Review Article

Nonalcoholic fatty liver disease: A precursor of the metabolic syndrome

Amedeo Lonardo, Stefano Ballestri, Giulio Marchesini, Paul Angulo, Paola Loria

Abstract

The conventional paradigm of nonalcoholic fatty liver disease representing the “hepatic manifestation of the metabolic syndrome” is outdated. We identified and summarized longitudinal studies that, supporting the association of nonalcoholic fatty liver disease with either type 2 diabetes mellitus or metabolic syndrome, suggest that nonalcoholic fatty liver disease precedes the development of both conditions.

Online Medical databases were searched, relevant articles were identified, their references were further assessed and tabulated data were checked.

Although several cross-sectional studies linked nonalcoholic fatty liver disease to either diabetes and other components of the metabolic syndrome, we focused on 28 longitudinal studies which provided evidence for nonalcoholic fatty liver disease as a risk factor for the future development of diabetes. Moreover, additional 19 longitudinal reports which nonalcoholic fatty liver disease precedes and is a risk factor for the future development of the metabolic syndrome.

Finally, molecular and genetic studies are discussed supporting the view that aetiology of steatosis and lipid intra-hepatic compartmentation are a major determinant of whether fatty liver is/is not associated with insulin resistance and metabolic syndrome.

Data support the novel paradigm of nonalcoholic fatty liver disease as a strong determinant for the development of the metabolic syndrome, which has potentially relevant clinical implications for diagnosing, preventing and treating metabolic syndrome.

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1. Introduction

Steatosis and steatohepatitis, in the absence of competing aetiologies such as high alcohol intake, hepatitis C virus (HCV) infection, drugs and other endocrine disorders, are both part of the nonalcoholic fatty liver disease (NAFLD) spectrum [1–3]. NAFLD may either occur in the absence of fatty changes – and is often alluded to either as “cryptogenic” or nonalcoholic steatohepatitis (NASH)–cirrhosis [4,5] – or encompass hepatic and extra-hepatic complications, from hepatocellular carcinoma to atherosclerosis [6]. NAFLD is presently recognized as one the most common causes of altered liver tests, of end-stage liver disease requiring liver transplantation, and is frequently associated with the constellation of clinico-laboratory features that comprise the metabolic syndrome [7–9].

The metabolic syndrome is a cluster of cardio-metabolic conditions, generally triggered by an expansion of the adipose visceral tissue [10], which include insulin resistance – with or without impaired glucose metabolism and type 2 diabetes (T2D) – atherosgenic dyslipidemia [low high density lipoprotein (HDL)-cholesterol and high triglycerides], and high blood pressure [11]. The present definition of metabolic syndrome, at variance with one of its earliest proposals [12], does not include hepatic steatosis despite evidence supporting this association [13]. The existence and the utility of metabolic syndrome as a separate entity has been challenged by some experts [14]. Moreover, it remains controversial whether the presence of the full-blown syndrome really adds to the cardiovascular risk dictated by its individual components, particularly T2D [15,16]. Nevertheless, the single traits of the metabolic syndrome tend to aggregate in the same individuals and, more importantly, the presence of each of them often anticipates the appearance of
additional components over time [17]. This, together with the possible progression to organ failure and of the development of some cancer types, including primary liver cancer [9,11,18,19], makes metabolic syndrome a relevant condition in clinical practice and a major public health concern worldwide.

In 1999, European researchers [20–22] provided biological, clinical and epidemiological evidence for the theory that NAFLD should be regarded as the hepatic manifestation of the metabolic syndrome (Table 1) [20]. Since then, a “chicken and egg” scientific debate has arisen, concerning the primacy of metabolic syndrome (and insulin resistance) over NAFLD or, conversely, of NAFLD over the metabolic syndrome [23]. Moreover, while most studies acknowledge NAFLD to be a risk factor for T2D [24], the notion that NAFLD anticipates the future development of components of the metabolic syndrome other than T2D is far less accepted, although it has been suggested by few authors [23,25,26] mainly based on their expert opinion.

