

We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,900

Open access books available

186,000

International authors and editors

200M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities



WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com



Diagnosis and Characterization of Non-Alcoholic Fatty Liver Disease

Paula Iruzubieta, Marta González, Joaquín Cabezas,
María Teresa Arias-Loste and Javier Crespo

Additional information is available at the end of the chapter

<http://dx.doi.org/10.5772/intechopen.72668>

Abstract

Non-alcoholic fatty liver disease (NAFLD) can develop cirrhosis and even hepatocellular carcinoma, resulting in a high liver-related morbidity and mortality, being important to know those risk factors for disease progression, among which the presence of diabetes stands out. In addition, it is a disease with multisystemic behavior, becoming an independent risk factor for cardiovascular disease and extrahepatic tumors. Hence, early diagnosis and multidisciplinary management of NAFLD are really important. In this chapter, we will expose the different diagnostic and follow-up tools available for this disease, and with them we will make an algorithm according to the recommendations and the current evidence.

Keywords: NAFLD, biomarkers, transient elastography, multisystemic disease

1. Introduction

Non-alcoholic fatty liver disease (NAFLD) includes a wide spectrum of liver damage whose distinctive feature is the accumulation of intrahepatic fat, especially triglycerides, which cannot be attributed to secondary causes such as alcohol and certain drugs. NAFLD is nowadays considered to be the most common cause of chronic liver disease in western countries, showing a prevalence of around 30% in the general population [1]. Within NAFLD, two histological subtypes can be distinguished: (a) non-alcoholic fatty liver (NAFL), which includes patients with simple steatosis with or without mild inflammation and (b) non-alcoholic steatohepatitis (NASH), characterized by the presence of hepatic inflammation and hepatocyte injury (ballooning) with or without fibrosis [2, 3]. NAFL is a generally benign condition, and NASH is the progressive subtype that can lead to cirrhosis and hepatocellular carcinoma

(HCC) [4]. However, several studies with paired liver biopsies have demonstrated that both patients with NASH and those with NAFL have the potential to develop a progressive hepatic disease, and in this risk of progression there are some key factors such as diabetes mellitus [5, 6]. In general, patients with NAFLD have a higher long-term mortality than the general population, cardiovascular disease (CVD) being the principal cause of death, followed by different types of cancer [7–9] and liver-related complications, as well as the cardiovascular risk caused by the different factors of the metabolic syndrome, very frequent in this type of subjects; NAFLD is itself an independent risk factor for CVD [10]. Liver-related mortality is increased up to 10-fold in patients with NAFLD. In this sense, it should be emphasized that cirrhosis and HCC are the fifth most prevalent cause of mortality in the world. Therefore, given the hepatic and cardiovascular morbi-mortality generated by NAFLD, the early identification of these patients is important to provide suitable management that can lower the mortality for all causes.

2. Screening and diagnostic criteria

The mechanisms leading to the development and progression of NAFLD are not completely known, but it is widely accepted that the initial events are dependent on the development of obesity and insulin resistance (IR) [11]. For this reason, NAFLD has a strong association with the factors constituting the metabolic syndrome, the prevalence in this group of patients being considerably heightened. This relation is especially close in morbid obesity, where NAFLD is present in more than 90% of the cases, this condition taking the form of steatohepatitis in a third of the cases, while in up to 5–10% of the subjects, the liver disease has progressed to cirrhosis [12, 13]. The association between NAFLD and IR or diabetes mellitus type 2 (DM2) has also been clearly established [14]. It has been demonstrated that DM2 is associated with a greater hepatic content of triglycerides independently of the body mass index (BMI) [15, 16]. Thus, the prevalence of NAFLD in DM2 patients can reach up to 70% [11, 17, 18]. Moreover, both prediabetes (glucose intolerance and altered glucose when fasting) and DM2 are related to the severity of liver damage, the presence of steatohepatitis, fibrosis and even HCC [1, 19, 20]. Overall, 80% of the NAFLD cases present with some of the cardiovascular risk factors that constitute the metabolic syndrome (IR, obesity, dyslipidemia and arterial hypertension), and its prevalence directly increases the number of these factors that are present [21].

As a consequence of its high prevalence, especially in subjects with the abovementioned risk factors, its prognostic implications, and given that NAFLD is generally an asymptomatic disease, some authors recommend the implantation of an NAFLD-screening programme within the risk population [22, 23]. However, this topic is at present controversial given the great load on the national health systems that could be caused by these screening programmes and the lack of efficacious treatments currently available. In fact, the principal associations for the study of liver diseases (American Association for the Study of Liver Diseases (AASLD) and European Association for the Study of the Liver (EASL)) in their guidelines for clinical practice do not recommend this screening in any population [24] or they recommend it, with an A2 level of evidence, only for patients with DM2 independently of the levels of hepatic

enzymes [25]. To attempt to answer this question, valid cost-utility studies are necessary in screening programmes. There is no discussion, however, about the need to act when faced with a patient suspected of having NAFLD and not to underestimate its discovery due to the limited clinical and analytical repercussion manifested at first. In our opinion, patients with NAFLD and suspected to have advanced disease must be evaluated in specialist units for their correct characterization in case of a prompt availability of specific treatment.

2.1. Clinical and analytical manifestations

A diagnosis of NAFLD is very often reached through a casual analytical discovery during a health examination after an alteration in tests of liver function, or an alteration in hepatic morphology detected through an image study done with another objective, given that it is generally an asymptomatic disease. In the cases in which the patient reports symptoms, they are usually mild and unspecific, asthenia and abdominal problems being frequent, especially in the right hypochondria. The physical exploration may be normal or detect a soft, painless hepatomegaly, although occasionally it is difficult to evaluate as these patients very often present with central-type obesity, and in the patients with advanced fibrosis and cirrhosis, we may find signs of portal hypertension such as ascites, splenomegaly or jaundice [26].

Analytically, most of the patients present tests with normal or discretely altered liver function, with a predominance of ALT (alanine aminotransferase) compared to AST (aspartate aminotransferase). On specific occasions, a discrete elevation can be appreciated in the markers of cholestasis, especially GGT (gamma-glutamyl transpeptidase), which has been related to obesity and IR [27]. Another frequent analytical discovery is the elevation of the levels of ferritin in blood and of the transferrin saturation index without having demonstrated a corresponding increase in the deposits of hepatic iron [28]. Something similar occurs with the presence of elevated autoantibodies, which appear quite frequently in NAFLD and are considered an epiphenomenon [29].

2.2. Diagnosis of steatosis

Hepatic steatosis is defined histologically as the deposit of fat $\geq 5\%$ of the hepatocytes and is classified in four grades depending on the percentage of hepatocytes with steatotic vacuoles. The normal liver (S0) contains fat in less than 5% of the hepatocytes while grade 1 steatosis (S1) corresponds to less than 33% of the steatotic hepatocytes. In grade 2 and 3 steatosis, fat is present in at least 33 or 66% of the hepatocytes, respectively.

The presence of risk factors such as DM2, metabolic syndrome and obesity with the elevation of the hepatic enzymes, especially ALT, increases the possibility of fatty liver presenting. Nevertheless, although the ALT is a useful test, it is not valid for predicting the presence of this disease, or even the risk of progression, given that it can occur with normal hepatic enzymes [30]. In fact, in patients with DM2 and normal levels of ALT, a high prevalence of NAFL and NASH has been reported [31].

In clinical practice, ultrasound scan is a first-rate image technique if NAFLD is suspected due to its wide availability, low cost and safety [32]. The sensitivity of this technique is 93%

when the steatosis is greater than 33%; however, this sensitivity decreases considerably when the steatosis affects less than 30% of the hepatocytes [33, 34]. Steatosis can also be diagnosed through computerized tomography (CT), but its cost and the patient's exposure to radiation make its systematic use in long-term follow-up unadvisable in this pathology; moreover, its sensitivity does not improve substantially if the steatosis is mild [32]. Magnetic resonance imaging (MRI), including spectroscopy, can diagnose content levels of hepatic fat >5% and it is reliable to determine changes ($\geq 0.5\%$) in the grade of steatosis after weight loss. Although its use has widened in many studies, its use in clinical practice is limited by its cost and duration [35, 36].

The recently developed CAP (controlled attenuation parameter), an application of transient elastography (TE), which will be discussed later, available in the latest generation devices, enables the immediate and easy quantification of steatosis. CAP measures the degree of attenuation of the ultrasound wave transmitted through the liver, which is proportional to the amount of hepatic fat, and is less influenced by the sampling error than the liver biopsy, since it explores a liver volume approximately 100 times greater. Its values oscillate between 100 and 400 dB/m and it is possible to measure the liver stiffness used for the evaluation of fibrosis simultaneously. The studies published to date indicate that CAP is capable of diagnosing steatosis in chronic liver diseases of diverse causes even in mild stages (>10%) and has a good correlation with the degree of steatosis [37–44]. These studies show different cut-offs of CAP for the different grades of steatosis, but all of them demonstrate that the cut-offs do not differ among the different causes of liver diseases, in contrast with what happens with transient elastography [40]. In this sense, a recent meta-analysis including 2735 patients has established a series of cut-offs for the different grades of steatosis: 248 dB/m for S1, 268 dB/m for S2 and 280 dB/m for S3, with a sensitivity of 69, 77 and 88%, respectively, and a specificity of 82, 81 and 78%, respectively. In this meta-analysis, etiology, BMI and diabetes showed a significant influence on the value of the CAP, so the authors suggested the cut-offs established but subtracting 10 dB/m from the value of the CAP for the patients with NAFLD, 10 dB/m for diabetics, and subtracting/adding 4.4 dB/m for each unit of BMI above/below 25 kg/m² in the interval of 20–30 kg/m² [45].

Another image technique is magnetic resonance imaging-estimated proton density fat fraction (MRI-PDFF), which is based on chemical shift-based water fat separation methods. MRI-PDFF has shown good correlation with histology-determined steatosis grade in NAFLD patients, so it could be used for follow-up and treatment-response evaluation [46].

Lastly, within the non-invasive diagnosis of steatosis, various serological tests of biomarkers have been developed to predict the existence of hepatic fat (**Table 1**) [47–51]. However, all these biomarkers may be influenced by inflammation and fibrosis, and given that they do not provide great advantages compared to image techniques and routine analysis, their use in clinical practice is not widespread; even so, the Fatty Liver Index (FLI) that uses easily available parameters could be considered, although CAP has demonstrated better performance than this test for the diagnosis of grade 2–3 steatosis [44]. Nevertheless, FLI has been associated independently to liver-related mortality, as well as to the mortality rates due to

Indices	Formula	Cut-offs	Sensitivity (%)	Specificity (%)
Hepatic	$8 \times \text{ALT/AST ratio} + \text{BMI}$	30 (low cut-off)	93	40
Steatosis	(+2, if DM; +2, if female)	36 (high cut-off)	45	93
Index (HSI)				
Fatty liver	$\exp(n)/1 + \exp(n) \times 100$	10 (low cut-off)	95	29
Index (FLI)	$(n) = 0.953 \times \ln(\text{TG}) + 0.139 \times \text{BMI} + 0.718 \times \ln(\text{GGT}) + 0.053 \times \text{waist circumference} - 15.745$	60 (high cut-off)	44	91
SteatoTest	Proprietary formula	0.3 (low cut-off)	90	54
	(a2-macroglobulin, haptoglobin, apolipoprotein A1, GGT, bilirubin, ALT, cholesterol, triglycerides, glucose, BMI, age, gender)	0.7 (high cut-off)	46	88
NAFLD liver	$-2.89 + 1.18 \times \text{metabolic}$	-0.640	86	71
Fat score	Syndrome (yes: 1, no: 0) + $0.45 \times \text{Type 2 diabetes}$ (yes: 2, no: 0) + $0.15 \times \text{insulin} + 0.04 \times \text{AST} - 0.94 \times \text{AST/ALT}$			
Lipid	LAP (men) = Waist	20 (low cut-off)	99	16
accumulation	circumference - 65	80 (high cut-off)	43	94
product (LAP)	LAP (women) = Waist			
	circumference - 58			

Table 1. Indices for diagnosis of steatosis.

cardiovascular disease and cancer, but it seems that these associations are interfered by the risk conferred by the state of insulin resistance [52].

When steatosis is suspected in the aforementioned non-invasive methods, the liver biopsy is still the gold-standard method to conclusively diagnose NAFLD and the only one capable of distinguishing between NAFL and NASH, thus enabling the classification of the disease

according to the grade of activity (inflammation and hepatocyte injury) and the stage of fibrosis, the best predictors of the disease progression [53, 54]. The advantages and, especially, the drawbacks of the liver biopsy are dealt with later.

2.3. Initial diagnosis of NAFLD

From what has been mentioned so far, we can specify a series of characteristics that indicate a patient with NAFLD: (1) radiological evidence of steatosis or CAP >248 dB/m \pm Abnormal liver blood test, (2) the presence of insulin resistance or another component of the metabolic syndrome, (3) consumption of alcohol of <30 g/d in men and <20 g/d in women and (4) exclusion from other causes of chronic liver disease (viral hepatitis, cholestatic diseases, autoimmune hepatitis, hemochromatosis, $\alpha 1$ antitrypsin deficiency, Wilson's disease, drug-induced liver injury and celiac disease) [24, 25]. Once the initial diagnosis of NAFLD has been made, our next step is to evaluate the stage of disease and the necessity of carrying out a liver biopsy.

3. Diagnosis of steatohepatitis

A key element in the diagnosis of NAFLD is the differentiation of NASH from NAFL and the staging of the liver fibrosis, given that patients with NASH and advanced fibrosis are those at the greatest risk of developing hepatic complications and cardiovascular disease [54–56].

3.1. Liver biopsy

As it was mentioned earlier, the chosen method to evaluate the grade of histological lesion is still the liver biopsy. However, liver biopsy has well-known limitations and cannot be proposed for all patients, given the high prevalence of NAFLD worldwide. Liver biopsy is invasive and is not without complications. Besides, there are other drawbacks: (1) sampling error, since a typical liver biopsy samples only 1/50,000 of all liver tissues, and histological lesions of NASH are unevenly distributed throughout the liver parenchyma [57]; (2) inter- and intra-observer variability, as observed by Gawrieh et al. although there was a high agreement ratio in the assessment of steatosis grading and fibrosis staging between pathologists, the agreement was suboptimal for lobular inflammation and hepatocellular ballooning [58]; and (3) the existence of different criteria for the definition of NASH. The Non-alcoholic Steatohepatitis Clinical Research Network (NASH-CRN) proposed the system termed the NAS scoring system in order to classify NAFLD according to severities of fatty change, inflammation and hepatocellular ballooning [3]. NAS is markedly reproducible and is useful for assessing therapeutic effects in Clinical trials, but it is incapable of diagnosing NASH in patients with burned-out NASH, in whom fatty changes and inflammatory cell infiltration resolving in fibrosis have progressed [59].

Given these limitations, non-invasive methods have been developed for the diagnosis of NASH and fibrosis as a first option to examine NAFLD patients and to help determine which require a liver biopsy. The ideal test should be economical, reproducible and capable of diagnosing the whole spectrum of lesions, including within NAFLD, and even reflecting the

changes produced on initiating specific treatment. Nowadays, we do not have a test available that has these characteristics, so these non-invasive methods are based on diverse complementary approaches: clinical factors, genetics, serological markers, image tests and transient elastography [60, 61].

