacologic, or other evidence developed using biomarkers, for example, or other scientific methods or tools." Clinical data should be provided to support the assertion that a relationship of the surrogate or intermediate clinical endpoint to the outcome is reasonably likely, and should be relevant to the relationship between the specific endpoint to be used and the specific intended clinical benefit of the drug.

What is clear is that there are pitfalls with using surrogates, i.e., examples of where a plausible surrogate that showed improvement with treatment resulted in an overall poor outcome for the patient. Some of these unexpected outcomes may be from off-target effects of a drug. Several things should be taken into consideration when evaluating a surrogate for use in the regular approval pathway. A surrogate that directly measures tissue loss or organ function e.g. liver failure is particularly plausible for use as an endpoint. Also helpful are the results of controlled trials that show the relationship of the effect on outcomes in trials of other drugs, especially trials involving drugs with different mechanisms, as is the case for example with antihypertensives.

See Guidance "Expedited Programs for Serious Conditions—Drugs and Biologics":http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM358301.pdf for further details.

4. Clinical Endpoints

A: Endpoints based on symptoms and functional status

Measurements of a change in clinically meaningful symptoms of a disease (how the patient feels and functions) are acceptable as primary endpoints for the regular approval pathway. NASH significantly impacts many aspects of patients' quality of life. Both adults and children with NASH have poorer health related quality of life scores than healthy controls (35–39).

Most subjects with NAFLD or NASH, are able to function, maintain a job and manage day-to-day activities. There is a need for additional research to identify the frequency and prevalence of physical and mental disability in those with NAFLD, while correcting for confounding variables such as obesity, diabetes and its complications, among others.

Consideration of the risk/benefit balance in this population is important as the majority of patients will not progress; therefore, potential interventions should have a low risk profile. However, as a subset of patients with NASH will progress to cirrhosis, it is important that

efforts continue in developing tools to identify high risk populations and target them for appropriate and safe interventions.

Once cirrhosis develops, there is a progressive impairment in liver function, which eventually leads to multiple symptoms that negatively impact patients' ability to function (40). With progression of disease, measurement of clinical endpoints that assess the patients' symptoms and ability to function becomes feasible. In studies targeting advanced stage disease, it will be valuable to capture the functional status of individual subjects.

In order to assess treatment benefit in clinical trials, well-defined and reliable clinical outcome assessments (COA) may be selected or developed to assess how patients with cirrhosis feel or function in their daily lives. Symptoms can be known only to the patients themselves, and should be assessed using well-defined and reliable patient-reported outcome (PRO) assessments (FDA PRO Guidance for Industry: http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM193282.pdf)

For patients unable to report for themselves (e.g., young children, those with cognitive impairments), other COAs (e.g., clinician-reported outcome (ClinRO) or other observer-reported outcome (ObsRO) assessments) may be used to assess patients' observable behaviors. All COAs require appropriate development and testing within the target patient population in order to ensure they are adequately defined and reliable to detect meaningful and interpretable changes within a clinical trial. Given the high prevalence of symptoms and impaired mental and physical health in patients with cirrhosis, it is important to evaluate outcomes from the patient perspective in the context of long-term clinical trials using appropriately selected or newly-developed instruments.

Potential targets of treatment were discussed and some options that were considered included prevention of progression of fibrosis, prevention of progression to cirrhosis, and prevention or treatment of symptoms. A measurable and objective composite liver-related outcome endpoint could be used to assess events of decompensation in compensated cirrhotic patients. The rates of development of events of decompensation in patients with compensated cirrhosis are well known (41, 42), and specific data for the rate of development of cirrhosis in NASH patients is also known (43). However, even in the cirrhotic population, in view of the rates at which these outcomes develop, large sample sizes or long trials will be required to demonstrate statistically significant differences in rates of development of the events of interest in a reasonable timeframe. These timeframes may be potentially reduced if the treatment effect is large or by enriching the eligible trial population. Enriching the cirrhosis population could be potentially achieved by using patients with a hepatic venous pressure gradient (HVPG) > 10 mm Hg or a MELD score > 10 (44).

Other clinical endpoints are likely to be meaningful to NASH patients with cirrhosis. These include the rates of hospitalization, unscheduled clinic and emergency room visits, tests performed, and lost work days (45). Together with an endpoint measuring a clinically meaningful change in health status, these might provide a more comprehensive picture of an intervention's potential benefit.

B. Endpoints based on “hard clinical outcomes”

B1: Survival as a Clinical Endpoint in Trials for NASH or Cirrhosis—All-cause mortality has long been held as the most important outcome and thus a key endpoint in clinical trials evaluating therapies for many chronic diseases that can lead to death, including NAFLD and NASH (11, 30, 46). Liver-related mortality is a component of all-cause mortality. It is closely linked to the development of hepatocellular cancer, ascites and related complications, variceal hemorrhage, hepatic encephalopathy and eventually acute or chronic liver failure, usually precipitated by sepsis (43, 46). It is well established that the development of any one of these complications heralds an immediate deterioration in health status (i.e., clinical decompensation) and an increase in mortality risk (41, 42). These complications are virtually all linked to the presence of cirrhosis. Hepatocellular cancer is an exception to this rule and can develop in the absence of cirrhosis. Recent studies suggest that almost 50% of hepatocellular cancers related to NAFLD occur in the absence of cirrhosis (47).

NASH progresses slowly to cirrhosis; 15–20% of subjects develop cirrhosis over many years (30, 48). Cirrhosis, the principal cause of liver-related deaths, is associated with a decompensation and mortality rate of approximately 4% annually (43, 49). Therefore, to demonstrate an improvement in mortality rates (all-cause or liver-related) a large number of subjects with early stage NASH would need to be followed for longer than 10 to 15 years. The costs and logistics of such an endeavor are prohibitive, making all-cause or liver-related mortality endpoints difficult to use for drug development and approval. Therefore, all-cause mortality may be impractical as a primary endpoint for most trials that will enroll patients who have NASH without cirrhosis. In subjects with NASH who have established cirrhosis, where decompensation, death or cancer is more imminent, all-cause or liver-related mortality are operationally feasible clinical endpoints but are still likely to require long time frames to measure.

B2: Surrogate Endpoints—As noted above, surrogate endpoints can be used as part of the regular approval pathway if these endpoints have been validated by definitive trials to predict outcomes on clinically meaningful endpoints. Surrogates may be used to support accelerated approval when there is not an established link of the surrogate to the clinical outcome.

The long duration over which NASH evolves before patients develop clinical outcomes, and the inability to identify the population that will progress, creates a major challenge in designing and conducting clinical trials for those with early stage NASH. This challenge, however, does not diminish the need to develop therapeutics in these patients, and identify who is at risk of progression, given the growing contribution of progressive NASH to the burden of chronic liver disease. It is also important to identify endpoints that can be achieved within a reasonable time interval and that are reliable surrogates for meaningful outcomes. Moreover, these endpoints should both reflect changes in the disease process and be ‘biologically plausible’, in other words, linked mechanistically to the disease’s pathogenesis.