In our systematic review of the literature, we discuss all available data supporting the view that NAFLD, rather than being a mere “manifestation of the metabolic syndrome” is indeed a necessary precursor of the future development of metabolic syndrome in humans. To this end, further to identifying all relevant cross-sectional studies, we specifically address the evidence from prospective studies showing that NAFLD is an independent risk factor for the future development of both T2D and other components of the metabolic syndrome. Methodological limitations of such data are briefly analysed. Finally, we discuss how some examples of dissociation of NAFLD from metabolic syndrome – seemingly contradicting our thesis – eventually confirm the general paradigm of NAFLD being a precursor of the metabolic syndrome.

2. Research strategy

The PubMed data base was manually searched for the following terms: “Metabolic syndrome”; “Type 2 Diabetes”; “Obesity”; “Dyslipidemia” “Hyperlipidemia”; “Insulin resistance”; “Prediabetes”; “Fatty liver”; “Follow-up”; “Association” and “Dissociation”. The research was updated at the 28th of April 2014. Further bibliographic updates were performed whenever needed during the revision process.

All of the identified articles and their references were further checked in duplicate in order to identify appropriate articles. Pertinent studies were identified as a result of the agreement two investigators, who also tabulated the methods and chief findings of the selected material.

The accuracy of the tabulated data was independently performed by all authors.

3. Association of NAFLD with other conditions

3.1. Type 2 diabetes

Many cross-sectional studies demonstrate that NAFLD is associated with insulin resistance/pre-diabetes/T2D [27–39]. Despite a variable follow-up range, data appear consistent in disclosing that the “NAFLD pathway” to T2D follows a route characterized by insulin resistance developing at various anatomic sites, notably including hepatic insulin resistance. Interestingly, the NAFLD-T2D association occurs irrespective of potential confounders and particularly obesity and moderate alcohol drinking, suggesting that NAFLD may closely mirror the development of fatty pancreas disease [40] and thus reflect tissue lipotoxicity. Moreover, a quantitative relationship links fasting plasma glucose with NAFLD prevalence: the higher the former, the higher the latter [29].

However, widely acknowledged predictors of T2D in non-NAFLD populations (such as high fasting glucose and low HDL-cholesterol and adiponectin, mirroring insulin resistance) and low physical activity also predict T2D in NAFLD suggesting that NAFLD probably amplifies the physiological determinants of T2D. Finally, most findings appear to be confirmed irrespective of ethnicity (e.g. T2D-prone Asians vs. Caucasians) and of the diagnostic technique followed to capture NAFLD, spanning from the most invasive or accurate tests (liver histology, magnetic resonance imaging) to the most insensitive (liver tests), via the most widely performed ultrasonography scanning.

However, the above studies [27–39] are cross-sectional. Accordingly, they do not rule out the possibility that primarily impaired gluco-regulation may eventually result in the development of NAFLD. In order to clarify this issue, in Supplementary Table S–1 the studies showing NAFLD as a risk factor for the future development of T2D were summarized. Eleven out of 12 studies were conducted in Far East, most of them from Korea. With such a limitation, they confirm that NAFLD is an independent risk factor for the development of T2D, although confirmation from Western countries appears necessary. Moreover, the diagnostic technique used to detect NAFLD may lead to different results. For instance,