3.2. Risk factors associated with non-alcoholic steatohepatitis and progressive disease

The best predictor of the evolution of NAFLD is the presence of necroinflammation and fibrosis in liver biopsy; however, there are more and more studies reporting no insignificant rates of progression of simple steatosis [5, 6, 62]. A first study that analyzed patients with NAFLD and paired biopsies demonstrated that even patients with simple steatosis can progress to NASH and advanced fibrosis, especially in the presence of metabolic risk factors [6]. Therefore, there is a series of non-modifiable and modifiable factors in patients associated with a greater risk of development of NASH and more progressive disease.

Various transversal studies have demonstrated that the disease is more severe in older patients, although this phenomenon could be due to the sum of pathogenic factors and a greater duration of the liver disease itself and the associated diseases [8, 63, 64]. In fact, the longitudinal studies have not managed to demonstrate that age is a factor that aggravates the disease per se [65]. The association between sex and fibrosis progression is controversial; two transversal studies show that men and post-menopause women have a greater risk of fibrosis in comparison with pre-menopause women; moreover, precocious menopause is associated with a greater risk of fibrosis [66–69]. Other non-modifiable factors are genetic; dozens of genes with multiple polymorphisms associated with NAFLD have been discovered thanks to genome-wide association studies (GWAS), but the number of strongly validated genes in large independent cohorts is limited to two, *patatin-like phospholipase domain containing 3* (PNPLA3) and *transmembrane 6 superfamily member 2* (TM6SF2) [70]. The presence of the single nucleotide polymorphisms (SNPs) rs738409 and rs58542926 of the genes PNPLA3 and TM6SF2, respectively, has been associated with a greater risk of NAFLD, as well as a more severe disease [71–76]. Recently, an SNP of IL28b (also implicated in the response to interferon in chronic hepatitis C patients (VHC)) has been associated with an increment in fibrosis in NAFLD patients [77]. Moreover, in a control-case study carried out by our working group, we have observed that the presence of the variants rs1421085 and rs1558902 of the fat mass and obesity-associated (FTO) gene confer a high risk of liver inflammation particularly in patients of normal weight with NAFLD (unpublished).

On the other hand, NAFLD tends to be more severe in patients with various factors of the metabolic syndrome, particularly DM2 and obesity. In fact, the reduction in weight and good glycemic control are associated with an improvement in inflammation and liver fibrosis [11, 78, 79]. However, it is known that NASH can also be present in slim subjects although it is unknown whether the natural history of the disease in these slim subjects is similar to that present in obese subjects. As for arterial hypertension, it is arguable whether its treatment improves the histology of NASH [5, 80]. Another factor of the metabolic syndrome, frequent in NAFLD patients, is dyslipidemia, fundamentally in the form of hypertriglyceridemia and atherogenic dyslipidemia [64, 81]; but moreover, a recent study has related the very low-density lipoprotein (VLDL) profile with the NAFLD severity,

observing that a decrease in small VLDL particle concentration is associated with more advanced fibrosis [82]. Vitamin D deficiency is also frequent among NAFLD patients, and its levels have been correlated negatively with the severity of steatosis, inflammation and fibrosis [83, 84].

Another possible factor associated with NAFLD progression is the alcohol consumption, a controversial aspect as despite there being a limit above which the consumption of alcohol would define alcoholic steatohepatitis (≥ 60 g/d in women and ≥ 80 g/d in men), it is not clear that we are confronting a pathology different to NASH given that the pathogeny of these entities presents a great similarity. Moreover, the quantification of alcohol consumption is quite subjective, imprecise, habitually underestimated and not contrasted with objective determinations through biomarkers. At present, there is no agreement on the impact of light-moderate consumption of alcohol on NAFLD given that the literature available about this topic shows contradictory results relate to NAFLD progression [85, 86]. Nevertheless, it seems that all the relevant studies are in favor of a possible benefit from the moderate alcohol consumption, defined as the consumption of up to one drink a day for women and two drinks a day for men [87]. While the consumption of large doses of alcohol leads to the development of insulin resistance and to the infiltration of macrophages into the adipose tissue [88], moderate consumption has been associated with an improvement in the sensitivity to insulin and high concentrations of adiponectin [89–91]. Various studies suggest a significant association between the moderate consumption of alcohol and the less histological severity of NAFLD [92, 93]. As for the development of HCC, only one prospective study exists that evaluates the consumption of alcohol with the risk of HCC in NAFLD, finding a greater risk of this tumor with moderate use of alcohol; however, this study is carried out in patients with cirrhosis due to established NASH, without evaluating the impact of alcohol on patients with a less severe disease [94].

3.3. Non-invasive diagnosis of non-alcoholic steatohepatitis

There is still no available image test in clinical practice capable of differentiating NAFL from NASH, so various biomarkers have been evaluated to predict the existence of NASH, which are related to pathogenesis pathways of the disease (apoptosis/cellular death, inflammation and oxidative stress).

The most studied serum biomarker associated with the presence of NASH is cytokeratin 18 fragments (CK18-F), a product of the degradation resulting from the apoptosis of hepatocytes mediated by caspase 3 [95], which is measured using enzyme-linked immunosorbent assay (ELISA). Various studies have demonstrated a significant increase in CK18-F in NASH patients in comparison with NAFL patients, and a positive correlation with fibrosis and the histological components of NASH [96, 97]. However, the sensitivity and specificity of this test are quite low, around 60% [98]. Oxidized low-density lipoprotein (LDL), thiobarbituric acid reactive substances (TBARS) and malonaldehyde have been used as markers of oxidative stress, but the results are contradictory [99, 100]. Among the markers of inflammation studied include leptin, protein C reactive, interleukin 6, hyaluronic acid, adiponectin and tumor necrosis factor α (TNF α). All of them have been evaluated in short series or pilot studies in heterogeneous groups of patients with contradictory results [101].

With the aim of improving the diagnostic value of the biomarkers, predictive models have been developed that combine some of these serum biomarkers with analytical parameters and clinical variables, but they have not been adequately validated, so up to now, they are not recommendable in clinical practice [102–107] (**Table 2**).

3.3.1. Emerging fields

Emerging fields in the search for non-invasive biomarkers of NAFLD are proteomics, metabolomics and epigenetics.

Proteomics provides essential information about the biologically active entity named protein. Thanks to proteomic analysis, key changes in serum protein expression levels have been demonstrated between control subjects and patients with different stages of fatty liver [108].

In the last years, studies about the use of metabolomic to discover biomarkers of progression of NAFLD have received great interest, and not only in this liver disease [109–111]. In fact, a Spanish group has developed the so-called OWL Liver Test that consists in the determination

Model	Variables	Sensitivity (%)	Specificity (%)
HAIR score	Hypertension, ALT, insulin resistance	80	89
NASHTest	Age, gender, weight, height, cholesterol, triglycerides, α 2-macroglobulin, apolipoprotein A1, haptoglobin, GGT, ALT, AST, bilirubin	88	50
NASH score	PNPLA3 genotype, insulin, AST	75	74
Nice model	Ck18, ALT, metabolic syndrome	84	86
NAFLD diagnostic panel	Diabetes, gender, BMI, triglycerides, M30, M65-M30	91	47
OxNASH	Age, BMI, AST, 13-Hydroxyoctadecadienoic acid, linoleic acid	81	–

Table 2. Predictive models for non-alcoholic steatohepatitis.

of more than 500 serum metabolites through liquid chromatography coupled with mass spectrometry (LC–MS) in NAFLD patients obtaining a metabolomic profile that enables the differentiation between NAFL and NASH with good specificity and sensitivity [112]. Moreover, the same group thanks to the study of metabolomic profiles at the serum level observed two different subtypes of NAFLD according to the involvement of the methionine metabolism, subtype M and subtype no M, distinguishing those patients that could benefit from therapy with SAME (S-adenosyl methionine) [113].

Recently, studies in rodents suggest that epigenetic events, inheritable events not caused by changes in DNA sequence, may influence susceptibility to NASH. The three most commonly described epigenetic mechanisms are DNA (CpG) methylation, post-translational histone modifications and microRNAs (miRNAs). Several miRNAs have been identified in serum/plasma of NAFLD patients that show diagnostic potential for distinguishing NAFL from NASH and advanced fibrosis [114].

4. Diagnosis of hepatic fibrosis

The stage of fibrosis ranges from absent (F0) to cirrhosis (F4), with stages F2–F4 considered to be clinically significant and stages F3–F4 considered to be advanced fibrosis. Apart from liver biopsy, there are two broad categories of non-invasive markers used to determine the stage of liver fibrosis: serum and radiological markers. This stratification based on markers of fibrosis is more tractable than those used for NASH and so it is currently used to identify patients who are at risk of disease progression.

4.1. Serum biomarkers

There are two large groups of predictive models of advanced fibrosis: ‘simple bedside models’, which use a combination of routine blood tests and clinical variables, and ‘complex models’, which use serum markers of fibrosis (measures of extracellular matrix deposition and turnover).

Although several of these predictive models of advanced fibrosis have been evaluated (**Table 3**) [61, 66, 115], two of the tests have been more widely studied and have easily available parameters, *Fibrosis-4 index* (FIB-4) and *NAFLD Fibrosis Score* (NFS). FIB-4 is based on age, levels of AST, ALT and platelet count. Values of this index below -1.30 enable the exclusion of the presence of advanced fibrosis with a sensitivity of 74% and a specificity of 71%, while values above 2.67 indicate advanced fibrosis with a sensitivity and specificity of 33 and 98%, respectively [115]. NFS is another formula developed and validated for the detection of advanced fibrosis that includes age, BMI, presence of diabetes or hyperglycemia, platelet count, albumin and AST/ALT ratio (<http://naflscore.com/>). In a meta-analysis of 13 studies with more than 3000 patients, a value of NFS < -1.455 had a sensitivity of 90% and a specificity of 60% to exclude advanced fibrosis, while a value of >0.676 identified the presence of it with a sensitivity of 67% and a specificity of 97% [116].

Model	Variables	Cut-offs	Sensitivity (%)	Specificity (%)
FIB-4	Age, AST and ALT	1.30 (low cut-off)	74	71
	levels, platelets	2.67 (high cut-off)	33	98
NAFLD fibrosis score	Age, hyperglycemia,	-1.455 (low cut-off)	90	60
	BMV, AST/ALT ratio, albumin, platelet	0.675 (high cut-off)	67	97
AST to platelet ratio index (APRI)	AST, platelet	1	27	89
AST/ALT ratio	AST, ALT	0.8 (low cut-off)	90	60
		1 (high cut-off)	67	97
BAAT score	BMI, age, ALT, serum triglycerides	2	71	80
BARD score	BMI, AST/ALT ratio, diabetes	2	89	44
Enhanced liver fibrosis (ELF) test	Age, hyaluronic acid,	8.5 (low cut-off)	80	90
	TIMP-1, PIIINP	11.3 (high cut-off)		
FibroTest	α 2-macroglobulin,	0.3 (low cut-off)	77	77
	haptoglobin, GGT,	0.7 (high cut-off)	15	90
	bilirubin,			
	apolipoprotein			
Hepascore	Age, gender, bilirubin,	0.44	75	84
	hyaluronic acid, α 2- macroglobulin			

AST, aspartate aminotransferase; ALT, alanine aminotransferase; BMI, body mass index; TIMP-1, tissue inhibitor of metalloproteinase 1; PIIINP, procollagen III amino-terminal peptide; GGT, gamma glutamyl transferase.

Table 3. Predictive models for significant and advanced fibrosis in NAFLD patients.

The principal drawback of all these biomarkers is that none of them is specific of the liver and their results can be influenced by co-morbidities of patients, so a critical interpretation of the result is necessary.

4.2. Imaging methods to measure fibrosis

With respect to image techniques, transient elastography (FibroScan®) is the most widely used technique in the diagnosis of liver fibrosis, not only in NAFLD but also in different chronic

liver diseases [117]. TE measures the propagation velocity of low-frequency waves (50 Hz) through the hepatic parenchyma using ultrasounds and is expressed in kilo Pascal (kPa); the higher the propagation velocity, the greater the stiffness of the tissue. The advantages provided by this technique are its speed, the immediacy of the results and the ease of handling. However, proper results require careful interpretation of data, based on at least 10 successful measurements, a success rate above 60% and an interquartile range (IQR) of <30% of the median value. A limitation of TE in NAFLD is the high rate of technical failure due to the attenuation of the elastic wave by interposition of adipose tissue secondary to the central obesity, very frequent in these patients. Although an XL probe has been developed, which enables greater penetration of the wave, this difficulty is often insurmountable [118, 119]. Moreover, this technique has been initially validated in patients with chronic infection by VHC [120], while the studies focusing on evaluating its use in NAFLD are smaller and have often used different cut-offs [42, 118, 121–129] (**Table 4**). According to the results of several studies, the cut-offs with M probe accepted for NAFLD patients are 7.0 kPa for significant fibrosis ($\geq F2$), 8.7 kPa for advanced fibrosis ($\geq F3$) and 10.3 kPa for cirrhosis (F4) [124, 126, 128]. When using the XL probe, these cut-offs differ as the measure of liver stiffness with this probe is less than that with the M probe in the same patient; in this case, 6.2, 7.2 and 7.9 kPa are the cut-offs for significant fibrosis, advanced fibrosis and cirrhosis, respectively [119, 130, 131].

Another liver elasticity-based imaging technique is ARFI (*acoustic radiation force impulse imaging*). Although for the time being there are few studies that have evaluated its utility in NAFLD patients, its great advantage is that it can be easily connected to traditional ultrasound scan enabling the positioning of the zone of interest under visual control [132, 133]. Another method suitable for studying the elastic properties of the hepatic parenchyma is magnetic resonance elastography (MRE). MRE can be more reliable than TE to diagnose advanced fibrosis; moreover, it has the advantage of being able to evaluate the whole hepatic parenchyma even in obese patients, but the technique is expensive and not widely available [134, 135]. Magnetic resonance imaging is more widely available and is the basis of new software called DEMILI (*Detection of Metabolic-Induced Liver Injury*), which through computerized optical analysis of its images determines a series of optical biomarkers enabling the detection of the presence of NASH (NASHMRI) and predicting significant fibrosis (FibroMRI) in NAFLD patients. For the detection of NASH, a cut-off has been established with NASHMRI of >0.5 , presenting a sensitivity and specificity of 87 and 60%, respectively. In the case of FibroMRI, the cut-off is also >0.5 for the prediction of significant fibrosis with a sensitivity of 77% and a specificity of 80% [136]. Given that this technique enables the analysis of the total volume of the liver, as well as its use in the diagnosis of NASH and significant fibrosis, it enables the potential effects of a therapy to be monitored.

A recently developed technique is multiparametric magnetic resonance (MR) that includes T_1 mapping for fibrosis/inflammation imaging, T_2^* mapping for liver iron quantification and proton magnetic resonance spectroscopy (^1H -MRS) for liver fat quantification. In a recent study, it has demonstrated good correlation with disease severity in NAFLD patients, showing excellent accuracy in quantifying both the inflammatory and fibrotic components of NAFLD [137].