B2.1 Histology-based Endpoints in NASH Trials: Liver histology currently offers the best short-term method for tracking the progression of NASH. Certain features on histopathology provide some prognostic information regarding risk for progression. Steatohepatitis, not isolated fatty liver, is associated with a substantial increase in the long-term risk of developing cirrhosis and liver-related outcomes (30, 48). This is believed to be related to the underlying inflammation and activation of pro-fibrogenic pathways in NASH. Based on this current understanding of the pathogenesis of NASH, one would expect that reversal of steatohepatitis would reduce the risk of developing cirrhosis. However, steatosis and inflammation can decrease as fibrosis advances (50). Therefore, the reversal of steatohepatitis with no evidence of progression to advanced fibrosis (stage 3 or 4), may be an acceptable surrogate endpoint suitable both for phase 2b and 3 trials that enroll patients with NASH and evidence of early fibrosis. In a recent clinical trial of vitamin E for NASH, patients who resolved their steatohepatitis also were observed to have improvement in fibrosis (51). Note that due to the uncertainty about the ability of reversal of steatohepatitis to predict a true clinical benefit, to establish the clinical benefit of a drug, outcome trials will be eventually needed to demonstrate that reversal of steatohepatitis does predict clinical outcomes.

A decrease in the NAFLD activity score (NAS) has also been used as an endpoint in clinical trials (52), however, its value is limited by a lack of data linking changes in NAS and its individual component scores, to either progression to advanced fibrosis or clinical outcomes. The advantage of NAS is that it is quantifiable and relatively more reproducible than the diagnosis of steatohepatitis. Additionally, there is ongoing discussion about the need to generate data to either validate the use of the NAS as a valid determinant of NASH clinical outcomes and/or the response to therapy. This would best be accomplished by incorporating a change in NAS as a secondary endpoint in randomized clinical trials.

B2.2 Development of Cirrhosis as Potential Surrogate Endpoint for NASH Clinical Trials: There is a body of data that supports that the development of histologically identified cirrhosis predicts a significant worsening of health status. Differences in progression to cirrhosis between treatment and control groups were discussed at the workshop as a potential clinically meaningful outcome measure for clinical trials (40, 41, 43, 46, 49, 53–56). However, there are known weaknesses that are well documentated and discussed in the literature with using histopathology assessments in clinical trials and limitations to their use in clinical practice. However, because there is a substative body of evidence to support that cirrhosis on histolology is predictive of clinical outcomes, it was proposed by some at the workshop as a acceptable surrogate endpoint for “regular” approval under FDA guidelines or as the endpoint for required post-approval studies for treatments approved via the Accelerated Approval pathway.

Development of cirrhosis can be measured by transient elastography methods; the potential therefore exists that an approval based on non-invasive measures may be acceptable in some cases. There are some data to support the correlation of liver stiffness with clinical outcomes of events of decompensation, HCC and death (57). This would avoid the need for repeated liver biopsies in trials that utilize the difference in progression to histologically defined cirrhosis as an endpoint. However, more data will be required to support this endpoint for

use in clinical trials. Including such measures as secondary or exploratory endpoints in current trials will provide the necessary data to establish the validity of these potential noninvasive markers.

B2.3: Surrogate Endpoints for Clinical Trials for patients with Cirrhosis (Table 2): There are also several surrogates that were put forward for consideration at the workshop that may be acceptable as surrogates "likely to predict" mortality or morbidity risk in subjects with compensated cirrhosis, for example the Child Pugh Turcotte score (CPT) and the Model for End Stage Liver Disease (MELD) score, and the hepatic venous pressure gradient (HVPG).

i. Child-Pugh-Turcotte Score: The Child-Pugh-Turcotte score was originally developed as a tool to evaluate mortality risk following portacaval shunt surgery in those with cirrhosis (58). It has been shown to be associated with mortality over 1–5 years. Progression from Child class A to class B accurately measures worsening of a given patient's health status and an increase in mortality risk (59). The CPT score suffers from several limitations however; for example, a bilirubin of 5 mg/dl and 15 mg/dl would both be scored identically while they clearly represent different health states. CPT also has a subjective element, which may introduce bias in assessment of outcomes. Despite these limitations, it remains a time-tested way to assess 1–5 year mortality risk in subjects with advanced liver disease. Progression from Child class A to B or a 2-point worsening of the CPT score were discussed as potential surrogate endpoints, but the precise change in score to be used as a responder criterion in clinical trials remains uncertain.

ii. Model for End Stage Liver Disease (MELD) Score: The MELD score is among the best predictors of short-term (3 month) mortality risk in those with decompensated cirrhosis. MELD has a more sensitive dynamic range than the CPT score and is used to determine organ allocation for liver transplantation (60, 61). MELD is objective, easy to measure, widely available and supported by a very large body of evidence as a predictor for mortality risk over a 3 month period (60, 62–67).

Patients with NASH and compensated cirrhosis generally have low MELD scores, and the ability of low MELD scores to predict changes in mortality are not as robust as high MELD scores. Subjects with a MELD score > 10 are however more likely to experience decompensation and a liver-related clinical outcome than those with lower scores (44, 68). The benefits of liver transplantation only emerge once the MELD exceeds 14 (69), and therefore a MELD score > 14 is typically the minimal listing threshold for transplantation. While many other factors also determine whether a patient is listed for transplant, a MELD of 14 or higher is an endpoint that meets the criteria for a surrogate that may be acceptable for clinical trials, in that it has a strong relationship to mortality and outcomes, is objective, easy to measure, sensitive and widely available. There was discussion of the potential of MELD as a surrogate endpoint based on sufficient evidence to establish a link to "need for transplant" and "mortality"; specifically, a MELD of 15 or higher with a minimal 2 point change from baseline would establish these links in a population with cirrhosis. Because "need for transplant" is an irreversible morbidity, if MELD score is the criterion used to make a decision to transplant, achieving the transplantation-qualifying MELD score was

discussed as a potentially reasonable clinical trial endpoint for clinical trials intended to support drug approval.

iii: Hepatic venous pressure gradient (HVPG): The HVPG was discussed as a potential surrogate endpoint that might be considered reasonably likely to predict clinical outcome to support accelerated approval. HVPG measures the pressure differential from the portal to the hepatic vein and thus provides a physiological readout that integrates the hemodynamic consequences of increased sinusoidal resistance to flow due to hepatic fibrosis and/or increased portal inflow. Its methodology is well established and it is reproducible (70, 71), with a large body of literature that demonstrates its concordance with liver related outcomes (70, 72–76). It is limited by being an invasive procedure and requiring standardization of the procedures used to obtain the pressure measurement. Cirrhosis related complications heralding clinical decompensation largely occur above a threshold HVPG of 10 mm Hg, which has been established as the cutoff for "clinically significant portal hypertension" (44, 68). Reduction of the proportion of subjects that progress to developing a HVPG > 10 mm Hg was posed as a treatment objective and surrogate primary endpoint for consideration, in the setting of accelerated approval in trials of therapeutics for patients with NASH with advanced fibrosis who have baseline HVPG values below 10 mm Hg at study entry. Lowering HVPG from values > 10 mm Hg to less than this threshold value was also posed for consideration as a surrogate endpoint to support accelerated approval in patients with NASH with bridging fibrosis or advanced cirrhosis and elevated HVPG.