<table>
<thead>
<tr>
<th>Table 1</th>
<th>Nonalcoholic fatty liver disease and metabolic syndrome – the original spectrum of similarities.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Epidemiology</td>
<td>Metabolic syndrome</td>
</tr>
<tr>
<td>Prevalence in the general population</td>
<td>Up to 25% of adults</td>
</tr>
<tr>
<td>Prevalence grows with age</td>
<td>Yes</td>
</tr>
<tr>
<td>Males more affected</td>
<td>Yes (adults, children)</td>
</tr>
<tr>
<td>Anthropometry</td>
<td>Abdominal adiposity is an independent predictor</td>
</tr>
<tr>
<td>Association with central obesity</td>
<td>Documented</td>
</tr>
<tr>
<td>Metabolism – Association with</td>
<td></td>
</tr>
<tr>
<td>Hyperinsulinemia</td>
<td>Yes</td>
</tr>
<tr>
<td>Hypertension</td>
<td>Yes</td>
</tr>
<tr>
<td>Obesity</td>
<td>Yes</td>
</tr>
<tr>
<td>Hypertriglyceridemia</td>
<td>Yes</td>
</tr>
<tr>
<td>Low HDL Cholesterol</td>
<td>Circumstantial Evidence</td>
</tr>
<tr>
<td>Clinical features</td>
<td></td>
</tr>
<tr>
<td>Systemic disease</td>
<td>Yes</td>
</tr>
<tr>
<td>Accelerated atherosclerosis</td>
<td>Yes</td>
</tr>
<tr>
<td>Response to diet and/or exercise</td>
<td>Yes</td>
</tr>
<tr>
<td>Experimental pathology</td>
<td>Atherogenic diet induces steatosis</td>
</tr>
</tbody>
</table>

Adapted from Lonardo [20].
the less specific gamma-glutamyltransferase (GGT) values more accurately predict the development of T2D than aminotransferase levels. Similarly, the ultrasonographic severity of NAFLD is associated with increasing risks of T2D and, notably, NASH (more than simple steatosis) appears to be more closely associated with T2D. However, such a conclusion was challenged by a subsequent meta-analysis by Musso et al. showing that the risk of developing T2D is increased to a similar extent in both steatosis and NASH [41].

### 3.2. Other components of metabolic syndrome

Data reporting NAFLD as a risk factor for either some individual components of metabolic syndrome [arterial hypertension [42,43] and dyslipidemia [44]] or the full-blown syndrome are less established than NAFLD progression to T2D. Many cross-sectional studies support the view that NAFLD is associated with the metabolic syndrome [13,22,45–64].

Data universally confirm the strong NAFLD-metabolic syndrome association in humans. In addition, although each component of metabolic syndrome [including serum uric acid, not included in the diagnostic criteria but clearly a component of the syndrome from a pathophysiologic point of view [8]] is independently associated with NAFLD, a few features (visceral obesity, dyslipidemia and insulin resistance/T2D) are more closely related to NAFLD. The association of NAFLD with the metabolic syndrome appears to be stronger in lean than in obese individuals, especially in women. NAFLD specifically anticipates the presence of metabolic disorders and independently predicts insulin resistance, identifying those individuals with insulin resistance who may be missed by metabolic syndrome criteria in certain populations. The frequency of metabolic syndrome significantly increases with quintiles of ALT and GGT, even within the normal range of these enzymes [51]. Moreover, T2D and hypertriglyceridemia are more commonly associated with raised hepato-biliary enzymes, and particularly GGT [65], an association which appears to be independently mediated by insulin resistance. In turn, the presence of metabolic syndrome is associated with the development of fibrosis or NASH.

Again, association in cross-sectional studies does not provide evidence as to which condition comes first. In order to address this issue, studies supporting the view that NAFLD precedes the development of metabolic syndrome are summarized in Supplementary Table S-2.

### Table 2

Nonalcoholic fatty liver disease and metabolic syndrome – the expanded spectrum of similarities.