A summary of the approach to the management and characterization of NAFLD patients is shown in **Figure 1**.

Study	Patients, <i>n</i>	Probe	Fibrosis Stage	Cut-off (kPa)	Sensitivity (%)	Specificity (%)
Yoneda et al. [122]	67	M	$F \geq 2$	6.65	82	91
			$F \geq 3$	8.0	87	84
			$F = 4$	17.0	100	98
Yoneda et al. [125]	97	M	$F \geq 2$	6.65	74	97
			$F \geq 3$	9.8	85	81
			$F = 4$	17.5	100	97
Nobili et al. [129]	52	M	$F \geq 2$	7.4	100	92
			$F \geq 3$	10.2	100	100
			$F = 4$	–		
Wong et al. [128]	246	M	$F \geq 2$	7.0	79	76
			$F \geq 3$	8.7	84	83
			$F = 4$	10.3	92	88
Lupsor et al. [124]	72	M	$F \geq 2$	6.8	67	84
			$F \geq 3$	10.4	100	97
			$F = 4$	–		
Petta et al. [118]	169	M	$F \geq 2$	7.25	69	70
			$F \geq 3$	8.75	76	78
			$F = 4$	–		
Kumar et al. [126]	205	M	$F \geq 2$	7.0	78	79
			$F \geq 3$	9.0	85	88
			$F = 4$	11.8	90	88
Pathik et al. [121]	110	M	$F \geq 2$	9.1	–	–
			$F \geq 3$	12.0	90	80
			$F = 4$	20.0	90	80
Cassinotto et al. [123]	291	M	$F \geq 2$	6.2	90	–
			$F \geq 3$	8.2	90	–
			$F = 4$	9.5	90	–
Imajo et al. [42]	142	M	$F \geq 2$	11.0	62	100
			$F \geq 3$	11.4	86	84
			$F = 4$	14.0	100	76

Table 4. Comparative studies of FibroScan with liver biopsy in the detection of fibrosis in NAFLD.

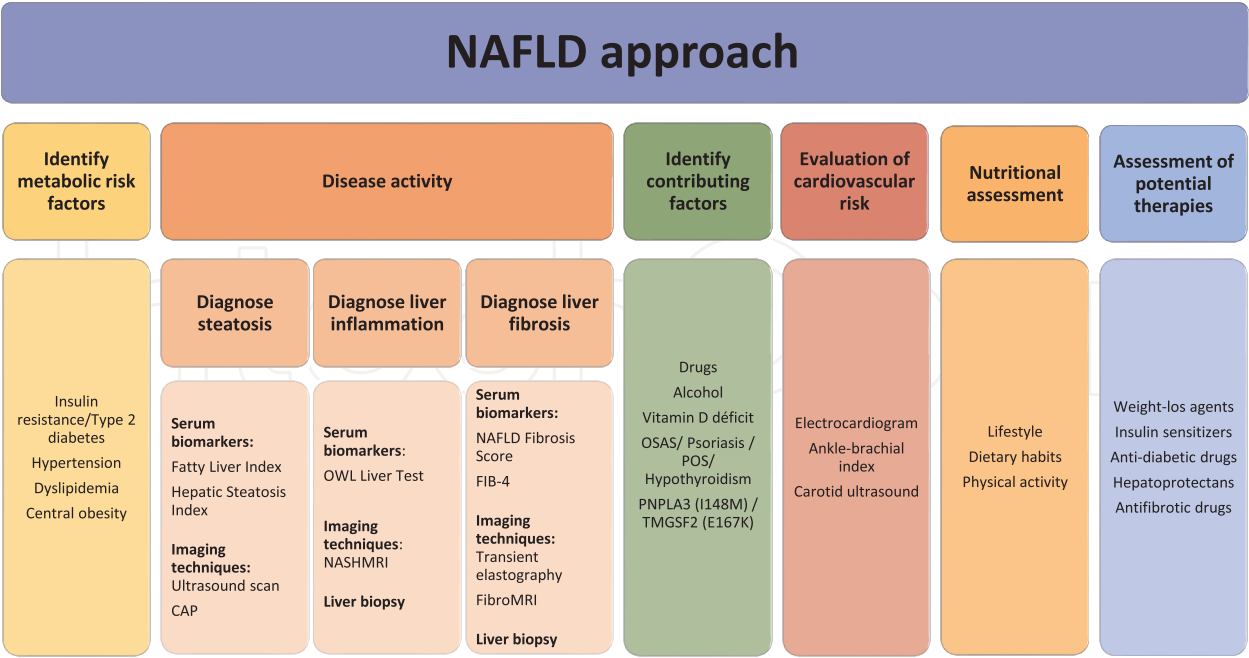


Figure 1. Practical approach to the management of patient with NAFLD.

5. Monitoring disease progression

A recent meta-analysis of 11 studies that evaluated the progression of NAFLD through the use of paired liver biopsies revealed that patients with NAFL and NASH presented a progression of fibrosis of 33.6% and an improvement of it of 22.3%, the rate of fibrosis progression being greater in patients with NASH than with NAFL (progression in a stage of 7.1 years compared to 14.3 years, respectively) [5]. However, there is a lack of homogenization in the speed of fibrosis progression in all these studies with paired biopsy, which is mostly due to the presence of characteristics of the metabolic syndrome in the patients [5–7, 62, 65, 138–144]. For this reason, due to the lack of studies that provide complete data about the differential progression of the disease in patients with different stages of NAFLD, there is no guide on the frequency of follow-up of these patients or on the means available to monitor the progression. Nevertheless, once the diagnosis of NAFLD is made, the follow-up will depend on the presence of metabolic risk factors and the severity of the hepatic disease, which will be determined by the presence of NASH and, especially, the stage of fibrosis.

The principal metabolic factor of the risk of NAFLD progression is DM2 [1, 19, 145]. Patients with DM2 have a more severe grade of NAFLD than the patients without DM2, with rates of NASH up to 80% and of advanced fibrosis of 30–40% [146, 147]. These data confirm the need for closer monitoring in these patients. Bazick et al. [147] developed a clinical model to detect NASH and advanced fibrosis in patients with NAFLD and DM2 with a sensitivity and specificity of 57 and 90%, respectively. This model includes easily accessible parameters such as BMI, circumference at the waist, HbA1c, insulin resistance, ALT, AST, albumin and ferritin, for NASH; and age, BMI, waist/hip ratio, arterial hypertension, ALT/AST ratio, alkaline

phosphatase, bilirubin, globulin, albumin, serum insulin, hematocrit, INR and platelets, to predict advanced fibrosis. However, further studies are still necessary to externally validate this model. Other metabolic factors described with more evidence for the disease progression are central obesity, arterial hypertension and high levels of LDL cholesterol [7, 148–150]. No study shows cost-efficacy in the monitoring of the progression in these at-risk patients, but we recommend carrying out NFS and/or FIB-4 every 2 or 3 years in these patients with non-significant fibrosis, and if NASH and/or significant fibrosis is presented in the initial diagnosis, the follow-up will not differ from the rest of the patients.

The other factor having the greatest effect on the disease progression is liver fibrosis [151]. In general, in a period of 15 years, 13% of the patients with stage F2 and 25% of those presenting with F3 will develop cirrhosis [6, 7, 62]. These patients with significant fibrosis should be considered for pharmacological treatment, besides lifestyle modifications (diet and exercise). Moreover, NAFLD patients may develop HCC even in the absence of cirrhosis [152], given that it is the continuous hepatocyte injury that leads to a compensatory proliferation, key driver of the development of HCC [153]. Therefore, patients with NASH and significant fibrosis, which is indicative of important cellular damage, are also at risk of developing this liver tumor.

With all this information, we recommend recalculating NFS and/or FIB-4 every 4–5 years for patients with NAFL without risk factors or if the patient develops DM2; in patients with NASH without significant fibrosis, we recommend an annual follow-up with a calculation of NFS and/or FIB-4 and carrying out TE and ultrasound, and in patients with significant fibrosis, a 6-monthly follow-up is recommended with special interest in screening for HCC. The management and follow-up of the patients with advanced fibrosis/cirrhosis due to NASH does not differ from the rest of etiologies [25].

Another important question is the evaluation of the response to the therapy provided. The non-invasive methods available currently have not been reliable or have not been validated to document efficacy of the treatments, so liver biopsy is still necessary to determine this efficacy, especially in a clinical trial setting.

6. Screening of associated diseases

In recent years, several studies have confirmed that the morbimortality associated with NAFLD is not limited only to hepatic injury, yet it is a disease with multisystemic behavior with affectation of different organs.

6.1. Insulin resistance and metabolic syndrome

As was previously mentioned, the concurrent characteristics of metabolic syndrome increase the risk of developing NAFLD, and a recent study of the HepaMet group relates the severity of NAFLD with the number of factors of the metabolic syndrome present (publication pending). However, the presence of NAFLD in itself also increases the risk of developing complications such as dyslipidemia and insulin resistance [154–156]. In this sense, the diagnosis and

quantification of hepatic fat can be useful in the prediction of future development of diabetes and other cardiovascular risk factors [56].

Insulin resistance is a key in the physiopathology of NAFLD, associated with the increase in the deposit of fat and fibrosis, and it substantially increases the risk of developing DM2, which indicates that NAFLD can precede the development of diabetes. Moreover, and as it was mentioned earlier, several studies have demonstrated that, especially in patients with insulin resistance and/or diabetes, liver fibrosis can progress even when a baseline hepatic histology described only simple steatosis without hepatocellular damage [62, 141]. All in all, in daily clinical practice the use of screening tools is necessary to detect the presence of diabetes (fasting blood glucose levels, HbA1c or, if available, the oral glucose tolerance test) or insulin resistance. The reference technique for the diagnosis of IR in non-diabetic patients is the hyperinsulinemic-euglycemic clamp test, although this procedure is expensive and complicated, so it is not routinely used in daily clinical practice [157]. In these cases, the calculation of HOMA-IR (*homeostatic model assessment*) is an acceptable alternative to evaluate the IR, although there is no agreement on the threshold that defines insulin resistance using this formula [158]. Nevertheless, HOMA-IR can help us during the follow-up to identify patients at risk of fibrosis progression [6, 62]. The next question once the patients with IR are identified is whether it is necessary to treat them pharmacologically or not; and, whether in diabetic patients is necessary to intensify the anti-diabetic treatment to avoid liver disease progression or not. As expected, several insulin-sensitizing agents have demonstrated an improvement in the hepatic histology [159–161], even in patients without DM2 [162, 163], given that both entities share multiple physiopathological mechanisms, so this treatment can be considered in patients with NASH and/or multiple factors of progression in which a decrease of IR cannot be achieved with diet and exercise, although the EASL and AASLD guidelines do not contemplate it. Given that IR plays an essential role in NAFLD progression but not the only one, we do not believe that it is necessary to treat DM2 differently/intensely in patients with NAFLD, independently of the grade, provided that the IR is controlled.

6.2. Cardiovascular disease

Cardiovascular disease is quantitatively the main cause of death in NAFLD patients. Besides the risk itself of the characteristics of the metabolic syndrome, multiple pathogenic conditions of NAFLD contribute to the development of cardiovascular disease. In fact, patients with NAFLD often present elevation in the markers implicated in the development of atherosclerosis, such as CD36 in its soluble form (sCD36), a membrane receptor responsible for, among other things, the transport of fatty acids [164]. The spectrum of CVD in NAFLD includes atherosclerotic coronary heart disease, heart failure and cardiac arrhythmias. This necessitates the study of probable CVD, especially subclinical atherosclerosis, in all these patients [10]. There are little data to define the optimal means of screening NAFLD patients with CVD, but it is important to be aware that there are different techniques for the detection of subclinical atherosclerosis that are bloodless and some of which are very easily performed. Among these, the measurement of ankle-brachial index and carotid ultrasound are assessments especially useful for patients with intermediate cardiovascular risk, situation affecting a very important part of the population with NAFLD [165].

6.3. Extrahepatic cancer

The second most prevalent cause of death among patients with NAFLD is cancer, both gastrointestinal (colon, esophagus, stomach and pancreas) and extraintestinal (kidney and breast), which leads to the suspicion that this liver disease might promote the development of neoplasms.

The association of insulin resistance/diabetes, obesity and metabolic syndrome with an increase in the risk of a large number of cancers is well established [166–171]. These three characteristics are closely related to NAFLD and contribute significantly to the risk of developing HCC; nevertheless, various recent studies indicate that NAFLD can be an additional and independent risk factor for extrahepatic cancers [172, 173], especially colorectal cancer (CRC) [127, 174]. In several studies, colorectal lesions, particularly tubular adenomas and carcinomas, were significantly more prevalent in NAFLD patients, regardless of age, sex and manifestations of metabolic syndrome; even the presence of NASH has been related to a greater risk in comparison with those with NAFL [174, 175]. This rise in the risk of CRC in NAFLD can be explained by the increase in insulin and pro-inflammatory cytokines and the alteration of the adipokines metabolism predominantly leptin versus adiponectin that exists in these patients and which promotes cellular proliferation, inhibition of apoptosis and angiogenesis [176, 177].

Although these data clearly suggest more rigorous screening programmes for CRC in NAFLD patients, there are no well-designed prospective studies enabling the verification of a causal relation between NAFLD and CRC or studies that evaluate the usefulness of earlier screening in this liver disease, so no guidelines make a distinction with respect to CRC screening in these patients.

6.4. Other associated diseases

There is increasing interest in the possible contribution of NAFLD to the development and progression of chronic kidney disease (CKD) [178–181]. A recent meta-analysis has revealed that the presence and severity of NAFLD are associated with an increase in the risk and severity of CKD [181]. However, it is difficult to establish NAFLD as an independent risk factor of CKD given the close relation between NAFLD and other known risk factors of CKD such as obesity and IR. Obstructive sleep apnea syndrome (OSAS) is strongly associated with NAFLD independently of other traditional factors; it is a consequence of the decrease in the lipid metabolism provoked by intermittent hypoxia [182–185]. Other described diseases associated with NAFLD include osteoporosis [186], psoriasis [187], polycystic syndrome [188] and other endocrinopathies such as hypothyroidism [189], hypopituitarism [190] and hypogonadism [191]. Until now, there is no evidence for screening of all these pathologies for the mere fact that the subject presents NAFLD, so all that needs to be studied is the presence of them if the patient has clinical manifestations related to them. Moreover, a recent study by our group has demonstrated that psychotic patients with specific pharmacological treatment have a high risk of developing NAFLD in the first years, so its early detection will enable better prevention of cardiovascular events, which are so increased in this population [192].

7. Diagnostic algorithm and follow-up

While working on the different sections of this chapter, we have detailed the fundamental elements for the development of a diagnostic algorithm and a follow-up procedure for NAFLD (Figure 2). This algorithm is based on clinical evidence available in the current literature with respect to the topic and on different guidelines issued by the principal international associations for the study of the liver (EASL and AASLD). In the case of monitoring and follow-up of these patients where the existing evidence is not relevant in certain aspects, our recommendations are based on the experience of our clinical group in different high-quality studies in this field.