C. Other Surrogate Endpoints for disease progression

C1: Quantitative liver function tests—There has been considerable interest in the development of quantitative liver tests as markers of overall hepatic integrity and functional reserve, with several such tests in various phases of development. In order to be acceptable as surrogate endpoints for clinical trials intended to support Subpart H or E approval, all functional tests must have adequate data to support their use as a surrogate that is "reasonably likely" based on epidemiologic, therapeutic, pathophysiologic, or other evidence, to predict benefit" on irreversible morbidity or mortality.

5. Other Endpoints in Trials for NASH

The objectives of early phase trials are to assess safety and to obtain a signal of efficacy that will guide decision-making about further developing a specific drug. The endpoints for such trials should include traditional endpoints for evaluating safety, including potential hepatotoxicity (FDA Guidance - Drug-Induced Liver Injury: Premarketing Clinical Evaluation; http://www.fda.gov/downloads/Drugs/…/Guidances/UCM174090.pdf).

Other endpoints were discussed that may, with additional validation, be useful for supporting efficacy, and have the potential to be used to evaluate efficacy in early phase trials such as proof-of-concept and dose-ranging trials. In NASH, a combination of hepatic triglyceride quantification, liver enzymes and CK 18 are biologically plausible markers of improvement that are also objective, measurable and sensitive to change. Resolution of steatohepatitis almost never occurs without a decrease in hepatic steatosis. Moreover, serum CK18 levels, which are reflective of apoptotic activity, corresponded well to an improvement in liver

histology in two phase 2b clinical trials of NASH in adults and in children respectively but need additional validation (77, 78). This additional validation can best be achieved by incorporating these biomarkers as secondary endpoints in early phase clinical trials.

6. Identification of the population at risk in early stage disease (Table 4)

In studies of early stage NASH, it is important to study a relatively homogeneous population on the one hand, but the data must also be generalizable to the larger histological spectrum of NASH on the other. As noted above, more research is needed to be able to identify a high risk population among those with early stage disease.

The NASH Clinical Research Network classification distinguishes between definite and borderline NASH but it is not clear if these phenotypes imply different natural courses and outcomes to therapy (31). Data were presented at the workshop suggesting that a diagnosis of steatohepatitis based on presence of steatosis, inflammation and either ballooning or pericellular fibrosis has been shown to correlate with outcomes.(79). Also, if both patients with borderline and definite NASH are included in trials, appropriate statistical methods, for example, stratification of randomization could be used to account for these histological variants of steatohepatitis as variables that may impact outcomes with a given therapy.

Several small studies of subjects with multiple biopsies indicate that the disease progression is bidirectional, with some subjects regressing and others progressing (29, 30). Moreover, in view of the sampling variability associated with liver biopsies (80), there is confusion about whether it is feasible to identify and quantify disease progression within a 3–4 year time frame.

While the clinical implications of progression from stage 1 to 2 disease are unclear, there is general agreement in the literature that progression to advanced fibrosis or cirrhosis represents a clear worsening of the health status of the patient (43). Recently, the NIDDK NASH CRN reported the risk of progression to advanced fibrosis in 239 subjects within a 3–4 year time frame for subjects with early stage disease (81) These data demonstrated that the metabolic syndrome (but not type 2 diabetes alone), persistently elevated ALT, higher baseline inflammation scores, Mallory bodies and portal/periportal fibrosis were independent predictors of the risk of progression. Of note, over 20% of subjects with stage 1b or stage 2 disease progressed to advanced fibrosis. These data provide guidance on enrichment strategies for trial populations with early stage NASH.

7. Pathways for treatment development for advanced stage (stage 3–4) disease

There are two potential targets for therapy in those with advanced stage disease. These include therapies directed against the steatohepatitis (the abnormality that drives disease progression) or fibrosis, which is the consequence of disease pathogenesis that leads to clinical events.

Arguably, those with NASH and advanced stage disease are at greatest risk of adverse liver-related outcomes within 3–4 years. Trials for therapeutics directed against fibrosis would generally need to include patients with NASH and bridging fibrosis or cirrhosis (Table 4).

Presentations at the workshop supported the contention that the NAFLD fibrosis score and FIB4 (based on age, AST, ALT and platelet count) score have been shown to predict mortality and liver-related outcomes in subjects with NAFLD (82, 83), which makes these markers of interest for potential enrichment strategies. A MELD score has been clinically considered to reflect overall liver health, with a score > 10 associated with increased risk of liver-related outcomes in those with otherwise compensated cirrhosis (44). In trials in patients with advanced fibrosis or cirrhosis, inclusion of those with advanced fibrosis and a HVPG > 10 mm Hg might also serve as an approach to enrich the population for higher rates of outcome events, thereby maximizing the ability to show a difference between those on treatment and placebo.

8. Other Considerations in Trial Design and Endpoints

Early phase clinical trials that are attempting to provide evidence of proof-of-concept, or to identify appropriate doses, often utilize endpoints such as transaminases or other noninvasive biomarkers of disease. Toxicity merits particular attention in these trials.

In phase 2b and 3 trials, surrogate measures closely linked to liver-related outcomes may be considered. One such potential surrogate is the proportion of subjects with an increase in HVPG to values > 10 mm Hg. This would necessitate inclusion of subjects with advanced fibrosis and an HVPG between 6–10 mm Hg. In studies that will measure improvement in clinical liver-related outcomes, the ability to demonstrate a benefit may be maximized by including those with compensated cirrhosis and an HVPG > 10 mm Hg and/or a MELD score > 10. Both FIB4 and the NAFLD fibrosis score may also facilitate enrichment for those populations who are more likely to have outcomes. They have the added advantages of being easy to compute, and are widely available.

It is important to remember that purely anti-fibrotic drugs used to treat advanced stage NASH do not affect steatohepatitis, which is thought to be the driver of disease progression. Therefore even if an anti-fibrotic is effective, efforts to directly attenuate underlying disease pathogenesis must be considered. The nature of such ‘maintenance therapy’ is not yet defined and could include combinations of agents directed against steatohepatitis, in addition to, or instead of anti-fibrotics once some fibrosis regression has been achieved. Disease progression or regression is not simply reflective of fibrosis alone, however, and long-term studies will be necessary to demonstrate sustained improvement in outcomes, including a beneficial impact of fibrosis regression in reducing the risks of decompensation or hepatocellular carcinoma.

9. Biomarker and Diagnostics Development for NAFLD

At present, a liver biopsy is required for both the clinical diagnosis of NASH and assessment of the treatment response. Liver biopsies are invasive, painful, subject to sampling variability and occasionally associated with serious complications. Therefore, there is an urgent unmet

need to develop biomarkers that facilitate the diagnosis, identification of populations at risk, assessment of disease progression or regression, and/or response to treatment.