<table>
<thead>
<tr>
<th></th>
<th>NAFLD</th>
<th>Metabolic syndrome</th>
</tr>
</thead>
<tbody>
<tr>
<td>Increasing serum uric acid levels</td>
<td>Are associated with increased prevalence and histological severity of NAFLD [66–68]</td>
<td>Are associated with increased prevalence and incidence of metabolic syndrome [69–82]</td>
</tr>
<tr>
<td>High fructose consumption</td>
<td>Is associated with increased prevalence of NAFLD and increased NASH severity [83,84] owing to enhanced lipogenesis, hepatic fibrosis, inflammation, endoplasmic reticulum stress and lipopaoptosis [85]</td>
<td>Increases the risk of developing metabolic syndrome and its components [86–88]</td>
</tr>
<tr>
<td>TSH</td>
<td>Clinical/subclinical hypothyroidism and elevated TSH, though in the normal range, are associated with increased prevalence and histological severity of NAFLD [89–92]</td>
<td>There is a close dose-dependent relationship between increasing TSH and incident/prevalent metabolic syndrome in various ethnicities and across different age-groups [93–97]</td>
</tr>
<tr>
<td>Bile acids</td>
<td>Participate in the pathogenesis of NAFLD through regulating hepatic nutrients and energy homeostasis; stimulating GLP-1 secretion in the small intestine and energy expenditure in brown adipose tissue and skeletal muscle [98]. UDCA’s role in NASH management, however, remains controversial [99,100]</td>
<td>Are powerful regulators of metabolism and induce increased thermogenesis, protect from diet-induced obesity, hepatic lipid accumulation, and increased plasma triglyceride and glucose levels via activation of bile acids receptors (FXR and TGR5) [101–103]</td>
</tr>
<tr>
<td>Increased physical exercise</td>
<td>Protects from NAFLD, independent of weight loss [104–108] and improves histological steatosis [109]</td>
<td>Protects from the development of and cures established metabolic syndrome [110–116]</td>
</tr>
<tr>
<td>Light to moderate alcohol consumption</td>
<td>Is associated with a reduced prevalence of NAFLD and NASH [3,117–120]</td>
<td>Seemingly protects from the development of at least some components of the metabolic syndrome. Conversely, heavy drinking increases the risk of metabolic syndrome [121–122]</td>
</tr>
<tr>
<td>Caffeine</td>
<td>Seemingly protects from NAFLD and liver fibrosis [133–136] without decreasing the risk of NASH [134,135]. Beneficial liver outcomes are mediated by antioxidant, antifibrogenic, anti-inflammatory, insulin sensitizing, lipolytic, cyto, and genoprotective activity [137–142]</td>
<td>Reportedly to be a protective factor from metabolic syndrome, maybe through a lipolytic effects of caffeine on adipocytes [143–145].</td>
</tr>
<tr>
<td>Smoking</td>
<td>Risk factor for the development of NAFLD and its fibrotic progression [146–148]</td>
<td>Positively and reversibly associated in a dose-dependent manner with the risk of metabolic syndrome [149]</td>
</tr>
<tr>
<td>Altered sleep physiology</td>
<td>Shorter sleep duration and severe obesity-related obstructive sleep apnoea are independently associated with development and histological severity of NAFLD [150–152]</td>
<td>Altered sleeping time duration and continuity are associated with increased risk for the metabolic syndrome [153,155–156]</td>
</tr>
<tr>
<td>Brown adipose tissue</td>
<td>Subjects maintaining active brown adipose tissue are protected from NAFLD [157]</td>
<td>The presence of brown adipose tissue is inversely related to BMI. Brown adipose tissue activity plays a role in metabolic homeostasis between glucose metabolism [158–161]</td>
</tr>
<tr>
<td>Intestinal microbiota</td>
<td>May promote NASH by several mechanisms including improved energy efficiency; increased gut permeability and production of lipopolysaccharides with activation of the innate immune system; altered bile acids pool; reduced bioavailability of dietary choline and increased endogenous ethanol production [162–172]</td>
<td>Plays a significant role in the development of metabolic syndrome via increased energy harvesting, metabolic endotoxia; control of lipid and glucose metabolism via the composition of bile-acid pools and the modulation of FXR and TGR5 signalling; modulation of intestinal microbial composition by innate immune system; &quot;brain-gut enteric microbiota axis&quot; [170,173–188]</td>
</tr>
<tr>
<td>Vitamin D deficiency</td>
<td>Is associated with NAFLD development and the severity of hepatic histological changes [189–192]</td>
<td>May increase the risk for metabolic syndrome, type 2 diabetes, insulin resistance, obesity, hypertension and cardiometabolic outcomes [193–198]</td>
</tr>
</tbody>
</table>

