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# Non-alcoholic fatty liver disease: risk factors and relationship with the metabolic syndrome

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# Non-alcoholic fatty liver disease: risk factors and relationship with the metabolic syndrome

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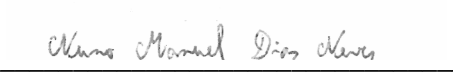
Assinatura: 

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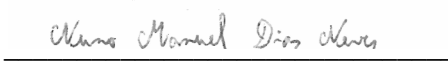
Assinatura: 

**Resumo**

Com o estilo de vida ocidental, a prevalência da síndrome metabólica encontra-se em ascensão, repercutindo-se na incidência de diabetes mellitus tipo 2 e de doenças cardiovasculares. Associadamente, está-se a assistir a um aumento da prevalência de esteatopatia hepática não alcoólica (EHNA) que pode progredir para condições graves como cirrose e hepatocarcinoma. Nesta revisão, pretende-se avaliar alguns fatores de risco para a EHNA, como a obesidade, a infeção pelo vírus da hepatite C e a toma de ácido valpróico e de amiodarona, analisando os mecanismos envolvidos e constatando se a associação com a síndrome metabólica está presente. A combinação de um estilo de vida sedentário com uma dieta desajustada e variações genéticas predisponentes podem impor um maior risco na progressão da EHNA nalguns grupos da população. Também se verifica o envolvimento de distúrbios metabólicos causados por fármacos e o efeito citopático do genótipo 3 do vírus da hepatite C.

**Palavras-chave:** esteatopatia hepática não alcoólica; síndrome metabólica; obesidade; hepatite C; ácido valpróico; amiodarona.

**Non-alcoholic fatty liver disease: risk factors and relationship with the metabolic syndrome**

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**Abstract**

With the western lifestyle, the prevalence of the metabolic syndrome is in an upward trend, with repercussions on the incidence of type 2 diabetes mellitus and cardiovascular diseases. In association, there is an increase of the prevalence of non-alcoholic fatty liver disease (NAFLD), which may progress to serious conditions such as cirrhosis and hepatocellular carcinoma. In this review, some risk factors for NAFLD such as obesity, hepatitis C virus infection and the intake of valproic acid and amiodarone will be assessed, analyzing the mechanisms involved and investigating whether the association with the metabolic syndrome is present. The combination of a sedentary lifestyle with an inadequate diet and predisposing genetic variations may pose more risk of progression of NAFLD to some groups of people. In addition, metabolic derangements caused by drugs and the cytopathic effect of hepatitis C virus genotype 3 are also implicated.

**Keywords:** non-alcoholic fatty liver disease; metabolic syndrome; obesity; hepatitis C; valproic acid; amiodarone.

**Abbreviations**

ALT - Alanine transaminase

BMI - Body mass index

CHCVI - Chronic hepatitis C virus infection

DAG - Des-acyl-ghrelin

FGF21 - Fibroblast growth factor 21

HCC - Hepatocellular carcinoma

HCV - Hepatitis C virus

HDL - High-density lipoprotein

IR - Insulin resistance

LPS - Lipopolysaccharide

MS - Metabolic syndrome

MTP - Microsomal triglyceride transfer protein

NAFLD - Non-alcoholic fatty liver disease

NASH - Non-alcoholic steatohepatitis

ROS - Reactive oxygen species

TNF-α - Tumor necrosis factor-α

VA - Valproic acid

## Introduction

The definition of the metabolic syndrome (MS) may differ according to several major organizations, such as the World Health Organization, the International Diabetes Federation, the European Group for the Study of Insulin Resistance and the National Cholesterol Education Program Adult Treatment Panel III. In common, they refer the importance of increased abdominal obesity, high serum triglyceride levels, low high-density lipoprotein (HDL) cholesterol, high blood pressure, impaired fasting glucose or the presence of insulin resistance (IR). These risk factors are associated with an increased likelihood for developing type 2 diabetes mellitus, atherosclerotic cardiovascular disease and chronic kidney disease [1-3]. The MS is characterized by a pro-inflammatory and pro-thrombotic state and it is related with reproductive disorders as well as non-alcoholic fatty liver disease (NAFLD). MS's components may reflect bad nutritional choices with overnutrition and a sedentary lifestyle with consequent excess adiposity [4,5].