There are multiple FDA Guidances related to biomarker development, biomarker implications for clinical trial designs, qualification of drug development tools, and considerations for using a diagnostic as part of a drug development program. Please refer to the FDA guidance webpage[2] for the following biomarker-related Guidances:

- Qualification Process for Drug Development Tools
- Clinical Pharmacogenomics: Premarketing Evaluation in Early Phase Clinical Studies
- *In vitro* Companion Diagnostic Devices
- Standards for Clinical Trial Imaging Endpoints
- Clinical Trial Designs Employing Enrichment Strategies to Support Approval of Human Drugs and Biological Products

In addition, FDA has a web navigational tool informing investigators about IND procedures[3] Studies assessing biomarkers should have clearly defined objectives, particularly with respect to how a biomarker is to be used to aid a drug development program. The most successful programs are those where the disease pathogenesis and the natural history of the disease are well understood. Biomarkers used as endpoints in clinical trials supporting drug approval should be validated to predict clinically meaningful outcomes. Furthermore, biomarkers for treatment response must be validated for individual drug classes (e.g., antifibrotics) used to treat NASH. Since biomarker development can be difficult for individual companies to pursue, especially in the setting of rare diseases, the creation of consortia of relevant stakeholders (e.g., Pharma, academia, NIH and other government entities, professional societies, and patient advocacy groups) can be most beneficial to share resources, clinical trial data, and patient samples. As a biomarker development program progresses, there are several pathways to enable continued dialogue with FDA and we encourage such exchanges to promote efficient development of effective biomarkers.

10. Pediatric populations with NAFLD and development of therapeutics for such populations

There is a substantial burden of disease due to NAFLD in the pediatric population (6, 84–86) but there are also special challenges in drug development for children. Children with NAFLD often have hepatic histology that is different from that seen in adults (33). However, given the long natural history of the disease before clinical outcomes occur, many children are no longer under the care of a pediatrician when such outcomes develop. The impacts of the physiological and behavioral changes associated with growth and sexual maturation on either the disease phenotype or progression have not been defined. Children also represent a

[2] www.fda.gov/Drugs/Guidance ComplianceRegulatoryInformation/Guidances/default
[3] http://www.fda.gov/Drugs/DevelopmentApprovalProcess/HowDrugsareDevelopedandApproved/ApprovalApplications/InvestigationalNewDrugINDApplication/ucm343349.htm

vulnerable population who mandate greater attention to potential toxicity, and specific studies may be required to determine pharmacokinetics and to optimize dosing for drug development in this population. Trials in children must comply with the standards as set out in 21 CFR 50.52, which state that “investigations involving greater than minimal risk to pediatric patients must present the prospect of direct benefit to individual subjects”. Initial pediatric studies should focus on the optimal dosing strategy before clinical trials for efficacy are planned. There is no clear consensus on the optimal pathway for drug development in children, which highlights the need for continued dialogue and discussion to clarify and accelerate drug development pathways for NASH in children.

11. Safety related issues in drug development for NASH

It is imperative to be vigilant about the potential for drug-induced liver injury in all stages of drug/biological development in NAFLD. NASH is associated with type II diabetes, increased cardiovascular risk and cancer-related mortality (12, 13, 87). For this reason, it seems important to monitor LDL- and HDL-cholesterol, triglycerides, and diabetes control (e.g. hemoglobin A_{1C}) in phase 2b and 3 NASH trials. Those engaged in drug development for NASH are also encouraged to follow FDA guidance related to assessment of safety such as those listed below:

Investigator Responsibilities — Protecting the Rights, Safety, and Welfare of Study Subjects at http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM187772.pdf

AND

Establishment and Operation of Clinical Trial Data Monitoring Committees at http://www.fda.gov/downloads/RegulatoryInformation/Guidances/ucm127073.pdf

Acknowledgments

The authors gratefully acknowledge the following individuals who provided thoughtful critique and comments on the multiple drafts of the manuscript: Dr. Robert Temple (FDA), Dr. Julie Bietz (FDA), Dr. Donna Griebel (FDA), Dr. Andrew Mulberg (FDA), Dr. Christopher Leptak (FDA) and Ashley Slagle PhD (SEALD, FDA).

Grant Support: This work was supported by the Food and Drug Administration of the US Federal Government and by the American Association for Study of Liver Diseases.

List of Abbreviations

NAFLD	nonalcoholic fatty liver disease
NASH	nonalcoholic steatohepatitis
NAFL	nonalcoholic fatty liver
HVPG	hepatic venous pressure gradient
MELD	model for end stage liver disease
T2DM	type 2 diabetes mellitus

METS	metabolic syndrome
FDA	Food and Drug Administration
AASLD	American Association for Study of Liver Disease

References

1. Loomba R, Sanyal AJ. The global NAFLD epidemic. Nat Rev Gastroenterol Hepatol. 2013; 10:686–690. [PubMed: 24042449]
2. Ahmed MH, Abu EO, Byrne CD. Non-Alcoholic Fatty Liver Disease (NAFLD): new challenge for general practitioners and important burden for health authorities? Prim Care Diabetes. 4:129–137. [PubMed: 20299294]
3. Neuschwander-Tetri BA, Clark JM, Bass NM, Van Natta ML, Unalp-Arida A, Tonascia J, Zein CO, et al. Clinical, laboratory and histological associations in adults with nonalcoholic fatty liver disease. Hepatology. 2010; 52:913–924. [PubMed: 20648476]
4. Brunt EM. Pathology of nonalcoholic fatty liver disease. Nat Rev Gastroenterol Hepatol. 2010; 7:195–203. [PubMed: 20195271]
5. Browning JD, Szczepaniak LS, Dobbins R, Nuremberg P, Horton JD, Cohen JC, Grundy SM, et al. Prevalence of hepatic steatosis in an urban population in the United States: impact of ethnicity. Hepatology. 2004; 40:1387–1395. [PubMed: 15565570]
6. Patton HM, Sirlin C, Behling C, Middleton M, Schwimmer JB, Lavine JE. Pediatric nonalcoholic fatty liver disease: a critical appraisal of current data and implications for future research. J Pediatr Gastroenterol Nutr. 2006; 43:413–427. [PubMed: 17033514]
7. Denzer C, Thiere D, Muche R, Koenig W, Mayer H, Kratzer W, Wabitsch M. Gender-specific prevalences of fatty liver in obese children and adolescents: roles of body fat distribution, sex steroids, and insulin resistance. J Clin Endocrinol Metab. 2009; 94:3872–3881. [PubMed: 19773396]
8. Bellentani S, Scaglioni F, Marino M, Bedogni G. Epidemiology of non-alcoholic fatty liver disease. Dig Dis. 2010; 28:155–161. [PubMed: 20460905]
9. Wanless IR, Lentz JS. Fatty liver hepatitis (steatohepatitis) and obesity: an autopsy study with analysis of risk factors. Hepatology. 1990; 12:1106–1110. [PubMed: 2227807]
10. Adams LA, Lymp JF, St Sauver J, Sanderson SO, Lindor KD, Feldstein A, Angulo P. The natural history of nonalcoholic fatty liver disease: a population-based cohort study. Gastroenterology. 2005; 129:113–121. [PubMed: 16012941]
11. Rafiq N, Bai C, Fang Y, Srishord M, McCullough A, Gramlich T, Younossi ZM. Long-term follow-up of patients with nonalcoholic fatty liver. Clin Gastroenterol Hepatol. 2009; 7:234–238. [PubMed: 19049831]
12. Treeprasertsuk S, Leverage S, Adams LA, Lindor KD, St Sauver J, Angulo P. The Framingham risk score and heart disease in nonalcoholic fatty liver disease. Liver Int. 2012; 32:945–950. [PubMed: 22299674]
13. Adams LA, Harmsen S, St Sauver JL, Charatcharoenwitthaya P, Enders FB, Therneau T, Angulo P. Nonalcoholic fatty liver disease increases risk of death among patients with diabetes: a community-based cohort study. Am J Gastroenterol. 2010; 105:1567–1573. [PubMed: 20145609]
14. Stepanova M, Rafiq N, Makhlouf H, Agrawal R, Kaur I, Younoszai Z, McCullough A, et al. Predictors of all-cause mortality and liver-related mortality in patients with non-alcoholic fatty liver disease (NAFLD). Dig Dis Sci. 2013; 58:3017–3023. [PubMed: 23775317]
15. Lattuada G, Ragogna F, Perseghin G. Why does NAFLD predict type 2 diabetes? Curr Diab Rep. 2011; 11:167–172. [PubMed: 21431854]
16. Manchanayake J, Chitturi S, Nolan C, Farrell GC. Postprandial hyperinsulinemia is universal in non-diabetic patients with nonalcoholic fatty liver disease. J Gastroenterol Hepatol. 2011; 26:510–516. [PubMed: 21155882]