List of abbreviations: BMI, body mass index; FXR, farnesoid X receptor; GLP-1, glucagon-like peptide-1; NAFLD, nonalcoholic fatty liver disease; NASH, nonalcoholic steatohepatitis; TGR5, membrane-type G-protein-coupled receptor for bile acids; TSH, thyroid stimulating hormone; UDCA, ursodeoxycholic acid.
Collectively, they suggest that NAFLD, either diagnosed via surrogate indices or by ultrasonography, antedates the presence of several components of metabolic syndrome, not only of T2D. This association is largely regulated by the presence of insulin resistance. However, in 13 out of 15 such studies, NAFLD was diagnosed on surrogate laboratory indices, on ultrasonographic findings in two and in none by the gold standard liver biopsy technique. Moreover, the diagnostic criteria tended to be heterogeneous and given that enlarged waist circumference is both a main pathogenic key for NAFLD and is central for the diagnosis of metabolic syndrome, it is virtually impossible to rule out the possibility that visceral obesity links the development of both NAFLD and metabolic syndrome. Finally, no specific traits of the metabolic syndrome (dysglycemia, dyslipidemia, high blood pressure and central obesity) are mandatory for the development of the full-blown metabolic syndrome (Supplementary Table S-2).

4. Does NAFLD precede metabolic syndrome? Insulin resistance as a major player

Compared to our “historical” perception (Table 1), present understanding of the epidemiological, clinical and pathogenic similarities between NAFLD and the metabolic syndrome has expanded to a substantial extent. Accordingly, it presently embraces a large number of risk/protective factors and biological mediators, i.e., serum uric acid, fructose, TSH, bile acids, physical exercise, alcohol, caffeine, smoking, altered sleep physiology, brown adipose tissue, intestinal microbiota and vitamin D deficiency (Table 2) [66–198]. Associations of these factors with NAFLD may mirror different pathophysiological mechanisms underlying the natural course leading from NAFLD to metabolic syndrome.

Phylogenetically evolved to promote survival, insulin resistance will lead to the deleterious manifestations of the metabolic syndrome whenever inappropriately activated by unhealthy lifestyles [199]. Clinically, insulin resistance is defined as the critical connector linking stress, visceral adiposity, and decreased cardio-respiratory fitness with increased cardiovascular disease [14]. Biologically, elevated fasting glucose will result from hepatic insulin resistance, whereas increased free fatty acids (FFA) concentrations are the expression of peripheral insulin resistance [200]. At molecular level, insulin resistance invariably results from the interaction of cell membrane receptors with either excess nutrients or inflammatory cytokines. Such an interaction will trigger JNK and IKK, protein kinase C, S6K, mTOR, and ERK. All these pathways are involved in the amplification of inflammation, phosphorylation of insulin receptor substrates 1 and 2, stress of endoplasmic reticulum, production of reactive oxygen species and mitochondrial dysfunction [201]. Once triggered, this vicious circle of systemic metabolic dysfunction of insulin and leptin action tends to self-perpetuate [202]. Impaired intracellular insulin signalling will eventually result in perturbed Glut-4-mediated glucose uptake with preserved/potentiated MAP kinase-mediated mitogenesis/cell survival pathways [200].

First proposed, as early as 1988, as the underlying factor in the metabolic syndrome [203], insulin resistance undoubtedly plays a major role in the pathophysiology of the metabolic syndrome [204]. The expansion of visceral adipose tissue will result in the liver being overflowed with FFA, thus leading to steatosis. A fatty liver, in turn, is the prerequisite for the development of highly atherogenic dyslipoproteinemia featuring low HDL-cholesterol, hypertriglyceridemia and increased small and dense low-density lipoprotein (LDL) particles, as well as for fasting hyper-glycemia, which stimulates the pancreas to compensatory insulin over-secretion. Collectively, increased FFA concentrations, hyperglycemia and hyper-insulinemia may directly (or indirectly via sympathetic over-activation) promote arterial hypertension and ectopic deposition of fat in extra-adipose tissues. More importantly, the described metabolic derangements are intimately associated with increased production of fibrinogen, PAI-1, TNF-alpha, IL-6 and decreased adiponectin levels. A subclinical pro-inflammatory proatherogenic state will ensue, which is a major biological feature of insulin resistance and a typical hallmark of the metabolic syndrome [204].