The prevalence of the MS has been found to be in an upward trend among American adults, especially women, in which it increased from 27.0% to 32.9% between 1988 –1994 and 1999–2000. The rise in blood pressure values, waist circumference and hypertriglyceridemia were the main cause for the increased prevalence of the MS [6].

Therefore, given the high prevalence of the MS and its associated features, this review will focus on one of them – the NAFLD.

NAFLD comprises a spectrum of liver pathologies from hepatic steatosis and non-alcoholic steatohepatitis (NASH) to cirrhosis, with a daily alcohol consumption <20g. When lipid content in the liver exceeds 5–10% by weight, it is considered hepatic steatosis, whereas in NASH there is steatosis, mixed lobular inflammation, ballooning degeneration of hepatocytes with or without Mallory bodies or perisinusoidal fibrosis. Cirrhosis is diagnosed through the presence of regenerating nodules of hepatocytes surrounded by fibrous bands. Since there is a risk of progression to hepatocellular carcinoma (HCC), NAFLD should be closely regarded as it is a common liver disease that affects approximately 15–30% of the general population and its prevalence is increasing worldwide [7-12].

There are several causes of NAFLD. Besides the MS and its components (for example obesity), also acute starvation, total parenteral nutrition, celiac disease and rapid weight loss or bacterial overgrowth, following ileal or gastric bypass, have been implicated. Moreover, autoimmune hepatitis, primary biliary cirrhosis, sclerosing cholangitis, hepatitis C, hypopituitarism and drugs like salicylates, tamoxifen, amiodarone, corticosteroids,

methotrexate, valproic acid (VA) and highly active antiretroviral therapy are involved in NAFLD as well [12,13].

NAFLD can be quite indolent. Patients may complain of dull right upper quadrant, abdominal discomfort, fatigue or malaise [14]. The diagnosis of NAFLD frequently occurs after routine liver function tests or it can be a finding in abdominal ultrasounds for other reasons. It may be suspected if aminotransferase serum levels are elevated. Other markers have also been suggested such as high serum ferritin, high plasminogen activator inhibitor-1 or low sex hormone binding globulin [15, 16]. As for the imaging methods, ultrasound is the most affordable choice and computed tomography or magnetic resonance imaging may be used specially for morbidly obese patients. The FibroScan can be used to estimate hepatic fibrosis. Nevertheless, the gold standard for evaluating the degree of hepatic necroinflammation and fibrosis is a liver biopsy, which presents as an invasive and potentially armful method [16-18].

A proposed explanation for the pathophysiology and progression of NAFLD is based on a “two hit” model. The “first hit” would be the accumulation of triglycerides in the hepatocytes. Then, when the liver is exposed to insults such as inflammatory cytokines/adipokines, mitochondrial dysfunction and oxidative stress, occurs the “second hit” resulting in liver damage and the development to NASH and/or fibrosis in about 10-20% of patients with NAFLD. Ultimately, in 15-25% of patients with NASH, cirrhosis develops and 30-40% of subjects with NASH will suffer from a liver related death over a 10-year period such as liver failure or HCC [19-22]. It has been reported that 13% of the cases of HCC are due to NAFLD [23].

Other theories emphasise the influence of gut bacteria. The effects of hepatic exposure to intestinal products originated from its flora such as ethanol and bacterial lipopolysaccharide (LPS) promote the production of reactive oxygen species (ROS) by hepatocytes and liver macrophages which release tumor necrosis factor-α (TNF-α), contributing for a low-grade systemic inflammation. Thus the “second hit” is increased [24].

This review will focus on some of the risk factors for NAFLD such as obesity, hepatitis C virus (HCV) infection, the intake of VA and amiodarone, analyzing the mechanisms involved and investigating whether the association with the MS is present.