Once the initial diagnosis of NAFLD is made, our posterior attitude will depend on the result of the non-invasive liver fibrosis methods. In general, current image techniques are quite reliable to distinguish between advanced fibrosis ($\geq F3$) and mild fibrosis or null ($F0-F1$), but they are insufficient to identify those patients with significant fibrosis ($\geq F2$). Therefore, in clinical practice, we recommend the combination of elastographic techniques with serum markers, more specifically TE and NFS due to their wide accessibility and ease of application. When these two parameters generate doubt about the grade of fibrosis or indicate possible significant fibrosis, liver biopsy is necessary. Depending on the result, we determine the posterior follow-up as can be seen in the algorithm (Figure 2). The presence of metabolic risk factors influences not only the therapeutic management but also the follow-up. If liver biopsy is

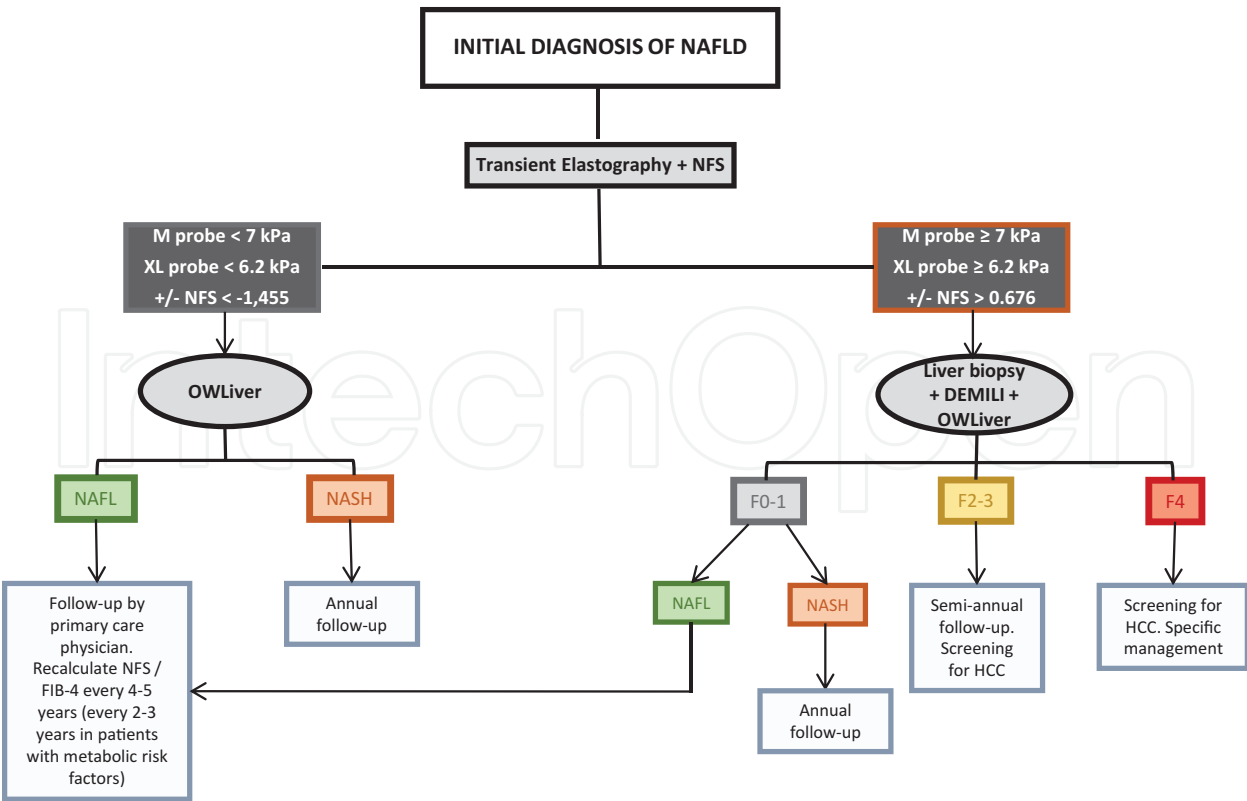


Figure 2. Clinical algorithm for the diagnosis of NAFLD and monitoring disease progression.

not to be performed on the patient with NAFLD, due to advanced age, to the absence of significant fibrosis in the non-invasive methods or to contraindication, we could evaluate the performance of the OWL Liver Test to help identify those patients with NASH who require a closer follow-up. If the patient does not present improvement in laboratory parameters even in imaging tests, we should evaluate to repeat liver biopsy 5 years after the last one, or even before if progression of the disease is suspected.

8. Conclusions

NAFLD is currently the primary cause of chronic liver disease in the western world and its growth is a consequence of its close relation to obesity and metabolic syndrome. One of the great challenges in this disease is to diagnose and classify it correctly, given that the characteristics defining NAFLD are the common denominator of many liver diseases. Its correct characterization is important as in spite of presenting a generally benign and slowly developing evolution from the hepatic viewpoint; the fatty liver can progress towards more severe forms with the development of inflammation, fibrosis, cirrhosis and HCC, thus conferring morbimortality. However, its potential morbimortality is not limited to this organ, but goes beyond; NAFLD is being considered a mediator of systemic diseases. Therefore, the early identification of these patients would help to improve its prognosis through an individualized intervention depending on the stage of liver disease, on the metabolic risk factors present and on the cardiovascular risk, which translates into the need for a systemic approach to the disease with multidisciplinary management including primary care physician, endocrinologists, nutritionists, psychologists and hepatologists.

Conflict of interest

The authors declared that they do not have anything to disclose regarding funding or conflict of interest with respect to this chapter.

Author details

Paula Iruzubieta, Marta González, Joaquín Cabezas, María Teresa Arias-Loste and Javier Crespo*

*Address all correspondence to: javiercrespo1991@gmail.com

Department of Gastroenterology and Hepatology, Marqués de Valdecilla University Hospital, Centro de Investigación Biomédica en Red de Enfermedades Hepáticas y Digestivas (CIBERehd), Infection, Immunity and Digestive Pathology Group, Research Institute Marqués de Valdecilla (IDIVAL), Santander, Spain

References

- [1] Vernon G, Baranova A, Younossi ZM. Systematic review: The epidemiology and natural history of non-alcoholic fatty liver disease and non-alcoholic steatohepatitis in adults. *Alimentary Pharmacology & Therapeutics*. [Internet]. Aug 2011;**34**(3):274-285. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/21623852> [Accessed: Jul 10, 2014]
- [2] Ludwig J, Viggiano TR, McGill DB, Oh BJ. Nonalcoholic steatohepatitis: Mayo Clinic experiences with a hitherto unnamed disease. *Mayo Clinic Proceedings* [Internet]. Jul 1980;**55**(7):434-438. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/7382552> [Accessed: Dec 26, 2016]
- [3] Kleiner DE, Brunt EM, Van Natta M, Behling C, Contos MJ, Cummings OW, et al. Design and validation of a histological scoring system for nonalcoholic fatty liver disease. *Hepatology* [Internet]. Jun 2005;**41**(6):1313-1321. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/15915461> [Accessed: Jun 17, 2015]
- [4] Cuadrado A, Orive A, García-Suárez C, Domínguez A, Fernández-Escalante JC, Crespo J, et al. Non-alcoholic steatohepatitis (NASH) and hepatocellular carcinoma. *Obesity Surgery* [Internet]. Mar 1, 2005;**15**(3):442-446. Available from: <http://www.springerlink.com/index/10.1381/0960892053576596> [Accessed: Dec 26, 2016]
- [5] Singh S, Allen AM, Wang Z, Prokop LJ, Murad MH, Loomba R. Fibrosis progression in nonalcoholic fatty liver vs nonalcoholic steatohepatitis: A systematic review and meta-analysis of paired-biopsy studies. *Clinical Gastroenterology and Hepatology* [Internet]. Apr 2015;**13**(4):643-54-9-40. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S1542356514006028> [Accessed: Dec 26, 2016]
- [6] Pais R, Charlotte F, Fedchuk L, Bedossa P, Lebray P, Poynard T, et al. A systematic review of follow-up biopsies reveals disease progression in patients with non-alcoholic fatty liver. *Journal of Hepatology* [Internet]. Sep 2013;**59**(3):550-556. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/23665288> [Accessed: Apr 23, 2016]
- [7] 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. *Journal of Hepatology* [Internet]. Jan 2005;**42**(1):132-138. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0168827804004350> [Accessed: Dec 26, 2016]
- [8] Ong JP, Pitts A, Younossi ZM. Increased overall mortality and liver-related mortality in non-alcoholic fatty liver disease. *Journal of Hepatology* [Internet]. Oct 2008;**49**(4):608-612. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/18682312> [Accessed: Jun 4, 2016]
- [9] Stepanova M, Rafiq N, Makhoul H, Agrawal R, Kaur I, Younoszai Z, et al. Predictors of all-cause mortality and liver-related mortality in patients with non-alcoholic fatty liver disease (NAFLD). *Digestive Diseases and Sciences* [Internet]. Oct 2013;**58**(10):3017-3023. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/23775317> [Accessed: Jun 4, 2016]
- [10] Francque SM, van der Graaff D, Kwanten WJ. Non-alcoholic fatty liver disease and cardiovascular risk: Pathophysiological mechanisms and implications. *Journal of Hepatology*

- [Internet]. Aug 2016;**65**(2):425-443. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0168827816301076> [Accessed: Dec 27, 2016]
- [11] Anstee QM, Targher G, Day CP. Progression of NAFLD to diabetes mellitus, cardiovascular disease or cirrhosis. *Nature Reviews Gastroenterology & Hepatology* [Internet]. Jun 2013;**10**(6):330-344. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/23507799> [Accessed: May 17, 2016]
- [12] Machado M, Marques-Vidal P, Cortez-Pinto H. Hepatic histology in obese patients undergoing bariatric surgery. *Journal of Hepatology* [Internet]. Oct 2006;**45**(4):600-606. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/16899321> [Accessed: Mar 1, 2016]
- [13] Boza C, Riquelme A, Ibañez L, Duarte I, Norero E, Viviani P, et al. Predictors of non-alcoholic steatohepatitis (NASH) in obese patients undergoing gastric bypass. *Obesity Surgery* [Internet]. Sep 2005;**15**(8):1148-1153. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/16197788> [Accessed: Jun 4, 2016]
- [14] Fabbrini E, Magkos F. Hepatic steatosis as a marker of metabolic dysfunction. *Nutrients* [Internet]. Jun 2015;**7**(6):4995-5019. Available from: <http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=4488828&tool=pmcentrez&rendertype=abstract> [Accessed: Jun 4, 2016]
- [15] Gastaldelli A, Cusi K, Pettiti M, Hardies J, Miyazaki Y, Berria R, et al. Relationship between hepatic/visceral fat and hepatic insulin resistance in nondiabetic and type 2 diabetic subjects. *Gastroenterology* [Internet]. Aug 2007;**133**(2):496-506. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/17681171> [Accessed: Mar 8, 2016]
- [16] Kotronen A, Juurinen L, Hakkarainen A, Westerbacka J, Cornér A, Bergholm R, et al. Liver fat is increased in type 2 diabetic patients and underestimated by serum alanine aminotransferase compared with equally obese nondiabetic subjects. *Diabetes Care* [Internet]. Jan 2008;**31**(1):165-169. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/17934148> [Accessed: Jun 4, 2016]
- [17] Targher G, Bertolini L, Rodella S, Tessari R, Zenari L, Lippi G, et al. Nonalcoholic fatty liver disease is independently associated with an increased incidence of cardiovascular events in type 2 diabetic patients. *Diabetes Care* [Internet]. Aug 2007;**30**(8):2119-2121. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/17519430> [Accessed: Jun 4, 2016]
- [18] Leite NC, Salles GF, Araujo ALE, Villela-Nogueira CA, Cardoso CRL. Prevalence and associated factors of non-alcoholic fatty liver disease in patients with type-2 diabetes mellitus. *Liver International* [Internet]. Jan 2009;**29**(1):113-119. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/18384521> [Accessed: Jun 4, 2016]
- [19] Loomba R, Abraham M, Unalp A, Wilson L, Lavine J, Doo E, et al. Association between diabetes, family history of diabetes, and risk of nonalcoholic steatohepatitis and fibrosis. *Hepatology* [Internet]. Sep 2012;**56**(3):943-951. Available from: <http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=3407289&tool=pmcentrez&rendertype=abstract> [Accessed: Jun 4, 2016]

- [20] Streba LAM, Vere CC, Rogoveanu I, Streba CT. Nonalcoholic fatty liver disease, metabolic risk factors, and hepatocellular carcinoma: An open question. *World Journal of Gastroenterology* [Internet]. Apr 14, 2015;**21**(14):4103-4110. Available from: <http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=4394070&tool=pmcentrez&rendertype=abstract> [Accessed: Jun 4, 2016]
- [21] Marchesini G, Bugianesi E, Forlani G, Cerrelli F, Lenzi M, Manini R, et al. Nonalcoholic fatty liver, steatohepatitis, and the metabolic syndrome. *Hepatology* [Internet]. Apr 2003;**37**(4):917-923. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/12668987> [Accessed: May 14, 2016]
- [22] Doycheva I, Cui J, Nguyen P, Costa EA, Hooker J, Hofflich H, et al. Non-invasive screening of diabetics in primary care for NAFLD and advanced fibrosis by MRI and MRE. *Alimentary Pharmacology & Therapeutics* [Internet]. Jan 2016;**43**(1):83-95. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/26369383> [Accessed: Jan 31, 2017]
- [23] Zhang E, Wartelle-Bladou C, Lepanto L, Lachaine J, Cloutier G, Tang A. Cost-utility analysis of nonalcoholic steatohepatitis screening. *European Radiology* [Internet]. Nov 21, 2015;**25**(11):3282-3294. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25994191> [Accessed: Jan 31, 2017]
- [24] Chalasani N, Younossi Z, Lavine JE, Diehl AM, Brunt EM, Cusi K, et al. The diagnosis and management of non-alcoholic fatty liver disease: Practice Guideline by the American Association for the Study of Liver Diseases, American College of Gastroenterology, and the American Gastroenterological Association. *Hepatology* [Internet]. Jun 2012;**55**(6):2005-2023. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/22488764> [Accessed: Feb 1, 2017]
- [25] European Association for the Study of the Liver (EASL), European Association for the Study of Diabetes (EASD), European Association for the Study of Obesity (EASO). EASL-EASD-EASO Clinical Practice Guidelines for the management of non-alcoholic fatty liver disease. *Journal of Hepatology* [Internet]. Jun 2016;**64**(6):1388-1402. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/27062661> [Accessed: Dec 27, 2016]
- [26] Angulo P. Nonalcoholic fatty liver disease. *The New England Journal of Medicine* [Internet]. Apr 18, 2002;**346**(16):1221-1231. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/11961152> [Accessed: Feb 21, 2015]
- [27] Tahan V, Canbakan B, Balci H, Dane F, Akin H, Can G, et al. Serum gamma-glutamyltranspeptidase distinguishes non-alcoholic fatty liver disease at high risk. *Hepatogastroenterology* [Internet]. Feb 1, 2017;**55**(85):1433-1438. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/18795706>
- [28] O'Brien J, Powell LW. Non-alcoholic fatty liver disease: Is iron relevant? *Hepatology International* [Internet]. Jan 12, 2012;**6**(1):332-341. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/22020821> [Accessed: Feb 1, 2017]
- [29] Ravi S, Shoreibah M, Raff E, Bloomer J, Kakati D, Rasheed K, et al. Autoimmune markers do not impact clinical presentation or natural history of steatohepatitis-related