17. Speliotes EK, Massaro JM, Hoffmann U, Vasan RS, Meigs JB, Sahani DV, Hirschhorn JN, et al. Fatty liver is associated with dyslipidemia and dysglycemia independent of visceral fat: the Framingham Heart Study. Hepatology. 2010; 51:1979–1987. [PubMed: 20336705]
18. Kasturiratne A, Weerasinghe S, Dassanayake AS, Rajindrajith S, de Silva AP, Kato N, Wickremasinghe AR, et al. Influence of non-alcoholic fatty liver disease on the development of diabetes mellitus. J Gastroenterol Hepatol. 2013; 28:142–147. [PubMed: 22989165]
19. Adams LA, Waters OR, Knuiman MW, Elliott RR, Olynyk JK. NAFLD as a risk factor for the development of diabetes and the metabolic syndrome: an eleven-year follow-up study. Am J Gastroenterol. 2009; 104:861–867. [PubMed: 19293782]
20. Anstee QM, Targher G, Day CP. Progression of NAFLD to diabetes mellitus, cardiovascular disease or cirrhosis. Nat Rev Gastroenterol Hepatol. 2013; 10:330–344. [PubMed: 23507799]
21. Charlton MR, Burns JM, Pedersen RA, Watt KD, Heimbach JK, Dierkhising RA. Frequency and outcomes of liver transplantation for nonalcoholic steatohepatitis in the United States. Gastroenterology. 2011; 141:1249–1253. [PubMed: 21726509]
22. Wong RJ, Cheung R, Ahmed A. Nonalcoholic steatohepatitis is the most rapidly growing indication for liver transplantation in patients with hepatocellular carcinoma in the U.S. Hepatology. 2013
23. Kemmer N, Neff GW, Franco E, Osman-Mohammed H, Leone J, Parkinson E, Cece E, et al. Nonalcoholic fatty liver disease epidemic and its implications for liver transplantation. Transplantation. 2013; 96:860–862. [PubMed: 24247899]
24. Ludwig J, Viggiano TR, McGill DB, Oh BJ. Nonalcoholic steatohepatitis: Mayo Clinic experiences with a hitherto unnamed disease. Mayo Clin Proc. 1980; 55:434–438. [PubMed: 7382552]
25. Chalasani N, Younossi Z, Lavine JE, Diehl AM, Brunt EM, Cusi K, Charlton M, et al. The diagnosis and management of non-alcoholic fatty liver disease: practice guideline by the American Gastroenterological Association, American Association for the Study of Liver Diseases, and American College of Gastroenterology. Gastroenterology. 2012; 142:1592–1609. [PubMed: 22656328]
26. Dunn W, Schwimmer JB. The obesity epidemic and nonalcoholic fatty liver disease in children. Curr Gastroenterol Rep. 2008; 10:67–72. [PubMed: 18417045]
27. Patton HM, Lavine JE, Van Natta ML, Schwimmer JB, Kleiner D, Molleston J. Clinical correlates of histopathology in pediatric nonalcoholic steatohepatitis. Gastroenterology. 2008; 135:1961–1971 e1962. [PubMed: 19013463]
28. Mofrad P, Contos MJ, Haque M, Sargeant C, Fisher RA, Luketic VA, Sterling RK, et al. Clinical and histologic spectrum of nonalcoholic fatty liver disease associated with normal ALT values. Hepatology. 2003; 37:1286–1292. [PubMed: 12774006]
29. Adams LA, Sanderson S, Lindor KD, Angulo P. The histological course of nonalcoholic fatty liver disease: a longitudinal study of 103 patients with sequential liver biopsies. J Hepatol. 2005; 42:132–138. [PubMed: 15629518]
30. Ekstedt M, Franzen LE, Mathiesen UL, Thorelius L, Holmqvist M, Bodemar G, Kechagias S. Long-term follow-up of patients with NAFLD and elevated liver enzymes. Hepatology. 2006; 44:865–873. [PubMed: 17006923]
31. Kleiner DE, Brunt EM, Van Natta M, Behling C, Contos MJ, Cummings OW, Ferrell LD, et al. Design and validation of a histological scoring system for nonalcoholic fatty liver disease. Hepatology. 2005; 41:1313–1321. [PubMed: 15915461]
32. Brunt EM, Janney CG, Di Bisceglie AM, Neuschwander-Tetri BA, Bacon BR. Nonalcoholic steatohepatitis: a proposal for grading and staging the histological lesions. Am J Gastroenterol. 1999; 94:2467–2474. [PubMed: 10484010]
33. Schwimmer JB, Behling C, Newbury R, Deutsch R, Nievergelt C, Schork NJ, Lavine JE. Histopathology of pediatric nonalcoholic fatty liver disease. Hepatology. 2005; 42:641–649. [PubMed: 16116629]
34. Caldwell SH, Hespenheide EE. Subacute liver failure in obese women. Am J Gastroenterol. 2002; 97:2058–2062. [PubMed: 12190177]