**Obesity**

Obesity is becoming a greater concern with its increasing prevalence during childhood, being estimated that 15% of the children in developed countries are obese [25]. In the general paediatric population, 9.6% present NAFLD and 4.2% MS. There is a significant higher prevalence of the MS in obese NAFLD children vs obese non-NAFLD children (37.6% vs 12.0%, respectively) and the prevalence of NAFLD in MS patients was 84.6% which was significantly higher than hypertension (57.5%) and glucose metabolic anomalies (22.6%) and similar to the prevalence of dyslipidemia (89.2%) [26]. Also, the more criteria of the MS are fulfilled, the more severe is NAFLD, in obese teens [27]. Nonetheless, in adults, hepatic IR has been found to be elevated only in NASH, while peripheral IR is significantly higher in all forms of NAFLD [28].

Obese patients frequently exhibit poor dietary habits with meals consisting of hypercaloric foods and with high glycemic index, which are related to IR and liver disorders. In patients with severe obesity is observed the highest prevalence of abnormalities in metabolic and hepatic markers. Qualitatively, these patients present an excessive energy intake from protein of animal source, which is associated with an increased risk of IR, simple carbohydrates and fat, although with low polyunsaturated fatty acids. This dietary pattern revealed the MS in 36% of the obese adult patients with 51% of the subjects presenting liver fibrosis. The prevalence of IR and hepatic steatosis is 63.5% and 63%, respectively. Still, in spite of the abnormalities found, only a third of all the subjects presents elevated alanine transaminase (ALT) and gamma-glutamyl transpeptidase activities, which may not be good parameters to evaluate these patients [29]. Also, saturated fatty acids were found to present a strong risk factor for developing NAFLD in obese adolescents [30].

There are also concerns about the role of fructose consumption, especially in soft drinks, and the pathogenesis of NAFLD. In fact, chronic fructose consumption lowers ghrelin and leptin to a lesser degree than does glucose, presenting a reduced satiety phenomenon, with consequent lower food intake suppression [31]. It was found that about 80% of adult patients with NAFLD, but only 20% of healthy subjects, present an excessive consumption of soft drink beverages, defined as more than 50 g/day of added sugar, which has been associated with increased visceral adiposity, decreased insulin sensitivity and dyslipidemia [32,33].

Patients with obesity often present earlier signs of cardiovascular disease. In adolescents, it was possible to assess that the median carotid artery intima-media thickness is

significantly increased in obese patients, especially on those with hepatic steatosis, reflecting a positive correlation with its severity [34]. Furthermore, adult female patients with NASH present 5.3 times higher Framingham risk scores ≥10% for 10-years compared to healthy individuals, even with an average body mass index (BMI) of 25.4. The severity of NAFLD is associated with a higher prevalence of risk factors for cardiovascular disease and with consequent worse Framingham risk score [35].

Useful anthropometric values were studied in korean adult patients. The cut-off point for detection of NAFLD was 89 cm of waist circumference for men and 84 cm for women, and the cut-off point of waist-to-height ratio was 0.52 for men and 0.53 for women, with high negative predictive values [36]. In men, the association between metabolic risk factors and NAFLD is stronger than in women, possibly due to a more visceral fat deposition in the former gender, which contributes more for the development of NAFLD than subcutaneous fat both in adolescent and adult subjects [37-39]. The same explanation is proposed for the fact that obese patients, either children or adults with Hispanic ethnicity, present higher prevalence of elevated ALT and triglyceride levels when compared to Caucasians and African Americans, who present the least amount of visceral fat for a given level of adiposity (most particularly the later group) [40,41]. Beyond the importance of visceral fat, also total body fat presents a determinant role, explaining why metabolic and hepatic parameters and the prevalence of NAFLD are progressively worse with peripheral obesity, abdominal obesity and mixed obesity, in school-age children [42]. That said, C-reactive protein, a marker of inflammation, was found to be related with the accumulation of fat as well, being associated with BMI and hepatic steatosis but not with the severity of NAFLD in adult obese patients [43].

Emphasizing the role of hypertriglyceridemia in the pathogenesis of NAFLD, Kashyap *et al*. found a positive association between the serum triglyceride levels and the histological severity of NAFLD in severely obese adult subjects [44]. Note that hepatic steatosis also occurs in patients with lipodystrophy who may not be obese but often present IR. The reduced storage of triglyceride in adipose tissue along with impaired adipokine production, such as adiponectin and leptin deficiencies, may explain the mechanism of NAFLD in these subjects [45].