While most individuals with NAFLD have insulin resistance [13,205], only a minority of those with NAFLD exhibit the full-blown metabolic syndrome [206]. This finding most likely results from fatty liver being a precursor [207] and a necessary prerequisite to the development of progressive metabolic disease [23,208–210]. Adipose tissue is the only physiologic reservoir of fatty acids, and the accumulation of fat in the extra-adipose tissues may be tissue-damaging and eventually lead to organ dysfunction (a concept alluded to as “lipotoxicity”). Data from a mouse model support the view of fatty liver being a protective mechanism which prevents progressive liver damage in NAFLD [211] suggesting that esterification into triglycerides might well be a mechanism which detoxifies histologically damaging FFA in the liver. Further evidence for the concept that fatty liver is an adaptive phenomenon derives from its occurrence in animal species other than humans suggesting that steatosis is evolutionarily “useful” [212–215].

Characterization of the role of PKCε is probably the most exciting progress in our understanding of the molecular pathogenesis of the connection linking liver steatosis with hepatic insulin resistance. In obese humans, fatty substrates such as diacylglycerol account for 64% of the variability of insulin sensitivity via the activation of PKCε [216], which impairs insulin signalling as shown in rats using antisense oligonucleotides [217]. Of outstanding interest, the intracellular compartmentation of diacylglycerol dictates whether or not PKCε will translocate to the plasma membrane so promoting hepatic insulin resistance [218]. Stated otherwise, various classes of lipids (such as mono-, di- and tri-acylglycerol) sequestered within lipid droplets/Endoplasmic reticulum fraction are deemed to be innocuous as far as the development of hepatic insulin resistance is concerned in the CGI-58 knockdown mouse model [219]. In humans, however, most cases of NAFLD are strongly associated with insulin resistance. In agreement, diacylglycerol acyl transferase 2 (DGAT2), the enzyme catalysing the final step in the biosynthesis of triglycerides, may link glycermia and triglyceridemia [220]. These data are fully consistent with the view that the development of a fatty liver occurs upstream in the chain of metabolic events eventually leading to the development of the metabolic syndrome [210] and help understand the mechanistic reason why subjects with NAFLD were 1.6 times more likely to develop T2D than NAFLD-free persons in a recent 3-year follow-up Japanese study [221]. Of clinical interest, a growing number of components of the metabolic syndrome are associated with an increased risk of hepatic fibrosis in the individual patient [222] thus enabling the identification of those who should undergo liver biopsy [223]. In particular, data have shown that a vicious circle links T2D and liver disease, with NAFLD contributing to the future development of T2D and the latter triggering progressive liver injury [40]. Fatty pancreas develops in the setting of metabolic syndrome, carrying amazing similarities to NAFLD in terms of organ inflammation, end-stage organ failure, susceptibility to cancer and post-surgical complications [224].

Lipid droplets display a unique protein repertoire including Adipophilin and TIP47 which, further to fatty adipocytes, are expressed in lipid droplets of hepatic stellate cells. Perilipin, once deemed to be characteristic of lipid droplets in the adipocytes and steroidogenic cells, is expressed de novo in liver cells of steatotic liver in humans [225]. The finding that the distribution of perilipin is predominantly perivenous in the liver lobule [225] may suggest
a specific role of this protein in the four-step model of histogenetic events originally proposed by Wanless to account for NASH pathogenesis and its progression to cirrhosis via hepatic venular obstruction [226]. Perilipin has been confirmed by following studies as one of the most prominent upregulated genes in NAFLD [227].

In summary, alterations in the regulation of lipid droplets physiology and metabolism influence the risk of developing metabolic diseases such as T2D and NAFLD [227]. Fig. 1 illustrates the hypothesis that NAFLD is a precursor of the metabolic syndrome via insulin resistance resulting from PKCε activation which conceivably occurs in most cases of NAFLD in humans.

If the “general rule” is that steatosis and insulin resistance are associated, are there any examples of such two conditions being dissociated from each other?