35. Kistler KD, Molleston J, Unalp A, Abrams SH, Behling C, Schwimmer JB. Symptoms and quality of life in obese children and adolescents with non-alcoholic fatty liver disease. Aliment Pharmacol Ther. 31:396–406. [PubMed: 19863497]
36. David K, Kowdley KV, Unalp A, Kanwal F, Brunt EM, Schwimmer JB. Quality of life in adults with nonalcoholic fatty liver disease: baseline data from the nonalcoholic steatohepatitis clinical research network. Hepatology. 2009; 49:1904–1912. [PubMed: 19434741]
37. Dan AA, Kallman JB, Wheeler A, Younoszai Z, Collantes R, Bondini S, Gerber L, et al. Health-related quality of life in patients with non-alcoholic fatty liver disease. Aliment Pharmacol Ther. 2007; 26:815–820. [PubMed: 17767465]
38. Younossi ZM, Boparai N, Price LL, Kiwi ML, McCormick M, Guyatt G. Health-related quality of life in chronic liver disease: the impact of type and severity of disease. Am J Gastroenterol. 2001; 96:2199–2205. [PubMed: 11467653]
39. Elwing JE, Lustman PJ, Wang HL, Clouse RE. Depression, anxiety, and nonalcoholic steatohepatitis. Psychosom Med. 2006; 68:563–569. [PubMed: 16868265]
40. Bajaj JS, Wade JB, Gibson DP, Heuman DM, Thacker LR, Sterling RK, Stravitz RT, et al. The multi-dimensional burden of cirrhosis and hepatic encephalopathy on patients and caregivers. Am J Gastroenterol. 2011; 106:1646–1653. [PubMed: 21556040]
41. Gines P, Quintero E, Arroyo V, Teres J, Bruguera M, Rimola A, Caballeria J, et al. Compensated cirrhosis: natural history and prognostic factors. Hepatology. 1987; 7:122–128. [PubMed: 3804191]
42. D'Amico G, Garcia-Tsao G, Pagliaro L. Natural history and prognostic indicators of survival in cirrhosis: a systematic review of 118 studies. J Hepatol. 2006; 44:217–231. [PubMed: 16298014]
43. Sanyal AJ, Banas C, Sargeant C, Luketic VA, Sterling RK, Stravitz RT, Shiffman ML, et al. Similarities and differences in outcomes of cirrhosis due to nonalcoholic steatohepatitis and hepatitis C. Hepatology. 2006; 43:682–689. [PubMed: 16502396]
44. Ripoll C, Groszmann R, Garcia-Tsao G, Grace N, Burroughs A, Planas R, Escorsell A, et al. Hepatic venous pressure gradient predicts clinical decompensation in patients with compensated cirrhosis. Gastroenterology. 2007; 133:481–488. [PubMed: 17681169]
45. Baumeister SE, Volzke H, Marschall P, John U, Schmidt CO, Flessa S, Alte D. Impact of fatty liver disease on health care utilization and costs in a general population: a 5-year observation. Gastroenterology. 2008; 134:85–94. [PubMed: 18005961]
46. Bhala N, Angulo P, van der Poorten D, Lee E, Hui JM, Saracco G, Adams LA, et al. The natural history of nonalcoholic fatty liver disease with advanced fibrosis or cirrhosis: an international collaborative study. Hepatology. 2011; 54:1208–1216. [PubMed: 21688282]
47. Sanyal A, Poklepovic A, Moyneur E, Barghout V. Population-based risk factors and resource utilization for HCC: US perspective. Curr Med Res Opin. 2010; 26:2183–2191. [PubMed: 20666689]
48. Matteoni CA, Younossi ZM, Gramlich T, Boparai N, Liu YC, McCullough AJ. Nonalcoholic fatty liver disease: a spectrum of clinical and pathological severity. Gastroenterology. 1999; 116:1413–1419. [PubMed: 10348825]
49. Ascha MS, Hanouneh IA, Lopez R, Tamimi TA, Feldstein AF, Zein NN. The incidence and risk factors of hepatocellular carcinoma in patients with nonalcoholic steatohepatitis. Hepatology. 2010; 51:1972–1978. [PubMed: 20209604]
50. Powell EE, Cooksley WG, Hanson R, Searle J, Halliday JW, Powell LW. The natural history of nonalcoholic steatohepatitis: a follow-up study of forty-two patients for up to 21 years. Hepatology. 1990; 11:74–80. [PubMed: 2295475]
51. Brunt E, Kleiner D, Wilson L, Unalp-Arida A, Neuschwander-Tetri B. Histological predictors of improvement of NASH: results from NASH Clinical Research Network PIVENS trial. Hepatology. 2012 Oct. A xxx.
52. Sanyal AJ, Chalasani N, Kowdley KV, McCullough A, Diehl AM, Bass NM, Neuschwander-Tetri BA, et al. Pioglitazone, vitamin E, or placebo for nonalcoholic steatohepatitis. N Engl J Med. 2010; 362:1675–1685. [PubMed: 20427778]