However, not all obese people develop NAFLD and progress to NASH, so several studies investigated what mechanisms were accounting for this fact.

One proposed explanation highlights the role of reduced exportation of triglyceride from hepatocytes, with accumulation of intracellular triglyceride. For example, through

genetic polymorphisms in the promoter of microsomal triglyceride transfer protein (MTP) from which result decreased MTP transcription, like the MTP gene -493 G/G genotype. This genotype is significantly more prevalent among obese children with simple steatosis and NASH than in healthy controls, while no T/T genotype is detected among NASH patients. Also, all NASH patients present the 1183 T/T genotype in the mitochondrial targeting sequence of manganese superoxide dismutase, which is associated with a reduced transportation of this enzymatic ROS scavenger to the mitochondria, therefore predisposing these patients to increased oxidative stress [46].

The patatin-like phospholipase domain–containing protein 3 rs738409 G allele has been associated with a significant higher prevalence of NAFLD in obese children, while not affecting IR and BMI. It was found to be related with an increased hepatocyte triglyceride content since it may be involved in the hydrolysis of triglycerides in adipose tissue [47].

In NAFLD, hepatic triglyceride exports are increased. Still, when its intrahepatic triglyceride content reaches 10%, this mechanism hits a plateau, revealing not be sufficient to normalize its fatty content [48]. Hepatic steatosis has been related with the oversecretion of very-low-density lipoprotein-apolipoprotein B-100 (VLDL-apoB-100), which is an important determinant of atherosclerosis. Yet, weight loss with subsequent reduction of liver fat allows a significant decrease in VLDL-apoB-100 [49,50].

Fibroblast growth factor 21 (FGF21) has been found to be positively associated with BMI, being also significantly increased in NAFLD. It is a hepatic protein that stimulates fatty acid oxidation in the liver and glucose uptake in adipocytes (after being released into the circulation). However, its serum levels are also increased with dyslipidemia and type 2 diabetes mellitus which may indicate a FGF21-resistant state [51].

For the development and progression of NAFLD, it is believed that further insults to the liver are implicated in the process. These may be considered the “second hit” of its pathogenesis by acting on a more susceptible liver.

Obese children with NAFLD present significantly lower adiponectin level compared to obese children without liver abnormalities, noticing that adiponectin is negatively correlated to the degree of liver steatosis and BMI. Adiponectin is thought to protect the progression of NAFLD to NASH by exhibiting anti-inflammatory properties through inhibition of hepatic TNF-$\alpha$ expression, as well as anti-fibrotic and anti-steatotic effects with improvement of glucose metabolism and increased insulin sensitivity. Hypoadiponectinemia

has been associated with IR [52,53] and with higher neuropeptide Y/agouti-related peptide ratio, which increases appetite [54].

Non-obese NAFLD patients exhibit a higher expression of genes related to fatty acid oxidation and ROS elimination when compared to obese NAFLD patients who are unable to compensate the mitochondrial ß-oxidation overload with ß-oxidation in peroxisomes and ω-oxidation in microsomes. So, while both groups generally overexpress genes related with fatty acid metabolism, obese subjects are more prone to severe liver injury, steatosis and IR. These patients present higher leptin levels that should stimulate fatty acid metabolism and peroxisome proliferator-activated receptor α gene expression, so leptin resistance may explain their inferior metabolic machinery [55].

Although patients with NAFLD have a decreased intestinal cholesterol absorption, its hepatic synthesis is increased, independently of BMI. This free cholesterol may contribute to the pathogenesis of NAFLD due to its cellular toxicity [56].

Obese adults with NAFLD present a significant elevation of serum LPS levels, which may be related with an increased intestinal permeability or bacterial overgrowth [57]. It was detected a positive association between the degree of hepatic steatosis and/or inflammation with a significant increase in interleukin-1α and TNF-α production by the monocyte-macrophage system, when stimulated by LPS. The authors state that this dysregulation in pro-inflammatory cytokines may promote the progression from steatosis to NASH [58].

