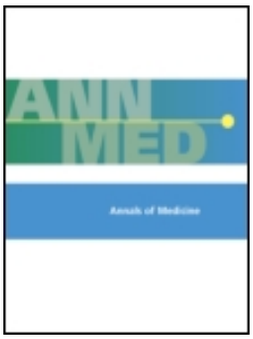

Annals of Medicine

ISSN: 0785-3890 (Print) 1365-2060 (Online) Journal homepage: informahealthcare.com/journals/iann20

# Is liver disease a threat to patients with metabolic disorders?

Giulio Marchesini, Gabriele Forlani & Elisabetta Bugianesi

**To cite this article:** Giulio Marchesini, Gabriele Forlani & Elisabetta Bugianesi (2005) Is liver disease a threat to patients with metabolic disorders?, Annals of Medicine, 37:5, 333-346, DOI: 10.1080/07853890510011445

**To link to this article:** https://doi.org/10.1080/07853890510011445

Published online: 08 Jul 2009.

*Annals of Medicine*. 2005; 37: 333–346

Taylor & Francis
Taylor & Francis Group

**REVIEW ARTICLE**

# Is liver disease a threat to patients with metabolic disorders?

GIULIO MARCHESINI[1], GABRIELE FORLANI[1] & ELISABETTA BUGIANESI[2]

[1]*Unit of Metabolic Diseases, Alma Mater Studiorum University, Bologna, and* [2]*Department of Gastroenterology, University of Turin, San Giovanni Battista Hospital, Turin, Italy*

**Abstract**
The association of metabolic disorders with liver disease is receiving increasing attention in the gastroenterological community. Cohort studies have shown that advanced liver disease may stem from metabolic disorders, via fatty liver, non-alcoholic steatohepatitis, cryptogenic cirrhosis, and eventually hepatocellular carcinoma. In both obesity and diabetes, deaths from cirrhosis are higher than expected, mainly in subjects with no or moderate alcohol consumption, but high rates of fatty liver disease have been associated with all features of the metabolic syndrome. Also the risk of hepatocellular carcinoma is higher than normal, being dependent on body mass index (BMI) in obesity, and independent of age, BMI, gender and race in diabetes. Finally, metabolic liver disease may interact with hepatitis C virus infection, increasing the risk of steatosis and liver disease progression, as well as reducing the chances of an effective antiviral treatment. There is evidence that treatments aimed at reducing insulin resistance are also effective in improving liver histology. Although cardiovascular disease remains the major cause of increased morbidity and excess mortality in metabolic disorders, the risk of progressive liver disease should no longer be underestimated, being a threat to millions of people at risk in the present epidemics of obesity and diabetes, and therapeutic strategies need to be tested.

**Key words:** *Diabetes, epidemiology, fatty liver, hepatocellular carcinoma, liver disease, metabolic syndrome*

## Introduction

The association of liver disease with metabolic disorders is receiving increased attention (1). Hepatic steatosis is a common feature in obesity and type 2 diabetes mellitus (T2DM), but its potential significance as a cause of advanced liver disease has long been underestimated. More recently, the identification of non-alcoholic fatty liver disease (NAFLD) as a clinical entity (2) and cohort studies suggesting a potential link between pure fatty liver, non-alcoholic steatohepatitis (NASH), cryptogenic cirrhosis (3), and eventually hepatocellular carcinoma (HCC) (4) have suggested that metabolic disorders *per se* may cause advanced liver failure.

Although cardiovascular risk remains the main cause accounting for increased morbidity and excess mortality in obesity and T2DM, many studies are in keeping with the hypothesis that liver failure might also be a threat to the millions of people at risk, whose number is further increasing due to the epidemics of obesity and T2DM in western countries (5). Metabolic disorders also interact with viral infections in the pathogenesis of liver failure, particularly in areas where hepatitis C (HCV) virus is endemic.

The aim of the present report is to review the existing evidence that advanced liver disease may develop in patients with metabolic disorders, and to review the pathogenic and therapeutic relevance of the association of metabolic liver disease with viral infection.

### *Search strategy and selection criteria*

We searched MEDLINE to May 2005, using different combinations of the following key-words: obesity, diabetes, survival, cirrhosis, liver disease, hepatocellular carcinoma, hepatitis B, hepatitis C, fatty liver, and related terms. We also searched the bibliographies of most recent articles for relevant references. Because of the large number of articles

Correspondence: Dr. Giulio Marchesini MD, Unit of Metabolic Diseases, Alma Mater Studiorum, University of Bologna, Azienda Ospedaliera S. Orsola-Malpighi, Via Massarenti 9, I-40138 Bologna, Italy. Fax: +39-51-6364502. E-mail: giulio.marchesini@unibo.it

ISSN 0785-3890 print/ISSN 1365-2060 online 
DOI: 10.1080/07853890510011445

**Abbreviations**

| | |
|---|---|
| ALF | acute liver failure |
| ALT | alanine aminotransferase |
| AR | attributable risk |
| AST | aspartate aminotransferase |
| BMI | body mass index |
| CC | cryptogenic cirrhosis |
| CI | confidence interval |
| DM | diabetes mellitus |
| FFA | free fatty acids |
| GGT | gamma-glutamyltransferase |
| HBV | hepatitis B virus |
| HCC | hepatocellular carcinoma |
| HCV | hepatitis C virus |
| HR | hazard ratio |
| ICD-9 | international classification of diseases ($9^{th}$ edition) |
| IRS-1 | insulin receptor substrate-1 |
| MS | metabolic syndrome |
| NAFLD | non-alcoholic fatty liver disease |
| NASH | non-alcoholic steatohepatitis |
| OR | odds ratio |
| PI-3K | phosphatidyl inosytol-3 kinase |
| PPAR | peroxisome-proliferator activated receptor |
| RR | relative risk |
| SIR | standardized incidence ratio |
| SMR | standardized mortality rate |
| T2DM | type 2 diabetes mellitus |
| TNF | tumor necrosis factor |

identified and limitations for quoting references, the final decision on what to include was based on personal judgment, with preferences for the most recent articles.