53. Marchesini G, Bianchi G, Amodio P, Salerno F, Merli M, Panella C, Loguercio C, et al. Factors associated with poor health-related quality of life of patients with cirrhosis. Gastroenterology. 2001; 120:170–178. [PubMed: 11208726]
54. Loria A, Escheik C, Gerber NL, Younossi ZM. Quality of life in cirrhosis. Curr Gastroenterol Rep. 2013; 15:301. [PubMed: 23250701]
55. Huang Y, de Boer WB, Adams LA, MacQuillan G, Bulsara MK, Jeffrey GP. Image analysis of liver biopsy samples measures fibrosis and predicts clinical outcome. J Hepatol. 2014; 61:22–27. [PubMed: 24607623]
56. Everhart JE, Wright EC, Goodman ZD, Dienstag JL, Hoefs JC, Kleiner DE, Ghany MG, et al. Prognostic value of Ishak fibrosis stage: findings from the hepatitis C antiviral long-term treatment against cirrhosis trial. Hepatology. 2010; 51:585–594. [PubMed: 20101752]
57. Singh S, Fujii LL, Murad MH, Wang Z, Asrani SK, Ehman RL, Kamath PS, et al. Liver stiffness is associated with risk of decompensation, liver cancer, and death in patients with chronic liver diseases: a systematic review and meta-analysis. Clin Gastroenterol Hepatol. 2013; 11:1573–1584. e1571–1572. quiz e1588–1579. [PubMed: 23954643]
58. Child, CG.; Turcotte, J. The liver and portal hypertension. In: Child, CG., editor. Surgery and Portal Hypertension. Philadelphia: Saunders; 1964. p. 50-58.
59. Durand F, Valla D. Assessment of prognosis of cirrhosis. Semin Liver Dis. 2008; 28:110–122. [PubMed: 18293281]
60. Kamath PS, Wiesner RH, Malinchoc M, Kremers W, Therneau TM, Kosberg CL, D'Amico G, et al. A model to predict survival in patients with end-stage liver disease. Hepatology. 2001; 33:464–470. [PubMed: 11172350]
61. Wiesner R, Edwards E, Freeman R, Harper A, Kim R, Kamath P, Kremers W, et al. Model for end-stage liver disease (MELD) and allocation of donor livers. Gastroenterology. 2003; 124:91–96. [PubMed: 12512033]
62. Angulo P, Lindor KD, Therneau TM, Jorgensen RA, Malinchoc M, Kamath PS, Dickson ER. Utilization of the Mayo risk score in patients with primary biliary cirrhosis receiving ursodeoxycholic acid. Liver. 1999; 19:115–121. [PubMed: 10220741]
63. Dickson ER, Grambsch PM, Fleming TR, Fisher LD, Langworthy A. Prognosis in primary biliary cirrhosis: model for decision making. Hepatology. 1989; 10:1–7. [PubMed: 2737595]
64. Grambsch PM, Dickson ER, Kaplan M, LeSage G, Fleming TR, Langworthy AL. Extramural cross-validation of the Mayo primary biliary cirrhosis survival model establishes its generalizability. Hepatology. 1989; 10:846–850. [PubMed: 2680867]
65. Malinchoc M, Kamath PS, Gordon FD, Peine CJ, Rank J, ter Borg PC. A model to predict poor survival in patients undergoing transjugular intrahepatic portosystemic shunts. Hepatology. 2000; 31:864–871. [PubMed: 10733541]
66. Merion RM, Schaubel DE, Dykstra DM, Freeman RB, Port FK, Wolfe RA. The survival benefit of liver transplantation. Am J Transplant. 2005; 5:307–313. [PubMed: 15643990]
67. Al Sibae MR, Cappell MS. Accuracy of MELD scores in predicting mortality in decompensated cirrhosis from variceal bleeding, hepatorenal syndrome, alcoholic hepatitis, or acute liver failure as well as mortality after non-transplant surgery or TIPS. Dig Dis Sci. 2011; 56:977–987. [PubMed: 20844956]
68. Ripoll C, Banares R, Rincon D, Catalina MV, Lo Iacono O, Salcedo M, Clemente G, et al. Influence of hepatic venous pressure gradient on the prediction of survival of patients with cirrhosis in the MELD Era. Hepatology. 2005; 42:793–801. [PubMed: 16175621]
69. Merion RM. When is a patient too well and when is a patient too sick for a liver transplant? Liver Transpl. 2004; 10:S69–73. [PubMed: 15382215]
70. Groszmann RJ, Wongcharatrawee S. The hepatic venous pressure gradient: anything worth doing should be done right. Hepatology. 2004; 39:280–282. [PubMed: 14767976]
71. Zipprich A, Winkler M, Seufferlein T, Dollinger MM. Comparison of balloon vs. straight catheter for the measurement of portal hypertension. Aliment Pharmacol Ther. 2010; 32:1351–1356. [PubMed: 21050237]
72. Atterbury CE, Groszmann RJ. Portal venous pressure measurement and its clinical significance. Lab Res Methods Biol Med. 1983; 7:567–585. [PubMed: 6353113]

73. Zipprich A, Garcia-Tsao G, Rogowski S, Fleig WE, Seufferlein T, Dollinger MM. Prognostic indicators of survival in patients with compensated and decompensated cirrhosis. Liver Int. 2012; 32:1407–1414. [PubMed: 22679906]
74. Bosch J, Garcia-Pagan JC. Prevention of variceal rebleeding. Lancet. 2003; 361:952–954. [PubMed: 12648985]
75. Moitinho E, Escorsell A, Bandi JC, Salmeron JM, Garcia-Pagan JC, Rodes J, Bosch J. Prognostic value of early measurements of portal pressure in acute variceal bleeding. Gastroenterology. 1999; 117:626–631. [PubMed: 10464138]
76. Ripoll C, Groszmann RJ, Garcia-Tsao G, Bosch J, Grace N, Burroughs A, Planas R, et al. Hepatic venous pressure gradient predicts development of hepatocellular carcinoma independently of severity of cirrhosis. J Hepatol. 2009; 50:923–928. [PubMed: 19303163]
77. Jain AK, Ross B, Yates K, Comerford M, Masuoka H, Molleston JP, Schwimmer JB, Lavine JE, Brunt EM, Tonascia J, Kleiner D, Chalasani N. Serum keratin fgragment 18 (CK18) levels significantly predict changes in liver histology in children and adolescents with nonalcoholic fatty liver disease: results from teh TONIC trial. Hepatology. 2013; 58:A114.
78. Vuppulanchi R, Deppe RB, Yates K, Unalp A, Kleiner D. Changes in Serum Cytokeratin 18 Levels Significantly Predict Changes in Liver Histology in Adults With Nonalcoholic Steatohepatitis: Results From the Pivens Trial. Gastroenterology. 2013; 144:S951.
79. Younossi ZM, Stepanova M, Rafiq N, Makhlouf H, Younoszai Z, Agrawal R, Goodman Z. Pathologic criteria for nonalcoholic steatohepatitis: interprotocol agreement and ability to predict liver-related mortality. Hepatology. 2011; 53:1874–1882. [PubMed: 21360720]
80. Ratziu V, Charlotte F, Heurtier A, Gombert S, Giral P, Bruckert E, Grimaldi A, et al. Sampling variability of liver biopsy in nonalcoholic fatty liver disease. Gastroenterology. 2005; 128:1898–1906. [PubMed: 15940625]
81. Brunt EM, Belt PH, Wilson L, Guy CD, Yeh MM, Kowdley KV, Sanyal AJ, Neuschwander-Tetri B, Kleiner DE. Progression to bridging fibrosis in nonalcoholic fatty liver disease over 4 years in the NASH CRN. Hepatology. 2013; 58:495A.
82. Angulo P, Bugianesi E, Bjornsson ES, Charatcharoenwitthaya P, Mills PR, Barrera F, Haflidadottir S, et al. Simple noninvasive systems predict long-term outcomes of patients with nonalcoholic fatty liver disease. Gastroenterology. 2013; 145:782–789 e784. [PubMed: 23860502]
83. Kim D, Kim WR, Kim HJ, Therneau TM. Association between noninvasive fibrosis markers and mortality among adults with nonalcoholic fatty liver disease in the United States. Hepatology. 2013; 57:1357–1365. [PubMed: 23175136]
84. Patton HM, Yates K, Unalp-Arida A, Behling CA, Huang TT, Rosenthal P, Sanyal AJ, et al. Association between metabolic syndrome and liver histology among children with nonalcoholic Fatty liver disease. Am J Gastroenterol. 2010; 105:2093–2102. [PubMed: 20372110]
85. Alisi A, Manco M, Vania A, Nobili V. Pediatric nonalcoholic fatty liver disease in 2009. J Pediatr. 2009; 155:469–474. [PubMed: 19772998]
86. Koebnick C, Getahun D, Reynolds K, Coleman KJ, Porter AH, Lawrence JM, Punyanitya M, et al. Trends in nonalcoholic fatty liver disease-related hospitalizations in US children, adolescents, and young adults. J Pediatr Gastroenterol Nutr. 2009; 48:597–603. [PubMed: 19412009]
87. Dunn W, Xu R, Wingard DL, Rogers C, Angulo P, Younossi ZM, Schwimmer JB. Suspected nonalcoholic fatty liver disease and mortality risk in a population-based cohort study. Am J Gastroenterol. 2008; 103:2263–2271. [PubMed: 18684196]

SUMMARY

The AASLD-FDA workshop discussed several elements in the development of diagnostics and therapeutics for NASH and will hopefully accelerate drug development for the disease. While there were discussions on several important issues, a full consensus was not reached, for example, with respect to specific pathways for validation of biomarkers. These uncertainties underscore the need for continuing dialogue between all the stakeholders in this arena to identify gaps in knowledge and unmet needs, and to address areas of ambiguity in development of diagnostics and therapies against NASH.