- liver disease. *Digestive Diseases and Sciences* [Internet]. Dec 15, 2015;**60**(12):3788-3793. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/26173506> [Accessed: Feb 2, 2017]
- [30] Amarapurkar DN, Patel ND. Clinical spectrum and natural history of non-alcoholic steatohepatitis with normal alanine aminotransferase values. *Tropical Gastroenterology* [Internet]. Feb 2, 2017;**25**(3):130-134. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/15682660>
- [31] Portillo-Sanchez P, Bril F, Maximos M, Lomonaco R, Biernacki D, Orsak B, et al. High prevalence of nonalcoholic fatty liver disease in patients with type 2 diabetes mellitus and normal plasma aminotransferase levels. *The Journal of Clinical Endocrinology and Metabolism* [Internet]. Jun 2015;**100**(6):2231-2238. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25885947> [Accessed: Feb 2, 2017]
- [32] Schwenzer NF, Springer F, Schraml C, Stefan N, Machann J, Schick F. Non-invasive assessment and quantification of liver steatosis by ultrasound, computed tomography and magnetic resonance. *Journal of Hepatology* [Internet]. Sep 2009;**51**(3):433-445. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/19604596> [Accessed: Feb 2, 2017]
- [33] Saadeh S, Younossi ZM, Remer EM, Gramlich T, Ong JP, Hurley M, et al. The utility of radiological imaging in nonalcoholic fatty liver disease. *Gastroenterology* [Internet]. Sep 2002;**123**(3):745-750. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/12198701> [Accessed: Feb 2, 2017]
- [34] Dasarathy S, Dasarathy J, Khiyami A, Joseph R, Lopez R, McCullough AJ. Validity of real time ultrasound in the diagnosis of hepatic steatosis: A prospective study. *Journal of Hepatology* [Internet]. Dec 2009;**51**(6):1061-1067. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0168827809005856> [Accessed: Feb 2, 2017]
- [35] Nouredin M, Lam J, Peterson MR, Middleton M, Hamilton G, Le T-A, et al. Utility of magnetic resonance imaging versus histology for quantifying changes in liver fat in nonalcoholic fatty liver disease trials. *Hepatology* [Internet]. Dec 2013;**58**(6):1930-1940. Available from: <http://doi.wiley.com/10.1002/hep.26455> [Accessed: Feb 2, 2017]
- [36] Raptis DA, Fischer MA, Graf R, Nanz D, Weber A, Moritz W, et al. MRI: The new reference standard in quantifying hepatic steatosis? *Gut* [Internet]. Jan 2012;**61**(1):117-127. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/21997548> [Accessed: Feb 2, 2017]
- [37] Sasso M, Beaugrand M, de Ledinghen V, Douvin C, Marcellin P, Poupon R, et al. Controlled attenuation parameter (CAP): A novel VCTE™ guided ultrasonic attenuation measurement for the evaluation of hepatic steatosis: Preliminary study and validation in a cohort of patients with chronic liver disease from various causes. *Ultrasound in Medicine and Biology* [Internet]. Nov 2010;**36**(11):1825-1835. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0301562910003546> [Accessed: Mar 4, 2017]
- [38] Lédíngheñ V, Vergniol J, Foucher J, Merrouche W, Bail B. Non-invasive diagnosis of liver steatosis using controlled attenuation parameter (CAP) and transient elastography. *Liver International* [Internet]. Jul 2012;**32**(6):911-918. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/22672642> [Accessed: Mar 4, 2017]

- [39] Shen F, Zheng R-D, Mi Y-Q, Wang X-Y, Pan Q, Chen G-Y, et al. Controlled attenuation parameter for non-invasive assessment of hepatic steatosis in Chinese patients. *World Journal of Gastroenterology* [Internet]. Apr 28, 2014;**20**(16):4702. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/24782622> [Accessed: Mar 4, 2017]
- [40] Kumar M, Rastogi A, Singh T, Behari C, Gupta E, Garg H, et al. Controlled attenuation parameter for non-invasive assessment of hepatic steatosis: Does etiology affect performance? *Journal of Gastroenterology and Hepatology* [Internet]. Jul 2013;**28**(7):1194-1201. Available from: <http://doi.wiley.com/10.1111/jgh.12134> [Accessed: Mar 4, 2017]
- [41] Myers RP, Pollett A, Kirsch R, Pomier-Layrargues G, Beaton M, Levstik M, et al. Controlled Attenuation Parameter (CAP): A noninvasive method for the detection of hepatic steatosis based on transient elastography. *Liver International* [Internet]. Jul 2012;**32**(6):902-910. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/22435761> [Accessed: Mar 4, 2017]
- [42] Imajo K, Kessoku T, Honda Y, Tomeno W, Ogawa Y, Mawatari H, et al. Magnetic resonance imaging more accurately classifies steatosis and fibrosis in patients with non-alcoholic fatty liver disease than transient elastography. *Gastroenterology* [Internet]. Mar 2016;**150**(3):626-637.e7. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0016508515017345> [Accessed: Feb 19, 2017]
- [43] Chan W-K, Nik Mustapha NR, Mahadeva S. Controlled attenuation parameter for the detection and quantification of hepatic steatosis in nonalcoholic fatty liver disease. *Journal of Gastroenterology and Hepatology* [Internet]. Jul 2014;**29**(7):1470-1476. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/24548002> [Accessed: Mar 4, 2017]
- [44] de Lédinghen V, Wong GL-H, Vergniol J, Chan HL-Y, Hiriart J-B, Chan AW-H, et al. Controlled attenuation parameter for the diagnosis of steatosis in non-alcoholic fatty liver disease. *Journal of Gastroenterology and Hepatology* [Internet]. Apr 2016;**31**(4):848-855. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/26514665> [Accessed: Mar 4, 2017]
- [45] Karlas T, Petroff D, Sasso M, Fan J-G, Mi Y-Q, de Lédinghen V, et al. Individual patient data meta-analysis of controlled attenuation parameter (CAP) technology for assessing steatosis. *Journal of Hepatology* [Internet]. May 2017;**66**(5):1022-1230. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0168827816307553> [Accessed: Feb 2, 2017]
- [46] Tang A, Desai A, Hamilton G, Wolfson T, Gamst A, Lam J, et al. Accuracy of MR imaging-estimated proton density fat fraction for classification of dichotomized histologic steatosis grades in nonalcoholic fatty liver disease. *Radiology* [Internet]. Feb 2015;**274**(2):416-425. Available from: <http://pubs.rsna.org/doi/10.1148/radiol.14140754> [Accessed: Oct 29, 2017]
- [47] Bedogni G, Bellentani S, Miglioli L, Masutti F, Passalacqua M, Castiglione A, et al. The fatty liver index: A simple and accurate predictor of hepatic steatosis in the general population. *BMC Gastroenterology* [Internet]. Nov 2, 2006;**6**(1):33. Available from: <http://bmcgastroenterol.biomedcentral.com/articles/10.1186/1471-230X-6-33> [Accessed: Feb 2, 2017]

- [48] Kotronen A, Peltonen M, Hakkarainen A, Sevastianova K, Bergholm R, Johansson LM, et al. Prediction of non-alcoholic fatty liver disease and liver fat using metabolic and genetic factors. *Gastroenterology* [Internet]. Sep 2009;**137**(3):865-872. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0016508509009135> [Accessed: Feb 2, 2017]
- [49] Lee J-H, Kim D, Kim HJ, Lee C-H, Yang JI, Kim W, et al. Hepatic steatosis index: A simple screening tool reflecting nonalcoholic fatty liver disease. *Digestive and Liver Disease* [Internet]. Jul 2010;**42**(7):503-508. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S1590865809003363> [Accessed: Feb 2, 2017]
- [50] Poynard T, Ratziau V, Naveau S, Thabut D, Charlotte F, Messous D, et al. The diagnostic value of biomarkers (SteatoTest) for the prediction of liver steatosis. *Comparative Hepatology* Dec 23, 2005;**4**(1):10. Available from: <http://comparative-hepatology.biomed-central.com/articles/10.1186/1476-5926-4-10> [Accessed: Feb 2, 2017]
- [51] Cuthbertson DJ, Weickert MO, Lythgoe D, Sprung VS, Dobson R, Shoaiee-Moradie F, et al. External validation of the fatty liver index and lipid accumulation product indices, using ¹H-magnetic resonance spectroscopy, to identify hepatic steatosis in healthy controls and obese, insulin-resistant individuals. *European Journal of Endocrinology* [Internet]. Nov 8, 2014;**171**(5):561-569. Available from: <http://www.eje-online.org/cgi/doi/10.1530/EJE-14-0112> [Accessed: Oct 29, 2017]
- [52] Calori G, Lattuada G, Ragogna F, Garancini MP, Crosignani P, Villa M, et al. Fatty liver index and mortality: The Cremona study in the 15th year of follow-up. *Hepatology* [Internet]. Jul 2011;**54**(1):145-152. Available from: <http://doi.wiley.com/10.1002/hep.24356> [Accessed: Mar 4, 2017]
- [53] Younossi ZM, Stepanova M, Rafiq N, Makhoul H, Younoszai Z, Agrawal R, et al. Pathologic criteria for nonalcoholic steatohepatitis: Interprotocol agreement and ability to predict liver-related mortality. *Hepatology* [Internet]. Jun 2011;**53**(6):1874-1882. Available from: <http://doi.wiley.com/10.1002/hep.24268> [Accessed: Dec 26, 2016]
- [54] Angulo P, Kleiner DE, Dam-Larsen S, Adams LA, Bjornsson ES, Charatcharoenwitthaya P, et al. Liver fibrosis, but no other histologic features, is associated with long-term outcomes of patients with nonalcoholic fatty liver disease. *Gastroenterology* [Internet]. Aug 2015;**149**(2):389-97.e10. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0016508515005995> [Accessed: Dec 26, 2016]
- [55] Angulo P. Long-term mortality in nonalcoholic fatty liver disease: Is liver histology of any prognostic significance? *Hepatology* [Internet]. Feb 2010;**51**(2):373-375. Available from: <http://doi.wiley.com/10.1002/hep.23521> [Accessed: Feb 7, 2017]
- [56] Arulanandan A, Ang B, Bettencourt R, Hooker J, Behling C, Lin GY, et al. Association between quantity of liver fat and cardiovascular risk in patients with nonalcoholic fatty liver disease independent of nonalcoholic steatohepatitis. *Clinical Gastroenterology and Hepatology* [Internet]. Aug 2015;**13**(8):1513-20.e1. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S1542356515001135> [Accessed: Feb 2, 2017]

- [57] Ratziu V, Charlotte F, Heurtier A, Gombert S, Giral P, Bruckert E, et al. Sampling variability of liver biopsy in nonalcoholic fatty liver disease. *Gastroenterology* [Internet]. Jun 2005;**128**(7):1898-1906. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/15940625> [Accessed: Feb 4, 2017]
- [58] Gawrieh S, Knoedler DM, Saeian K, Wallace JR, Komorowski RA. Effects of interventions on intra- and interobserver agreement on interpretation of nonalcoholic fatty liver disease histology. *Annals of Diagnostic Pathology* [Internet]. Feb 2011;**15**(1):19-24. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S1092913410001413> [Accessed: Oct 29, 2017]
- [59] Sumida Y, Nakajima A, Itoh Y. Limitations of liver biopsy and non-invasive diagnostic tests for the diagnosis of nonalcoholic fatty liver disease/nonalcoholic steatohepatitis. *World Journal of Gastroenterology* [Internet]. Jan 14, 2014;**20**(2):475-485. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/24574716> [Accessed: Oct 29, 2017]
- [60] Castera L, Vilgrain V, Angulo P. Noninvasive evaluation of NAFLD. *Nature Reviews Gastroenterology & Hepatology* [Internet]. Sep 24, 2013;**10**(11):666-675. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/24061203> [Accessed: Feb 7, 2017]
- [61] Kaswala DH, Lai M, Afdhal NH. Fibrosis assessment in nonalcoholic fatty liver disease (NAFLD) in 2016. *Digestive Diseases and Sciences* [Internet]. May 26, 2016;**61**(5):1356-1364. Available from: <http://link.springer.com/10.1007/s10620-016-4079-4> [Accessed: Feb 8, 2017]
- [62] McPherson S, Hardy T, Henderson E, Burt AD, Day CP, Anstee QM. Evidence of NAFLD progression from steatosis to fibrosing-steatohepatitis using paired biopsies: Implications for prognosis and clinical management. *Journal of Hepatology* [Internet]. May 2015;**62**(5):1148-1155. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25477264> [Accessed: Apr 19, 2016]
- [63] Frith J, Day CP, Henderson E, Burt AD, Newton JL. Non-alcoholic fatty liver disease in older people. *Gerontology* [Internet]. Jan 2009;**55**(6):607-613. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/19690397> [Accessed: May 30, 2016]
- [64] Assy N, Kaita K, Mymin D, Levy C, Rosser B, Minuk G. Fatty infiltration of liver in hyperlipidemic patients. *Digestive Diseases and Sciences* [Internet]. Oct 2000;**45**(10):1929-1934. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/11117562> [Accessed: May 23, 2016]
- [65] Hui AY, Wong VW-S, Chan HL-Y, Liew C-T, Chan JL-Y, Chan FK-L, et al. Histological progression of non-alcoholic fatty liver disease in Chinese patients. *Alimentary Pharmacology & Therapeutics* [Internet]. Feb 15, 2005;**21**(4):407-413. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/15709991> [Accessed: Dec 26, 2016]
- [66] McPherson S, Stewart SF, Henderson E, Burt AD, Day CP. Simple non-invasive fibrosis scoring systems can reliably exclude advanced fibrosis in patients with non-alcoholic fatty liver disease. *Gut* [Internet]. Sep 1, 2010;**59**(9):1265-1269. Available from: <http://gut.bmj.com/cgi/doi/10.1136/gut.2010.216077> [Accessed: Dec 26, 2016]