Recently, chronic intermittent hypoxia has been linked with more severe liver damage, presenting higher frequency of NASH and fibrotic lesions, with increased inflammation parameters. It was established a dose-response relationship according to the amount of nocturnal oxygen desaturations in morbidly obese subjects, independent of BMI. Therefore, hypoxia induced inflammation might be a part of the "second hit" for the development of NASH or might decrease the rate of oxidation of free fatty acids which therefore exert their hepatic lipotoxic effect [59].

Acylghrelin, des-acyl-ghrelin (DAG) and obestatin are common products of the ghrelin gene. Estep *et al.* found that morbidly obese patients with NASH present twice the serum levels of DAG than patients without NASH, which is positively correlated with fasting glucose, triglycerides and liver transaminase levels. Moreover, acylghrelin levels are also positively correlated with the fibrosis stage. The authors hypothesize that the expression of ghrelin gene is increased in response to ROS and pro-apoptotic signals since it is thought that ghrelin and DAG present an anti-apoptotic effect. Thus, while they might help to protect the liver from steatohepatitis, by inhibiting apoptosis of myofibroblast-like cells, these molecules

are believed to be fibrogenic. Additionally, interleukin-7 is found to be significantly reduced in NAFLD patients with type 2 diabetes mellitus or fibrosis, which seems to be in agreement with its role in suppressing the expression of several molecules that promote fibrosis and diabetes [60].

Ultimately, as simple steatosis progresses to more severe forms, hepatocyte apoptosis' rate is significantly increased, as well as caspase-2 and caspase-3 activation. Moreover, increased phosphorylation of c-Jun N-terminal kinases in muscle and liver in NASH subjects is a common route for IR and apoptosis and may pose a useful therapeutic target [61].

**Hepatitis C**

Features of NAFLD occur in chronic hepatitis C virus infection (CHCVI), namely hepatic steatosis, inflammation, increased oxidative stress, IR, fibrosis and HCC, that can be dependent of the virus genotype [62,65]. 8-Isoprostane, a marker of oxidative stress, has been shown to be increased both in NAFLD and in CHCVI, proving that lipid peroxidation is involved in the pathogenesis of these two entities [66].

A study of an endemic area of HCV genotypes 1 and 2, found that infected patients have a higher prevalence of MS than controls (24.7% vs 13.2%), presenting also an increased prevalence of hypertension and of high waist circumference, ALT and insulin levels, while having lower LDL-cholesterol [67]. Adiponectin levels have been determined to be lower in CHCVI patients which also have MS (61.5% of the HCV genotype 1 infected patients had MS). A significant negative correlation has been established between adiponectin and insulin and between adiponectin and IR [68].

However, another study determined that the prevalence of the MS is only significantly higher in NAFLD patients (not infected by HCV). In HCV genotypes 1- or 2- infected patients, its prevalence is similar to that of healthy nonsteatotic control subjects, even though both CHCVI and NAFLD have been associated with a high prevalence of IR. The authors retrieved that anthropometric (BMI) and metabolic (triglycerides) derangements are independently associated with IR in NAFLD patients as well as sex and fibrosis with IR in HCV infection [69]. Steatosis of genotype 1 infected patients has been associated with pre-existing risk factors for NAFLD, while genotype 3 independently associates with hepatic steatosis [70]. Other authors have found no association between CHCVI and MS. Yet, when

both pathologies are present, the homeostatic model assessment insulin value is higher than in those patients without CHCVI [71].

In fact, the concomitant presence of NAFLD and HCV in obese patients increases the likelihood of diabetes, hypertension and/or hypertriglycemia and reduces the efficacy of antiviral therapy. It has been reported that if a sustained virological response to therapy is attained there is a significant reduction of hepatic steatosis in patients with HCV genotype 3 (whereas genotype 1 showed no change). It has also been reported that IR can be ameliorated and beta-cell function improved (for the 3 genotypes) with therapy [72-75].