Table 1

Clinically meaningful outcomes in patients with nonalcoholic steatohepatitis

Outcome
Hard clinical outcomes Mortality: - All-cause - Liver-related Liver-related outcomes that drive mortality: - Ascites and its complications - Variceal hemorrhage - Hepatic encephalopathy - Hepatocellular cancer - Acute on chronic liver failure
How a patient functions? - Functional status (disability) - Days of work missed - Ability to manage activities of daily living
How a patient feels? - Symptoms - Physical health - Mental health

Table 2

Surrogate markers of the risk of mortality

Endpoint	Comment	Utility
Death	Strongest endpoint- but sample size and study duration will need to be very large	impractical
MELD score	Score of 14 identifies a point above which in absence of transplant, survival declines	Objective measure Validated
2 point CTP	Transition from Child A to B is clearly associated with poorer survival	Objective-subjective Validated Suffer from ceiling and floor effects
HVPG	Tracks risks of complications and progression	Objective measure Validated
Composite: Ascites Variceal bleed Encephalopathy HCC	**1** Strongly associated with mortality **2** It is quantifiable **3** Rates of development in controls are known	Objective-subjective Validated
Quantitative liver function tests	Quantitative – give an example	Needs more validation

Table 3

Endpoints (markers of outcomes) for early stage disease

Endpoint	Comment
Development of cirrhosis	• Clearly defines a worsening of health status and adversely affects how a patient feels, functions or survives • May serve as a primary endpoint for full approval • May be measured by biopsy or other modalities for the diagnosis of advanced fibrosis/cirrhosis (e.g., liver stiffness measurements)
Reversal of steatohepatitis without progression to advanced fibrosis (stage 3–4)	• Can be demonstrated within a 12–24 month time-frame • Is a surrogate for prevention of cirrhosis • May be suitable for subpart H approval process but will require long-term post-approval follow up to demonstrate that treatment prevents cirrhosis • Advanced fibrosis/cirrhosis may be measured by biopsy or other modalities for the diagnosis of advanced fibrosis/cirrhosis (e.g., liver stiffness measurements)
NAFLD Activity Score (NAS)	• Needs validation to demonstrate that a decrease reflects reduced risk of advanced fibrosis/cirrhosis. • Relative impact of improvement of steatosis vs. inflammation vs. ballooning is not clear
Reduction of hepatic steatosis by imaging studies along with decrease in ALT and/or CK18	• MR quantification of steatosis is now a community-accepted validated tool: requires FDA acceptance of the tool for validation and use in clinical trials used for registration * • ALT usually, but not always, improves with improvement in histology • Decreased CK18 has been shown to correlate with histological improvement in trials of subjects with stage 0–3 disease • Best suited for early phase proof of concept (phase 1/2a) studies

* It is recommended that sponsors planning to use this tool as an endpoint discuss their plans with FDA in accordance with FDA mechanisms for pre-trial consultation.

Table 4

Populations to study in clinical trials of NASH

Population	Comment
For advanced (stage 3–4) disease (mainly anti-fibrotic therapies)	
Biopsy-based populations - NASH - Bridging fibrosis or cirrhosis	Biopsy-based populations: • Biopsy remains gold standard despite several limitations
Non-biopsy based populations: - Steatosis (by MR spectroscopy or diffusion-weighted MR) - Elevated ALT (> 40 IU/L) or CK 18> (270 u/l) - Advanced fibrosis/cirrhosis by FDA-approved non-invasive method (e.g., elastography) - Absence of alternate cause of liver disease	Non-biopsy based populations: • These represent alternate approaches to identification of NASH with advanced fibrosis. • It is recommended that sponsors discuss with FDA during pre-IND process about these entry criteria
Populations may be enriched for risk of clinical outcomes by inclusion of subjects with: - MELD > 10 - HVPG > 10 mm Hg - High NAFLD fibrosis score or FIB4 Additional considerations: - fibrosis-related SNPs - type 2 diabetes	Enrichment of populations: • Most useful when a liver-related clinical outcome or meeting minimal listing criteria for transplant (MELD of 15 or higher) is the primary endpoint • Fibrosis-related SNPs may explain variability in treatment response and could be used as an ancillary entry criteria • Type 2 diabetes affects clinical outcomes and should be accounted for by stratification or randomization
For early (stage 0–2) stage disease (anti-NASH therapies) - Nonalcoholic steatohepatitis (biopsy-proven definite or borderline steatohepatitis) - Grade 2 or greater inflammation - Presence of ballooning with Mallory bodies - Stage 1–2 fibrosis - Persistently elevated ALT over last 6 months - Metabolic syndrome	• This population has up to 20% probability of developing advanced fibrosis/cirrhosis within 4 years. • Should be feasible to demonstrate reversal of steatohepatitis without fibrosis progression to bridging or cirrhosis within 1–2 years.

Table 5

Recommendations about safety-related parameters to be measured in clinical trials for NASH

Parameter	Comment
Cardiovascular: • Low density lipoprotein cholesterol (LDL-C) • Small dense LDL (sdLDL) particle concentration, sdLDL cholesterol, % sdLDL • High density cholesterol (HDL-C) and subclass II and III HDL • Triglycerides and very low density lipoprotein particle size (VLDL-P) • ApoB • Lp(a) • Coronary calcification scores	• Main goal in phase 2b/3 trials is to demonstrate that these parameters do not move in a direction suggesting increased risk. • It is imperative that any drug developed for NASH be at least neutral from a cardiovascular risk perspective and ideally also reduce cardiovascular risks
Metabolic: • Hemoglobin A1C • Fasting insulin and glucose • Fasting free fatty acids	• Main goal is to demonstrate stability over the course of phase 2b/3 trials
Cancer: • Enumerate cancers	• Best studied in phase 4 post-marketing studies • Will be valuable to obtain family history and prior history of cancer to better understand impact of treatment on cancer incidence • All subjects should follow established practice guidelines for cancer screening during long-term trials
Other safety parameters: • Drug-induced liver injury • Behavioral adverse events (e.g., depression) • Other off-target unexpected effects	• Refer to FDA guidance on drug toxicity and criteria for stopping therapy in an individual patient versus trial stopping rules for safety concerns (need citation) • Depression scores should be tracked during therapy