- [67] Hossain N, Afendy A, Stepanova M, Nader F, Srishord M, Rafiq N, et al. Independent predictors of fibrosis in patients with nonalcoholic fatty liver disease. *Clinical Gastroenterology and Hepatology* [Internet]. Nov 2009;**7**(11):1224-1229.e2. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/19559819> [Accessed: Dec 26, 2016]
- [68] Yang JD, Abdelmalek MF, Pang H, Guy CD, Smith AD, Diehl AM, et al. Gender and menopause impact severity of fibrosis among patients with nonalcoholic steatohepatitis. *Hepatology* [Internet]. Apr 2014;**59**(4):1406-1414. Available from: <http://doi.wiley.com/10.1002/hep.26761> [Accessed: Dec 26, 2016]
- [69] Klair JS, Yang JD, Abdelmalek MF, Guy CD, Gill RM, Yates K, et al. A longer duration of estrogen deficiency increases fibrosis risk among postmenopausal women with non-alcoholic fatty liver disease. *Hepatology* [Internet]. Jul 2016;**64**(1):85-91. Available from: <http://doi.wiley.com/10.1002/hep.28514> [Accessed: Dec 26, 2016]
- [70] Anstee QM, Day CP. The genetics of nonalcoholic fatty liver disease: Spotlight on PNPLA3 and TM6SF2. *Semin Liver Disease* [Internet]. Aug 2015;**35**(3):270-290. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/26378644> [Accessed: Jun 21, 2016]
- [71] Romeo S, Kozlitina J, Xing C, Pertsemlidis A, Cox D, Pennacchio LA, et al. Genetic variation in PNPLA3 confers susceptibility to nonalcoholic fatty liver disease. *Nature Genetics* [Internet]. Dec 2008;**40**(12):1461-1465. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/18820647> [Accessed: Jun 21, 2016]
- [72] Kawaguchi T, Sumida Y, Umemura A, Matsuo K, Takahashi M, Takamura T, et al. Genetic polymorphisms of the human PNPLA3 gene are strongly associated with severity of non-alcoholic fatty liver disease in Japanese. *PLoS One* [Internet]. 2012;**7**(6):e38322. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/22719876> [Accessed: Jun 21, 2016]
- [73] Liu Y-L, Patman GL, Leathart JBS, Piguet A-C, Burt AD, Dufour J-F, et al. Carriage of the PNPLA3 rs738409 C > G polymorphism confers an increased risk of non-alcoholic fatty liver disease associated hepatocellular carcinoma. *Journal of Hepatology* [Internet]. Jul 2014;**61**(1):75-81. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/24607626> [Accessed: Dec 27, 2016]
- [74] Liu Y-L, Reeves HL, Burt AD, Tiniakos D, McPherson S, Leathart JBS, et al. TM6SF2 rs58542926 influences hepatic fibrosis progression in patients with non-alcoholic fatty liver disease. *Nature Communication* [Internet]. 2014;**5**:4309. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/24978903> [Accessed: Jun 22, 2016]
- [75] Sookoian S, Castaño GO, Scian R, Mallardi P, Fernández Gianotti T, Burgueño AL, et al. Genetic variation in transmembrane 6 superfamily member 2 and the risk of nonalcoholic fatty liver disease and histological disease severity. *Hepatology* [Internet]. Feb 2015;**61**(2):515-525. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25302781> [Accessed: Jun 22, 2016]
- [76] Donati B, Dongiovanni P, Romeo S, Meroni M, McCain M, Miele L, et al. MBOAT7 rs641738 variant and hepatocellular carcinoma in non-cirrhotic individuals. *Science*

- Report [Internet]. Dec 3, 2017;7(1):4492. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/28674415> [Accessed: Oct 29, 2017]
- [77] Petta S, Grimaudo S, Cammà C, Cabibi D, Di Marco V, Licata G, et al. IL28B and PNPLA3 polymorphisms affect histological liver damage in patients with non-alcoholic fatty liver disease. *Journal of Hepatology* [Internet]. Jun 2012;56(6):1356-1362. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/22314430> [Accessed: Aug 3, 2015]
- [78] Zoppini G, Fedeli U, Gennaro N, Saugo M, Targher G, Bonora E. Mortality from chronic liver diseases in diabetes. *American Journal of Gastroenterology* [Internet]. Jul 3, 2014;109(7):1020-1025. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/24890439> [Accessed: Dec 26, 2016]
- [79] Vilar-Gomez E, Martinez-Perez Y, Calzadilla-Bertot L, Torres-Gonzalez A, Gra-Oramas B, Gonzalez-Fabian L, et al. Weight loss through lifestyle modification significantly reduces features of nonalcoholic steatohepatitis. *Gastroenterology* [Internet]. Aug 2015;149(2):367-378.e5. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25865049> [Accessed: Feb 8, 2017]
- [80] Williams CD, Stengel J, Asike MI, Torres DM, Shaw J, Contreras M, et al. Prevalence of non-alcoholic fatty liver disease and nonalcoholic steatohepatitis among a largely middle-aged population utilizing ultrasound and liver biopsy: A prospective study. *Gastroenterology* [Internet]. Jan 2011;140(1):124-131. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0016508510014162> [Accessed: Mar 2, 2016]
- [81] Loria P, Marchesini G, Nascimbeni F, Ballestri S, Maurantonio M, Carubbi F, et al. Cardiovascular risk, lipidemic phenotype and steatosis. A comparative analysis of cirrhotic and non-cirrhotic liver disease due to varying etiology. *Atherosclerosis* [Internet]. Jan 2014;232(1):99-109. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/24401223> [Accessed: Jun 4, 2016]
- [82] Jiang ZG, Tapper EB, Connelly MA, Pimentel CFMG, Feldbrügge L, Kim M, et al. Steatohepatitis and liver fibrosis are predicted by the characteristics of very low density lipoprotein in nonalcoholic fatty liver disease. *Liver International* [Internet]. Aug 2016;36(8):1213-1220. Available from: <http://doi.wiley.com/10.1111/liv.13076> [Accessed: Dec 26, 2016]
- [83] Targher G, Bertolini L, Scala L, Cigolini M, Zenari L, Falezza G, et al. Associations between serum 25-hydroxyvitamin D3 concentrations and liver histology in patients with non-alcoholic fatty liver disease. *Nutrition, Metabolism & Cardiovascular Diseases* [Internet]. Sep 2007;17(7):517-524. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0939475306001086> [Accessed: Dec 28, 2016]
- [84] Iruzubieta P, Terán Á, Crespo J, Fábrega E. Vitamin D deficiency in chronic liver disease. *World Journal of Hepatology* [Internet]. Dec 27, 2014;6(12):901. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25544877> [Accessed: Dec 26, 2016]
- [85] Hsu CC, Kowdley KV. The effects of alcohol on other chronic liver diseases. *Clinical Liver Disease* [Internet]. Aug 2016;20(3):581-594. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S1089326116300137> [Accessed: Dec 28, 2016]

- [86] Ajmera VH, Terrault NA, Harrison SA. Is moderate alcohol use in non-alcoholic fatty liver disease good or bad? a critical review. *Hepatology* [Internet]. Jun 2017;**65**(6): 2090-2099. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/28100008> [Accessed: Feb 18, 2017]
- [87] 2015-2020 Dietary Guidelines: health.gov [Internet]. 2015. Available from: <https://health.gov/dietaryguidelines/2015/guidelines/> [Accessed: Feb 18, 2017]
- [88] Kang L, Sebastian BM, Pritchard MT, Pratt BT, Previs SF, Nagy LE. Chronic ethanol-induced insulin resistance is associated with macrophage infiltration into adipose tissue and altered expression of adipocytokines. *Alcoholism: Clinical and Experimental Research* [Internet]. Sep 2007;**31**(9):1581-1588. Available from: <http://doi.wiley.com/10.1111/j.1530-0277.2007.00452.x> [Accessed: Feb 18, 2017]
- [89] Conigrave KM, Hu BF, Camargo CA, Stampfer MJ, Willett WC, Rimm EB. A prospective study of drinking patterns in relation to risk of type 2 diabetes among men. *Diabetes* [Internet]. Oct 2001;**50**(10):2390-2395. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/11574424> [Accessed: Feb 18, 2017]
- [90] Pischon T, Girman CJ, Rifai N, Hotamisligil GS, Rimm EB. Association between dietary factors and plasma adiponectin concentrations in men. *American Journal of Clinical Nutrition* [Internet]. Apr 2005;**81**(4):780-786. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/15817852> [Accessed: Feb 18, 2017]
- [91] Davies MJ, Baer DJ, Judd JT, Brown ED, Campbell WS, Taylor PR. Effects of moderate alcohol intake on fasting insulin and glucose concentrations and insulin sensitivity in postmenopausal women: A randomized controlled trial. *Journal of the American Medical Association* [Internet]. May 15, 2002;**287**(19):2559-2562. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/12020337> [Accessed: Feb 18, 2017]
- [92] Dunn W, Sanyal AJ, Brunt EM, Unalp-Arida A, Donohue M, McCullough AJ, et al. Modest alcohol consumption is associated with decreased prevalence of steatohepatitis in patients with non-alcoholic fatty liver disease (NAFLD). *Journal of Hepatology* [Internet]. Aug 2012;**57**(2):384-391. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0168827812002796> [Accessed: Feb 18, 2017]
- [93] Kwon HK, Greenson JK, Conjeevaram HS. Effect of lifetime alcohol consumption on the histological severity of non-alcoholic fatty liver disease. *Liver International* [Internet]. Jan 2014;**34**(1):129-135. Available from: <http://doi.wiley.com/10.1111/liv.12230> [Accessed: Feb 18, 2017]
- [94] Ascha MS, Hanouneh IA, Lopez R, Tamimi TA-R, Feldstein AF, Zein NN. The incidence and risk factors of hepatocellular carcinoma in patients with nonalcoholic steatohepatitis. *Hepatology* [Internet]. Jun 2010;**51**(6):1972-1978. Available from: <http://doi.wiley.com/10.1002/hep.23527> [Accessed: Dec 27, 2016]
- [95] Wieckowska A, Zein NN, Yerian LM, Lopez AR, McCullough AJ, Feldstein AE. In vivo assessment of liver cell apoptosis as a novel biomarker of disease severity in nonalcoholic fatty liver disease. *Hepatology* [Internet]. Jul 2006;**44**(1):27-33. Available from: <http://doi.wiley.com/10.1002/hep.21223> [Accessed: Feb 18, 2017]

- [96] Feldstein AE, Wieckowska A, Lopez AR, Liu Y-C, Zein NN, McCullough AJ. Cytokeratin-18 fragment levels as noninvasive biomarkers for nonalcoholic steatohepatitis: A multicenter validation study. *Hepatology* [Internet]. Oct 2009;**50**(4):1072-1078. Available from: <http://doi.wiley.com/10.1002/hep.23050> [Accessed: Feb 18, 2017]
- [97] Feldstein AE, Alkhouri N, De Vito R, Alisi A, Lopez R, Nobili V. Serum cytokeratin-18 fragment levels are useful biomarkers for nonalcoholic steatohepatitis in children. *The American Journal of Gastroenterology* [Internet]. Sep 11, 2013;**108**(9):1526-1531. Available from: <http://www.nature.com/doifinder/10.1038/ajg.2013.168> [Accessed: Feb 18, 2017]
- [98] Cusi K, Chang Z, Harrison S, Lomonaco R, Bril F, Orsak B, et al. Limited value of plasma cytokeratin-18 as a biomarker for NASH and fibrosis in patients with non-alcoholic fatty liver disease. *Journal of Hepatology* [Internet]. Jan 2014;**60**(1):167-174. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0168827813005965> [Accessed: Feb 18, 2017]
- [99] Yesilova Z, Yaman H, Oktenli C, Ozcan A, Uygun A, Cakir E, et al. Systemic markers of lipid peroxidation and antioxidants in patients with nonalcoholic fatty liver disease. *The American Journal of Gastroenterology* [Internet]. Apr 2005;**100**(4):850-855. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/15784031> [Accessed: Oct 23, 2017]
- [100] Chalasani N, Deeg MA, Crabb DW. Systemic levels of lipid peroxidation and its metabolic and dietary correlates in patients with nonalcoholic steatohepatitis. *The American Journal of Gastroenterology* [Internet]. Aug 2004;**99**(8):1497-1502. Available from: <http://www.nature.com/doifinder/10.1111/j.1572-0241.2004.30159.x> [Accessed: Oct 23, 2017]
- [101] Jayakumar S, Harrison SA, Loomba R. Noninvasive markers of fibrosis and inflammation in nonalcoholic fatty liver disease. *Current Hepatology Reports* [Internet]. Jun 21, 2016;**15**(2):86-95. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/27795938> [Accessed: Feb 18, 2017]
- [102] Dixon JB, Bhathal PS, O'Brien PE. Nonalcoholic fatty liver disease: Predictors of non-alcoholic steatohepatitis and liver fibrosis in the severely obese. *Gastroenterology* [Internet]. Jul 2001;**121**(1):91-100. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/11438497> [Accessed: Oct 30, 2017]
- [103] Poynard T, Ratziu V, Charlotte F, Messous D, Munteanu M, Imbert-Bismut F, et al. Diagnostic value of biochemical markers (NashTest) for the prediction of non alcoholic steato hepatitis in patients with non-alcoholic fatty liver disease. *BMC Gastroenterology* [Internet]. Nov 10, 2006;**6**(1):34. Available from: <http://bmcgastroenterol.biomedcentral.com/articles/10.1186/1471-230X-6-34> [Accessed: Oct 30, 2017]
- [104] Hyysalo J, Männistö VT, Zhou Y, Arola J, Kärjä V, Leivonen M, et al. A population-based study on the prevalence of NASH using scores validated against liver histology. *Journal of Hepatology* [Internet]. Apr 2014;**60**(4):839-846. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0168827813008738> [Accessed: Oct 30, 2017]
- [105] Anty R, Iannelli A, Patouraux S, Bonnafous S, Lavallard VJ, Senni-Buratti M, et al. A new composite model including metabolic syndrome, alanine aminotransferase and

- cytokeratin-18 for the diagnosis of non-alcoholic steatohepatitis in morbidly obese patients. *Alimentary Pharmacology & Therapeutics* [Internet]. Dec 2010;**32**(11-12):1315-1322. Available from: <http://doi.wiley.com/10.1111/j.1365-2036.2010.04480.x> [Accessed: Oct 30, 2017]
- [106] Younossi ZM, Page S, Rafiq N, Bireddi A, Stepanova M, Hossain N, et al. A biomarker panel for non-alcoholic steatohepatitis (NASH) and NASH-related fibrosis. *Obesity Surgery* [Internet]. Apr 8, 2011;**21**(4):431-439. Available from: <http://link.springer.com/10.1007/s11695-010-0204-1> [Accessed: Oct 30, 2017]
- [107] Feldstein AE, Lopez R, Tamimi TA-R, Yerian L, Chung Y-M, Berk M, et al. Mass spectrometric profiling of oxidized lipid products in human nonalcoholic fatty liver disease and nonalcoholic steatohepatitis. *Hepatology* [Internet]. Oct 2010;**51**(10):3046-3054. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/20631297> [Accessed: Oct 30, 2017]
- [108] Bell LN, Theodorakis JL, Vuppalanchi R, Saxena R, Bemis KG, Wang M, et al. Serum proteomics and biomarker discovery across the spectrum of nonalcoholic fatty liver disease. *Hepatology* [Internet]. Jan 2010;**51**(1):111-120. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/19885878> [Accessed: Oct 29, 2017]
- [109] Barr J, Vázquez-Chantada M, Alonso C, Pérez-Cormenzana M, Mayo R, Galán A, et al. Liquid chromatography–mass spectrometry-based parallel metabolic profiling of human and mouse model serum reveals putative biomarkers associated with the progression of nonalcoholic fatty liver disease. *Journal of Proteome Research* [Internet]. Sep 3, 2010;**9**(9):4501-4512. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/20684516> [Accessed: Feb 18, 2017]
- [110] Iruzubieta P, Arias-Loste MT, Barbier-Torres L, Martinez-Chantar ML, Crespo J. The need for biomarkers in diagnosis and prognosis of drug-induced liver disease: Does metabolomics have any role? *BioMed Research International* [Internet]. 2015;**2015**:1-8. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/26824035> [Accessed: Feb 18, 2017]
- [111] Safaei A, Arefi Oskouie A, Mohebbi SR, Rezaei-Tavirani M, Mahboubi M, Peyvandi M, et al. Metabolomic analysis of human cirrhosis, hepatocellular carcinoma, non-alcoholic fatty liver disease and non-alcoholic steatohepatitis diseases. *Gastroenterology and Hepatology from Bed to Bench* [Internet]. 2016;**9**(3):158-173. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/27458508> [Accessed: Feb 18, 2017]
- [112] Barr J, Caballería J, Martínez-Arranz I, Domínguez-Díez A, Alonso C, Muntané J, et al. Obesity-dependent metabolic signatures associated with nonalcoholic fatty liver disease progression. *Journal of Proteome Research* [Internet]. Apr 6, 2012;**11**(4):2521-2532. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/22364559> [Accessed: Feb 18, 2017]
- [113] Alonso C, Fernández-Ramos D, Varela-Rey M, Martínez-Arranz I, Navasa N, Van Liempd SM, et al. Metabolomic identification of subtypes of nonalcoholic steatohepatitis. *Gastroenterology* [Internet]. 2017;**152**(6):1449-1461.e7. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0016508517300720> [Accessed: Mar 5, 2017]