5. Dissociation of NAFLD from insulin resistance and the metabolic syndrome

The first clinical evidence for conditions where high-grade steatosis occurs without insulin resistance or low-grade liver fat content is nonetheless associated with insulin resistance was reported in 2006. This conclusion was reached by comparing insulin resistance (assessed by HOMA-IR) and steatosis grade in three naturally occurring different conditions displaying a variable extent of steatosis: familial hypobetalipoproteinemia (FHBL), NAFLD and steatosis (evaluated histologically) associated with HCV infection [228]. In 2010 Amaro et al. measuring insulin resistance with the hyperinsulinemic-euglycemic clamp procedure coupled with glucose tracer infusion and liver fat content with magnetic resonance (MR) spectroscopy, confirmed the results in FHBL individuals [229].

Of interest, circumstantial evidence suggests that FHBL is associated with progressive liver disease only in those patients featuring components of the metabolic syndrome [230].

Consistent with the potential occurrence of steatosis dissociated from insulin resistance, Kantartzis et al. reported that single nucleotide polymorphisms in the diacylglycerol acyltransferase 2 (DGAT2) gene accounted for variability in liver fat content rather than for insulin resistance. These results confirm and extend to humans the finding that DGAT2 mediates the dissociation between fatty liver and insulin resistance [231]. The same group of researchers reported that the patatin-like phospholipase 3 gene (PNPLA3) is a key factor in determining whether fatty liver is associated or not with metabolic derangements such as insulin resistance [232].

Finally, PNPLA3 gene polymorphisms have recently been associated with those NAFLD cases occurring in the absence of metabolic syndrome [233], indirectly suggesting that genetic conditions which promote the development of fatty changes in the liver may occur independently of insulin resistance.

A detailed survey of those studies demonstrating a dissociation of fatty liver and T2D in both experimental conditions and humans has recently been published by Sun et al. [234]. Collectively, findings from both genetically engineered animal models and humans with genetic conditions confirm the view that the aetiology of steatosis is a major determinant of whether steatosis is associated (such as seen in most cases of NAFLD observed in clinical practice) or dissociated from insulin resistance and features of the metabolic syndrome (such as seen in NAFLD occurring in carriers of PNPLA3 gene polymorphisms and in FHBL individuals) [228,233,234]. As schematically depicted in Fig. 1, compartmentation of intrahepatocytic lipids may potentially account for the end result of NAFLD being either associated or dissociated from insulin resistance [218,219] and therefore from the risk of developing the metabolic syndrome. Once fully characterized in humans, the sub-cellular mechanisms of dissociation of steatosis from insulin resistance may become a preferential target for the prevention and management of insulin resistance and the metabolic syndrome.

6. Conclusions

A systematic review of the literature shows that the presence of NAFLD is intimately linked with the metabolic syndrome, including
T2D. Moreover, NAFLD is a strong determinant for the future development of the metabolic syndrome. This is likely to occur as a result of its being a key factor associated with insulin resistance in naturally occurring conditions in humans. Exceptions to such a theory are found in some genetic polymorphisms where the steatosis-insulin resistance-metabolic syndrome triad is dissociated in humans. The molecular biology of events dictating NAFLD being associated with or dissociated from insulin resistance is increasingly being elucidated.

On these grounds, the diagnostic importance of NAFLD as a criterion for the presence, as well as for risk of future development of metabolic syndrome, needs to be emphasized. From a therapeutic point of view, those pathogenic-based interventions aimed at reversing NAFLD are likely to be a rational approach in the prevention and treatment of hepatic insulin resistance, metabolic syndrome and related complications.

Conflict of interest
None declared.

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Appendix A. Supplementary data
Supplementary data associated with this article can be found in the online version, at http://dx.doi.org/10.1016/j.jdl.2014.09.020.

References
[38] Li X, Xia M, Ma H, et al. Liver fat content, evaluated through semi-quantitative ultrasound measurement, is associated with impaired glucose profiles: a community-based study in Chinese. PLOS ONE 2013;8:e65210.