Moreover, the development of liver fibrosis is prior in CHCVI than in patients with NAFLD only. However, with the progression of the disease, fibrosis becomes as severe in both groups [76]. In CHCVI patients (including the 3 genotypes), type 2 diabetes mellitus and superimposed steatohepatitis are independently associated with advanced fibrosis [77]. MS features, including obesity and type 2 diabetes mellitus, play a critical role in the pathogenesis of fibrosis in CHCVI (genotypes 1 and 2). The Nagasaki score (the total number of specific risk factors, namely an older age, obesity and type 2 diabetes mellitus) is a significant predictor of severe fibrosis in CHCVI [78].

**Valproic acid**

VA is a drug commonly used in the treatment of epilepsy and bipolar disorder. It has been associated with hepatic steatosis as well as with an increase in plasma insulin, triglyceride and BMI levels or a decrease in HDL levels, indicating that VA treatment is related with the risk of developing metabolic disturbances [79-81]. VA, and some of its metabolites, inhibit mitochondrial β-oxidation of fatty acids and exhibit detrimental metabolic functions by reducing the levels of free coenzyme A and carnitine in the liver (and total serum carnitine) [82].

A study has presented that from 114 paediatric patients being treated with VA, for at least 24 months, 40% became obese and 43.5% of whom developed MS [83]. Other studies achieved similar results with the prevalence of MS in VA-treated patients being 45.5%, compared with the 15.4% in the carbamazepine and 27.3% in the control groups [84]. When compared with weight-matched controls, patients taking VA have significantly lower HDL-cholesterol and, oddly, higher adiponectin levels [85].

Despite the fact that patients treated with VA have more prevalence of NAFLD than normal-weight controls (36.0% vs 7.5%, respectively), when compared with weight-matched controls the prevalence appears to be similar (36.0% vs 34.9%, respectively). Actually, VA can lead to a substantial weight gain by increasing appetite and thirst for high-calorie beverages, which might explain the frequency of NAFLD related to the treatment with VA [86].

In a comparison between lean patients and lean controls, the former group has higher serum insulin (relative to BMI) and uric acid levels but lower HDL-cholesterol than the latter suggesting that hyperinsulinemia is not a consequence of obesity. Nonetheless, obese patients have worse outcomes than lean patients [87].

A case report of an 11-year-old girl shows that the patient developed obesity and IR after a year of taking VA. This drug was then replaced by levetiracetam. After 6 months there was a complete regression of NAFLD (evaluated by ultrasound) and normalization of metabolic and endocrine parameters. Notice that weight loss was achieved without any specific diet or increased physical activity [88].

It has also been reported a case of a patient that had taken VA for 17 years and that was diagnosed an hepatic tumor with 13 cm in diameter in a noncirrhotic NASH liver. He was 64 years old and obese (34.1 $kg/m^2$) without any other risk factor for developing an HCC, what suggests that chronic inflammation in itself could be an important risk factor in the development of HCC [89].

**Amiodarone**

Amiodarone is an antiarrhythmic drug which is associated with varying degrees of hepatotoxicity. Seventy-four percent of patients that were prescribed amiodarone for longer then 60 days present MS criteria [90]. Amiodarone has been demonstrated to inhibit the mitochondrial respiratory chain and β-oxidation and to uncouple the oxidative phosphorylation. Moreover, there is an increase of ROS and mitochondrial swelling, leading to apoptosis and necrosis of cells [91-93]. Due to the reduced β-oxidation of fatty acids, hepatic triglyceride accumulation occurs, preceding hepatic steatosis [82].

In a subject with amiodarone-induced steatohepatitis with advanced fibrosis, there was a temporal association between the starting of the amiodarone therapy, the diagnosis of diabetes, hypertension and the development of abnormal liver function tests with progression

to cirrhosis and portal hypertension. This 77-year-old woman had no other risk factor such as history of hepatitis B or C infection, autoantibodies, weight loss, obesity or family history of liver disease [94].

There is another case report of a patient in similar conditions that developed NASH with micronodular cirrhosis. When amiodarone was withdrawn the serum aminotransferase level and ascites were slightly improved [95].

**Discussion**

Although only a fraction of patients with NAFLD may suffer from complications related to it, the sheer number of its prevalence should be a concern.