- [114] Tan Y, Ge G, Pan T, Wen D, Gan J. A pilot study of serum microRNAs panel as potential biomarkers for diagnosis of nonalcoholic fatty liver disease. *PLoS One* [Internet]. Aug 20, 2014;**9**(8):e105192. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25141008> [Accessed: Oct 29, 2017]
- [115] Shah AG, Lydecker A, Murray K, Tetri BN, Contos MJ, Sanyal AJ, et al. Comparison of noninvasive markers of fibrosis in patients with nonalcoholic fatty liver disease. *Clinical Gastroenterology and Hepatology* [Internet]. Oct 2009;**7**(10):1104-1112. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/19523535> [Accessed: Oct 26, 2017]
- [116] Musso G, Gambino R, Cassader M, Pagano G. Meta-analysis: Natural history of non-alcoholic fatty liver disease (NAFLD) and diagnostic accuracy of non-invasive tests for liver disease severity. *Annals of Medicine* [Internet]. Dec 2, 2011;**43**(8):617-649. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/21039302> [Accessed: Feb 18, 2017]
- [117] Sandrin L, Fourquet B, Hasquenoph J-M, Yon S, Fournier C, Mal F, et al. Transient elastography: A new noninvasive method for assessment of hepatic fibrosis. *Ultrasound in Medicine and Biology* [Internet]. Dec 2003;**29**(12):1705-1713. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/14698338> [Accessed: Feb 19, 2017]
- [118] Petta S, Di Marco V, Cammà C, Butera G, Cabibi D, Craxì A. Reliability of liver stiffness measurement in non-alcoholic fatty liver disease: The effects of body mass index. *Aliment Pharmacol Ther* [Internet]. Jun 2011;**33**(12):1350-1360. Available from: <http://doi.wiley.com/10.1111/j.1365-2036.2011.04668.x> [Accessed: Feb 19, 2017]
- [119] Myers RP, Pomier-Layrargues G, Kirsch R, Pollett A, Duarte-Rojo A, Wong D, et al. Feasibility and diagnostic performance of the FibroScan XL probe for liver stiffness measurement in overweight and obese patients. *Hepatology* [Internet]. Jan 2012;**55**(1):199-208. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/21898479> [Accessed: Feb 19, 2017]
- [120] Castera L, Pawlotsky J-M. Noninvasive diagnosis of liver fibrosis in patients with chronic hepatitis C. *MedGenMed* [Internet]. Nov 9, 2005;**7**(4):39. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/16614661> [Accessed: Feb 19, 2017]
- [121] Pathik P, Ravindra S, Ajay C, Prasad B, Jatin P, Prabha S. Fibroscan versus simple noninvasive screening tools in predicting fibrosis in high-risk nonalcoholic fatty liver disease patients from Western India. *Annals of Gastroenterology* [Internet]. Apr-Jun 2017;**28**(2):281-286. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25830783>
- [122] Yoneda M, Fujita K, Inamori M, Nakajima A, Tamano M, Hiraishi H, et al. Transient elastography in patients with non-alcoholic fatty liver disease (NAFLD). *Gut* [Internet]. Apr 5, 2007;**56**(9):1330-1331. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/17470477> [Accessed: Feb 19, 2017]
- [123] Cassinotto C, Boursier J, de Lédinghen V, Lebigot J, Lapuyade B, Cales P, et al. Liver stiffness in nonalcoholic fatty liver disease: A comparison of supersonic shear imaging, FibroScan, and ARFI with liver biopsy. *Hepatology* [Internet]. Jun 2016;**63**(6):1817-1827. Available from: <http://doi.wiley.com/10.1002/hep.28394> [Accessed: Feb 19, 2017]

- [124] Lupsor M, Badea R, Stefanescu H, Grigorescu M, Serban A, Radu C, et al. Performance of unidimensional transient elastography in staging non-alcoholic steatohepatitis. *Journal of Gastrointestinal and Liver Diseases* [Internet]. Mar 2010;**19**(1):53-60. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/20361076> [Accessed: Feb 19, 2017]
- [125] Yoneda M, Yoneda M, Mawatari H, Fujita K, Endo H, Iida H, et al. Noninvasive assessment of liver fibrosis by measurement of stiffness in patients with nonalcoholic fatty liver disease (NAFLD). *Digestive and Liver Disease* [Internet]. May 2008;**40**(5):371-378. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/18083083> [Accessed: Feb 19, 2017]
- [126] Kumar R, Rastogi A, Sharma MK, Bhatia V, Tyagi P, Sharma P, et al. Liver stiffness measurements in patients with different stages of nonalcoholic fatty liver disease: Diagnostic performance and clinicopathological correlation. *Digestive Diseases and Sciences* [Internet]. Jan 12, 2013;**58**(1):265-274. Available from: <http://link.springer.com/10.1007/s10620-012-2306-1> [Accessed: Feb 19, 2017]
- [127] Lin X-F, Shi K-Q, You J, Liu W-Y, Luo Y-W, Wu F-L, et al. Increased risk of colorectal malignant neoplasm in patients with nonalcoholic fatty liver disease: A large study. *Molecular Biology Reports* [Internet]. May 22, 2014;**41**(5):2989-2997. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/24449368> [Accessed: Mar 7, 2017]
- [128] Wong VW-S, Vergniol J, Wong GL-H, Foucher J, Chan HL-Y, Le Bail B, et al. Diagnosis of fibrosis and cirrhosis using liver stiffness measurement in nonalcoholic fatty liver disease. *Hepatology* [Internet]. Feb 2010;**51**(2):454-462. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/20101745> [Accessed: Feb 19, 2017]
- [129] Nobili V, Vizzutti F, Arena U, Abraldes JG, Marra F, Pietrobattista A, et al. Accuracy and reproducibility of transient elastography for the diagnosis of fibrosis in pediatric nonalcoholic steatohepatitis. *Hepatology* [Internet]. Aug 2008;**48**(2):442-448. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/18563842> [Accessed: Mar 8, 2017]
- [130] de Lédinghen V, Wong VW-S, Vergniol J, Wong GL-H, Foucher J, Chu SH-T, et al. Diagnosis of liver fibrosis and cirrhosis using liver stiffness measurement: Comparison between M and XL probe of FibroScan®. *Journal of Hepatology* [Internet]. Apr 2012;**56**(4):833-839. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/22173167> [Accessed: Feb 19, 2017]
- [131] Wong VW-S, Vergniol J, Wong GL-H, Foucher J, Chan AW-H, Chermak F, et al. Liver stiffness measurement using XL probe in patients with nonalcoholic fatty liver disease. *The American Journal of Gastroenterology* [Internet]. Dec 2, 2012;**107**(12):1862-1871. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/23032979> [Accessed: Feb 19, 2017]
- [132] Palmeri ML, Wang MH, Rouze NC, Abdelmalek MF, Guy CD, Moser B, et al. Noninvasive evaluation of hepatic fibrosis using acoustic radiation force-based shear stiffness in patients with nonalcoholic fatty liver disease. *Journal of Hepatology* [Internet]. Sep 2011;**55**(3):666-672. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0168827811000079> [Accessed: Feb 19, 2017]
- [133] Liu H, Fu J, Hong R, Liu L, Li F. Acoustic radiation force impulse elastography for the non-invasive evaluation of hepatic fibrosis in non-alcoholic fatty liver disease patients:

- A systematic review & meta-analysis. PLoS One [Internet]. Jul 1, 2015;**10**(7):e0127782. Available from: <http://dx.plos.org/10.1371/journal.pone.0127782> [Accessed: Feb 19, 2017]
- [134] Talwalkar JA, Yin M, Fidler JL, Sanderson SO, Kamath PS, Ehman RL. Magnetic resonance imaging of hepatic fibrosis: Emerging clinical applications. *Hepatology* [Internet]. Dec 27, 2007;**47**(1):332-342. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/18161879> [Accessed: Feb 19, 2017]
- [135] Yin M, Glaser KJ, Talwalkar JA, Chen J, Manduca A, Ehman RL. Hepatic MR elastography: Clinical performance in a series of 1377 consecutive examinations. *Radiology* [Internet]. Jan 2016;**278**(1):114-124. Available from: <http://pubs.rsna.org/doi/10.1148/radiol.2015142141> [Accessed: Feb 19, 2017]
- [136] Gallego-Durán R, Cerro-Salido P, Gomez-Gonzalez E, Pareja MJ, Ampuero J, Rico MC, et al. Imaging biomarkers for steatohepatitis and fibrosis detection in non-alcoholic fatty liver disease. *Scientific Reports* [Internet]. Aug 12, 2016;**6**:31421. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/27514671> [Accessed: Dec 26, 2016]
- [137] Pavlides M, Banerjee R, Tunnicliffe EM, Kelly C, Collier J, Wang LM, et al. Multiparametric magnetic resonance imaging for the assessment of non-alcoholic fatty liver disease severity. *Liver International* [Internet]. Jul 2017;**37**(7):1065-1073. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/27778429> [Accessed: Oct 29, 2017]
- [138] Evans CDJ, Oien KA, MacSween RNM, Mills PR. Non-alcoholic steatohepatitis: A common cause of progressive chronic liver injury? *Journal of Clinical Pathology* [Internet]. Sep 2002;**55**(9):689-692. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/12195000> [Accessed: Dec 26, 2016]
- [139] Harrison SA, Torgerson S, Hayashi PH. The natural history of nonalcoholic fatty liver disease: A clinical histopathological study. *The American Journal of Gastroenterology* [Internet]. Sep 2003;**98**(9):2042-2047. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/14499785> [Accessed: Dec 26, 2016]
- [140] Fassio E, Alvarez E, Domínguez N, Landeira G, Longo C. Natural history of nonalcoholic steatohepatitis: A longitudinal study of repeat liver biopsies. *Hepatology* [Internet]. Oct 2004;**40**(4):820-826. Available from: <http://doi.wiley.com/10.1002/hep.20410> [Accessed: Dec 26, 2016]
- [141] Ekstedt M, Franzén LE, Mathiesen UL, Thorelius L, Holmqvist M, Bodemar G, et al. Long-term follow-up of patients with NAFLD and elevated liver enzymes. *Hepatology* [Internet]. Oct 2006;**44**(4):865-873. Available from: <http://doi.wiley.com/10.1002/hep.21327> [Accessed: Dec 26, 2016]
- [142] Hamaguchi E, Takamura T, Sakurai M, Mizukoshi E, Zen Y, Takeshita Y, et al. Histological course of nonalcoholic fatty liver disease in Japanese patients: Tight glycemic control, rather than weight reduction, ameliorates liver fibrosis. *Diabetes Care* [Internet]. Feb 1, 2010;**33**(2):284-286. Available from: <http://care.diabetesjournals.org/cgi/doi/10.2337/dc09-0148> [Accessed: Dec 26, 2016]

- [143] Wong VW-S, Wong GL-H, Choi PC-L, Chan AW-H, Li MK-P, Chan H-Y, et al. Disease progression of non-alcoholic fatty liver disease: A prospective study with paired liver biopsies at 3 years. *Gut* [Internet]. Jul 1, 2010;**59**(7):969-974. Available from: <http://gut.bmj.com/cgi/doi/10.1136/gut.2009.205088> [Accessed: Dec 26, 2016]
- [144] Chan W-K, Ida NH, Cheah P-L, Goh K-L. Progression of liver disease in non-alcoholic fatty liver disease: A prospective clinicopathological follow-up study. *Journal of Digestive Diseases* [Internet]. Oct 2014;**15**(10):545-552. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25060399> [Accessed: Dec 26, 2016]
- [145] Fracanzani AL, Valenti L, Bugianesi E, Andreoletti M, Colli A, Vanni E, et al. Risk of severe liver disease in nonalcoholic fatty liver disease with normal aminotransferase levels: A role for insulin resistance and diabetes. *Hepatology* [Internet]. Sep 2008;**48**(3):792-798. Available from: <http://doi.wiley.com/10.1002/hep.22429> [Accessed: Feb 20, 2017]
- [146] Goh GB-B, Pagadala MR, Dasarathy J, Unalp-Arida A, Sargent R, Hawkins C, et al. Clinical spectrum of non-alcoholic fatty liver disease in diabetic and non-diabetic patients. *BBA Clinical* [Internet]. Jun 2015;**3**:141-145. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S221464741400018X> [Accessed: Feb 20, 2017]
- [147] Bazick J, Donithan M, Neuschwander-Tetri BA, Kleiner D, Brunt EM, Wilson L, et al. Clinical model for NASH and advanced fibrosis in adult patients with diabetes and NAFLD: Guidelines for referral in NAFLD. *Diabetes Care* [Internet]. Jul 2015;**38**(7):1347-1355. Available from: <http://care.diabetesjournals.org/lookup/doi/10.2337/dc14-1239> [Accessed: Feb 20, 2017]
- [148] Motamed N, Sohrabi M, Ajdarkosh H, Hemmasi G, Maadi M, Sayeedian FS, et al. Fatty liver index vs waist circumference for predicting non-alcoholic fatty liver disease. *World Journal of Gastroenterology* [Internet]. Mar 14, 2016;**22**(10):3023-3030. Available from: <http://www.wjgnet.com/1007-9327/full/v22/i10/3023.htm> [Accessed: Feb 21, 2017]
- [149] DeFilippis AP, Blaha MJ, Martin SS, Reed RM, Jones SR, Nasir K, et al. Nonalcoholic fatty liver disease and serum lipoproteins: The multi-ethnic study of atherosclerosis. *Atherosclerosis* [Internet]. Apr 2013;**227**(2):429-436. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/23419204> [Accessed: Feb 21, 2017]
- [150] Marchesini G, Marzocchi R. Metabolic syndrome and NASH. *Clinical Liver Disease* [Internet]. Feb 2007;**11**(1):105-117, ix. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/17544974> [Accessed: Aug 3, 2015]
- [151] Ekstedt M, Hagström H, Nasr P, Fredrikson M, Stål P, Kechagias S, et al. Fibrosis stage is the strongest predictor for disease-specific mortality in NAFLD after up to 33 years of follow-up. *Hepatology* [Internet]. May 2015;**61**(5):1547-1554. Available from: <http://doi.wiley.com/10.1002/hep.27368> [Accessed: Dec 26, 2016]
- [152] Sanyal A, Poklepovic A, Moyneur E, Barghout V. Population-based risk factors and resource utilization for HCC: US perspective. *Current Medical Research and Opinion*

- [Internet]. Sep 29, 2010;**26**(9):2183-2191. Available from: <http://www.tandfonline.com/doi/full/10.1185/03007995.2010.506375> [Accessed: Dec 27, 2016]
- [153] Maeda S, Kamata H, Luo J-L, Leffert H, Karin M. IKK β couples hepatocyte death to cytokine-driven compensatory proliferation that promotes chemical hepatocarcinogenesis. *Cell* [Internet]. Jul 1, 2005;**121**(7):977-990. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0092867405003946> [Accessed: Feb 21, 2017]
- [154] Adams LA, Lymp JF, St Sauver J, Sanderson SO, Lindor KD, Feldstein A, et al. The natural history of nonalcoholic fatty liver disease: A population-based cohort study. *Gastroenterology* [Internet]. Jul 2005;**129**(1):113-121. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/16012941> [Accessed: May 3, 2016]
- [155] Chitturi S, Abeygunasekera S, Farrell GC, Holmes-Walker J, Hui JM, Fung C, et al. NASH and insulin resistance: Insulin hypersecretion and specific association with the insulin resistance syndrome. *Hepatology* [Internet]. Feb 2002;**35**(2):373-379. Available from: <http://doi.wiley.com/10.1053/jhep.2002.30692> [Accessed: Feb 21, 2017]
- [156] Pagano G, Pacini G, Musso G, Gambino R, Mecca F, Depetris N, et al. Nonalcoholic steatohepatitis, insulin resistance, and metabolic syndrome: Further evidence for an etiologic association. *Hepatology* [Internet]. Feb 2002;**35**(2):367-372. Available from: <http://doi.wiley.com/10.1053/jhep.2002.30690> [Accessed: Feb 21, 2017]
- [157] DeFronzo RA, Tobin JD, Andres R. Glucose clamp technique: A method for quantifying insulin secretion and resistance. *American Journal of Physiology* [Internet]. Sep 1979;**237**(3):E214-E223. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/382871> [Accessed: Dec 27, 2016]
- [158] Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: Insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia* [Internet]. Jul 1985;**28**(7):412-419. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/3899825> [Accessed: Dec 27, 2016]
- [159] Armstrong MJ, Gaunt P, Aithal GP, Barton D, Hull D, Parker R, et al. Liraglutide safety and efficacy in patients with non-alcoholic steatohepatitis (LEAN): A multicentre, double-blind, randomised, placebo-controlled phase 2 study. *Lancet (London, England)* [Internet]. Feb 13, 2016;**387**(10019):679-690. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S014067361500803X> [Accessed: Mar 6, 2017]
- [160] Belfort R, Harrison SA, Brown K, Darland C, Finch J, Hardies J, et al. A placebo-controlled trial of pioglitazone in subjects with nonalcoholic steatohepatitis. *The New England Journal of Medicine* [Internet]. Nov 30, 2006;**355**(22):2297-2307. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/17135584> [Accessed: Mar 6, 2017]
- [161] Cusi K, Orsak B, Bril F, Lomonaco R, Hecht J, Ortiz-Lopez C, et al. Long-term pioglitazone treatment for patients with nonalcoholic steatohepatitis and prediabetes or Type 2 diabetes mellitus. *Annals of Internal Medicine* [Internet]. Sep 6, 2016;**165**(5):305. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/27322798> [Accessed: Mar 6, 2017]

- [162] Sanyal AJ, Chalasani N, Kowdley KV, McCullough A, Diehl AM, Bass NM, et al. Pioglitazone, vitamin E, or placebo for nonalcoholic steatohepatitis. *The New England Journal of Medicine* [Internet]. May 6, 2010;**362**(18):1675-1685. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/20427778> [Accessed: Mar 6, 2017]
- [163] Aithal GP, Thomas JA, Kaye PV, Lawson A, Ryder SD, Spendlove I, et al. Randomized, placebo-controlled trial of pioglitazone in nondiabetic subjects with nonalcoholic steatohepatitis. *Gastroenterology* [Internet]. Oct 2008;**135**(4):1176-1184. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0016508508011013> [Accessed: Mar 6, 2017]
- [164] García-Monzón C, Lo Iacono O, Crespo J, Romero-Gómez M, García-Samaniego J, Fernández-Bermejo M, et al. Increased soluble CD36 is linked to advanced steatosis in nonalcoholic fatty liver disease. *European Journal of Clinical Investigation* [Internet]. Jan 2014;**44**(1):65-73. Available from: <http://doi.wiley.com/10.1111/eci.12192> [Accessed: Dec 26, 2016]
- [165] Fernández-Friera L, Ibáñez B, Fuster V. Imaging subclinical atherosclerosis: Is it ready for prime time? A review. *Journal of Cardiovascular Translational Research* [Internet]. Oct 14, 2014;**7**(7):623-634. Available from: <http://link.springer.com/10.1007/s12265-014-9582-4> [Accessed: Dec 27, 2016]
- [166] Røsbjerg TE, Aagnes B, Hjartaker A, Langseth H, Bray FI, Larsen IK. Body mass index, physical activity, and colorectal cancer by anatomical subsites: A systematic review and meta-analysis of cohort studies. *European Journal of Cancer Prevention* [Internet]. Nov 2013;**22**(6):492-505. Available from: <http://content.wkhealth.com/linkback/openurl?sid=WKPTLP:landingpage&an=00008469-201311000-00002> [Accessed: Mar 7, 2017]
- [167] Kaminski MF, Polkowski M, Kraszewska E, Rupinski M, Butruk E, Regula J. A score to estimate the likelihood of detecting advanced colorectal neoplasia at colonoscopy. *Gut* [Internet]. Jul 2014;**63**(7):1112-1119. Available from: <http://gut.bmj.com/lookup/doi/10.1136/gutjnl-2013-304965> [Accessed: Mar 7, 2017]
- [168] Peeters PJHL, Bazelier MT, Leufkens HGM, de Vries F, De Bruin ML. The risk of colorectal cancer in patients with Type 2 diabetes: Associations with treatment stage and obesity. *Diabetes Care* [Internet]. Mar 2015;**38**(3):495-502. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25552419> [Accessed: Mar 7, 2017]
- [169] Aune D, Greenwood DC, Chan DSM, Vieira R, Vieira AR, Navarro Rosenblatt DA, et al. Body mass index, abdominal fatness and pancreatic cancer risk: A systematic review and non-linear dose-response meta-analysis of prospective studies. *Annals of Oncology* [Internet]. Apr 1, 2012;**23**(4):843-852. Available from: <https://academic.oup.com/annonc/article-lookup/doi/10.1093/annonc/mdr398> [Accessed: Mar 7, 2017]
- [170] Rose DP, Vona-Davis L. Biochemical and molecular mechanisms for the association between obesity, chronic inflammation, and breast cancer. *Biofactors* [Internet]. Jan 2014;**40**(1):1-12. Available from: <http://doi.wiley.com/10.1002/biof.1109> [Accessed: Mar 7, 2017]

- [171] Schmid D, Ricci C, Behrens G, Leitzmann MF. Adiposity and risk of thyroid cancer: A systematic review and meta-analysis. *Obesity Reviews* [Internet]. Dec 2015;**16**(12):1042-1054. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/26365757> [Accessed: Mar 7, 2017]
- [172] Sørensen HT, Møllekjaer L, Jepsen P, Thulstrup AM, Baron J, Olsen JH, et al. Risk of cancer in patients hospitalized with fatty liver: A Danish cohort study. *Journal of Clinical Gastroenterology* [Internet]. Apr 2003;**36**(4):356-359. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/12642745> [Accessed: Mar 7, 2017]
- [173] Bilici A, Özgüroğlu M, Mihmanlı I, Turna H, Adaletli I. A case-control study of non-alcoholic fatty liver disease in breast cancer. *Medical Oncology* [Internet]. 2007;**24**(4):367-371. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/17917083> [Accessed: Mar 7, 2017]
- [174] Stadlmayr A, Aigner E, Steger B, Scharinger L, Lederer D, Mayr A, et al. Nonalcoholic fatty liver disease: An independent risk factor for colorectal neoplasia. *Journal of Internal Medicine* [Internet]. Jul 2011;**270**(1):41-49. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/21414047> [Accessed: Mar 7, 2017]
- [175] Wong VW-S, Wong GL-H, Tsang SW-C, Fan T, Chu WC-W, Woo J, et al. High prevalence of colorectal neoplasm in patients with non-alcoholic steatohepatitis. *Gut* [Internet]. Jun 1, 2011;**60**(6):829-836. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/21339204> [Accessed: Dec 27, 2016]
- [176] Hwang ST, Cho YK, Park JH, Kim HJ, Park II D, Sohn II C, et al. Relationship of non-alcoholic fatty liver disease to colorectal adenomatous polyps. *Journal of Gastroenterology and Hepatology* [Internet]. Mar 2010;**25**(3):562-567. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/20074156> [Accessed: Mar 7, 2017]
- [177] Kim S, Keku TO, Martin C, Galanko J, Woosley JT, Schroeder JC, et al. Circulating levels of inflammatory cytokines and risk of colorectal adenomas. *Cancer Research* [Internet]. Jan 1, 2008;**68**(1):323-328. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/18172326> [Accessed: Mar 7, 2017]
- [178] Machado MV, Gonçalves S, Carepa F, Coutinho J, Costa A, Cortez-Pinto H. Impaired renal function in morbid obese patients with nonalcoholic fatty liver disease. *Liver International* [Internet]. Feb 2012;**32**(2):241-248. Available from: <http://doi.wiley.com/10.1111/j.1478-3231.2011.02623.x> [Accessed: Feb 21, 2017]
- [179] Targher G, Chonchol MB, Byrne CD. CKD and nonalcoholic fatty liver disease. *American Journal of Kidney Diseases* [Internet]. Oct 2014;**64**(4):638-652. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25085644> [Accessed: Feb 21, 2017]
- [180] Choudhary NS, Saraf N, Kumar N, Rai R, Saigal S, Gautam D, et al. Nonalcoholic fatty liver is not associated with incident chronic kidney disease. *European Journal of Gastroenterology & Hepatology* [Internet]. Dec 2015;**28**(4):1. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/26636408> [Accessed: Feb 21, 2017]

- [181] Musso G, Gambino R, Tabibian JH, Ekstedt M, Kechagias S, Hamaguchi M, et al. Association of non-alcoholic fatty liver disease with chronic kidney disease: A systematic review and meta-analysis. *PLOS Medicine* [Internet]. Jul 22, 2014;**11**(7):e1001680. Available from: <http://dx.plos.org/10.1371/journal.pmed.1001680> [Accessed: Feb 21, 2017]
- [182] Musso G, Cassader M, Olivetti C, Rosina F, Carbone G, Gambino R. Association of obstructive sleep apnoea with the presence and severity of non-alcoholic fatty liver disease. A systematic review and meta-analysis. *Obesity Reviews* [Internet]. May 2013;**14**(5):417-431. Available from: <http://doi.wiley.com/10.1111/obr.12020> [Accessed: Feb 21, 2017]
- [183] Minville C, Hilleret M-N, Tamisier R, Aron-Wisnewsky J, Clement K, Trocme C, et al. Nonalcoholic fatty liver disease, nocturnal hypoxia, and endothelial function in patients with sleep apnea. *Chest* [Internet]. Mar 1, 2014;**145**(3):525-533. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0012369215343646> [Accessed: Feb 21, 2017]
- [184] Sookoian S, Pirola CJ. Obstructive sleep apnea is associated with fatty liver and abnormal liver enzymes: A meta-analysis. *Obesity Surgery* [Internet]. Nov 7, 2013;**23**(11):1815-1825. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/23740153> [Accessed: Jun 4, 2016]
- [185] Drager LF, Jun JC, Polotsky VY. Metabolic consequences of intermittent hypoxia: Relevance to obstructive sleep apnea. *Best Practice & Research: Clinical Endocrinology & Metabolism* [Internet]. Oct 2010;**24**(5):843-851. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S1521690X10001119> [Accessed: Feb 21, 2017]
- [186] Targher G, Lonardo A, Rossini M. Nonalcoholic fatty liver disease and decreased bone mineral density: Is there a link? *Journal of Endocrinological Investigation* [Internet]. Aug 24, 2015;**38**(8):817-825. Available from: <http://link.springer.com/10.1007/s40618-015-0315-6> [Accessed: Dec 27, 2016]
- [187] Miele L, Vallone S, Cefalo C, La Torre G, Di Stasi C, Vecchio FM, et al. Prevalence, characteristics and severity of non-alcoholic fatty liver disease in patients with chronic plaque psoriasis. *Journal of Hepatology* [Internet]. Oct 2009;**51**(4):778-786. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/19664838> [Accessed: Dec 27, 2016]
- [188] Cerda C, Pérez-Ayuso RM, Riquelme A, Soza A, Villaseca P, Sir-Petermann T, et al. Nonalcoholic fatty liver disease in women with polycystic ovary syndrome. *Journal of Hepatology* [Internet]. Sep 2007;**47**(3):412-417. Available from: <http://linkinghub.elsevier.com/retrieve/pii/S0168827807002899> [Accessed: Dec 27, 2016]
- [189] Xu L, Ma H, Miao M, Li Y. Impact of subclinical hypothyroidism on the development of non-alcoholic fatty liver disease: A prospective case-control study. *Journal of Hepatology* [Internet]. Nov 2012;**57**(5):1153-1154. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/22940010> [Accessed: Jun 4, 2016]
- [190] Adams LA, Feldstein A, Lindor KD, Angulo P. Nonalcoholic fatty liver disease among patients with hypothalamic and pituitary dysfunction. *Hepatology* [Internet]. Apr

2004;**39**(4):909-914. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/15057893> [Accessed: Jun 4, 2016]

- [191] Barbonetti A, Caterina Vassallo MR, Cotugno M, Felzani G, Francavilla S, Francavilla F. Low testosterone and non-alcoholic fatty liver disease: Evidence for their independent association in men with chronic spinal cord injury. *The Journal of Spinal Cord Medicine* [Internet]. Feb 25, 2016;1-7. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25614040> [Accessed: Jun 4, 2016]
- [192] Morlán-Coarasa MJ, Arias-Loste MT, Ortiz-García de la Foz V, Martínez-García O, Alonso-Martín C, Crespo J, et al. Incidence of non-alcoholic fatty liver disease and metabolic dysfunction in first episode schizophrenia and related psychotic disorders: A 3-year prospective randomized interventional study. *Psychopharmacology (Berlin)* [Internet]. Dec 12, 2016;**233**(23-24):3947-3952. Available from: <http://link.springer.com/10.1007/s00213-016-4422-7> [Accessed: Dec 26, 2016]

IntechOpen