NAFLD and the MS overlap quite often, even on different risk factors for the former, reinforcing the vital role of the liver in metabolism.

The different aetiologies of NAFLD imply that patients should be studied individually and the underlying cause should be established since it is crucial to prevent further progression of the disease, initiating treatment whenever possible. In this review, several mechanisms that may explain the pathogenesis of NAFLD were presented. In the obesity section, the "two hit" model seemed apparent where there is a positive balance of calories in the liver and hepatocytes accumulate triglycerides that may come from diet, adipose tissue or being synthesized *de novo*. The loss of balance between their consumption and influx as well as the additional degeneration of hepatocytes can be promoted through individual genetic variations, which can contribute for the susceptibility for developing NAFLD. This fact may elucidate why certain people may be more prone to progress to more hazardous conditions such as NASH, cirrhosis and HCC. Cytokines also appear to present an important role in modulating metabolic and inflammatory parameters, posing a probable key stage in the pathophysiology of NAFLD.

In hepatitis C, we can discriminate the genotype of the virus, since their means of action are different. HCV infection ought to be prevented, especially with information about the methods of transmission as well as risk behaviours, and pharmacological treatment must be started as soon as possible. Since other metabolic comorbidities further exacerbate the progression of the liver disease, these patients should also be evaluated and manage their other conditions.

VA seems to contribute for the development of NAFLD through changes in the dietary habits of patients who may seek an increase of calories ingested with consequent weight gain as well as through metabolic disturbances with the decrease of fatty acid catabolism leading to hepatic steatosis.

Amiodarone shares this last feature with VA, meaning that processes that block the metabolism of fatty acids are important mechanisms which promote NAFLD. However, there is scarce literature regarding this subject and a large number of the articles presented relied on case studies, with their inherent drawbacks. Another point that should be highlighted is that patients taking amiodarone often present cardiovascular risk factors that also contribute to NAFLD.

Current treatments are not specific for NAFLD. Obese patients may benefit from weight loss programs as well as from the use of insulin sensitizers, such as metformin and glitazones [96]. There are also positive results with bariatric surgery [97]. Liver transplantation due to NAFLD-related cirrhosis has been associated with recurrence of NAFLD in 39% of the recipients, 74% of whom had NASH, although with mild histologic activity. NAFLD recurrence is not only significantly associated with BMI of the pretransplanted patients but also with the patients' BMI after the transplantation, as well as high triglyceride levels and prednisone dose [98].

Recent studies evaluated the benefits of nutritional modulation with adherence to the Mediterranean diet with improvements of the MS features. The replacement of saturated fats for mono or polyunsaturated fatty acids and the inclusion of fruits, vegetables, legumes, fatty fish and olive oil, providing the recommended dosages of fibre, antioxidants and minerals (magnesium, zinc and calcium, for example), favour the success of the diet [99,102].

There are other causes of NAFLD, but this review focused on these four due to the large prevalence of individuals exposed to these factors and due to their relative simplicity, since there are not many variables involved.

It would be interesting to see future studies evaluating the effectiveness of current treatments on patients with NAFLD of different known aetiologies, verifying whether there are differences in the outcomes.

In conclusion, it is never enough to highlight the importance of having a balanced diet and physical exercise. These simple preventive measures are more and more imperative as the enormous costs of health care could be avoided with a sensible lifestyle. This empowers the society to be more responsible for its own health, allowing a more sustainable health care system. Otherwise, especially in individuals with genetic predisposition, it is expected to

witness a significant rise in cryptogenic cirrhosis and HCC, as well as other diseases associated with the MS. HCV infection must be rather prevented or at least treated in a timely manner and patients who are taking drugs that are known or suspected of causing NAFLD should be carefully monitored. One has to weigh the benefits and risks of continuing with such medication and wonder if it is possible to change to a more benign one or reduce the dosage.

## Declaration of interests

Nothing to declare.

## Contributor statements

Maria João Martins and Nuno Dias Neves: conception of the article. Nuno Neves: drafting of the article. Maria João Martins: revision of the article's structure, organization and coherence. Both authors approved the final version of the manuscript.

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**Anexo**

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