## Association of liver disease with diabetes, obesity and the metabolic syndrome

Fatty liver is the marker of metabolic liver disease, with or without raised aminotransferase levels, namely alanine aminotransferase (ALT). Although an association between the severity of liver disease and raised aminotransferases has not been unequivocally demonstrated (6,7), raised enzymes are the most common reason for patients' concern and extensive diagnostic workup. Several epidemiological data have convincingly associated the whole spectrum of NAFLD with the metabolic syndrome (MS) (6,8–10), and with its individual components as suggested by Adult Treatment Panel III proposal (11). There is now evidence that fatty liver and insulin resistance may be early predictors of metabolic disorders, even in non-obese, non-diabetic subjects, particularly in the normal-weight population (12).

**Key messages**

- Fatty liver, commonly observed in metabolic disorders, may progress to advanced liver disease, via non-alcoholic steatohepatitis, cryptogenic cirrhosis, and eventually hepatocellular carcinoma.
- The prevalence of liver disease, death rates for cirrhosis and the risk of hepatocellular carcinoma are higher than expected in subjects with features of the metabolic syndrome, particularly in obesity and diabetes, and metabolic liver disease may also interact with hepatitis C virus infection, favoring disease progression and reducing the chances of an effective antiviral treatment.
- Although cardiovascular disease remains the major cause of increased morbidity and excess mortality in metabolic disorders, the risk of progressive liver disease should no longer be underestimated, being a threat to millions of people at risk in the present epidemics of obesity and diabetes.

### *Diabetes*

The relationship of liver disease to T2DM is difficult to evaluate. End-stage liver disease can cause glucose abnormalities and overt diabetes (the so-called 'hepatogenous diabetes') (13), being a source of bias in prevalence studies. Old studies, based on liver biopsy, were also biased by the selection of cases with clinically suspected liver disease (14). Limiting the analysis to fatty liver, the estimated prevalence of diabetes ranges from 21% to 78% in recent studies, the difference being largely dependent upon diagnostic criteria (15–17). Ultrasonography is not sensitive enough to detect all cases with fatty liver infiltration, the detection limit being around 25%–30% of hepatocytes with fatty droplets (18,19). No systematic studies are available on the prevalence of diabetes in subjects with more severe histological alterations (necroinflammation, fibrosis), potentially progressive to advanced liver disease, which would require the extensive use of liver biopsy.

Surprisingly, very few cohort studies have been published on the prevalence of clinically-detectable liver disease or raised liver enzymes in subjects with T2DM. The overall incidence of elevated

aminotransferase levels in T2DM treated with oral antidiabetic agents is reported in the order of 50/10,000 person-years, but the criteria of ascertainment were questionable, and mild liver enzyme elevation was not considered (20). In the records of the Department of Veteran Affairs, the incidence of chronic non-alcoholic liver disease was doubled among patients with diabetes (21). In turn, elevated ALT (22,23) or GGT (24) were prospectively associated with the development of type 2 diabetes in the general population, and the predictive role of ALT was maintained after adjustment for insulin resistance (25).

### *Obesity*

More data are available in obesity. Ultrasound studies show evidence of hepatic steatosis in 76% of cases not drinking alcohol in toxic amounts (26), and that liver histology is compatible with NAFLD of variable severity in over 80% of morbidly obese subjects (27,28), where bariatric surgery provides an easy access to liver biopsy. Among 551 patients prospectively examined, steatosis was found in 86%, fibrosis in 74%, mild inflammation or NASH in 24%, and unexpected cirrhosis in 2% (27). In overweight subjects with abnormal aminotransferase levels but without overt liver disease, steatosis $\geqslant$10% was observed in 79% of cases, fibrosis outside the portal tracts in 30%, necroinflammation in 81% (29). The prevalence of fatty degeneration is progressively higher in relation to glucose control (from normoglycemia, to impaired glucose tolerance, to T2DM) (30,31).

A role of obesity in high aminotransferase cases has been repeatedly demonstrated. High levels of alanine and aspartate aminotransferases (ALT and AST), as well as raised gamma-glutamyltranspeptidase (GGT) activity, are frequently observed, in association with body mass index (BMI) and raised insulin levels (32,33), also in apparently healthy obese subjects with fatty liver at ultrasonography (34), the prevalence being higher in males and in relation to raised BMI. In a cohort study, Vozarova et al. (23) showed that raised liver enzymes were associated with body fat and insulin resistance, measured by the clamp technique. Prati et al. (35) confirmed that ALT levels are significantly associated with BMI in healthy individuals; their data suggested revising the upper limits of normal ALT to improve the sensitivity for identifying liver disease. In the general population, Stranges et al. (36) demonstrated an independent role of central adiposity in predicting increased levels of aminotransferases and GGT, a possible expression of an unrecognized hepatic disease.

### *Hyperlipidemia*

In hyperlipidemic patients admitted to an urban hospital-based clinic for the evaluation and management of hyperlipidemia, fatty liver was detected at ultrasounds in 50%, the prevalence being higher in subjects with severe hypertriglyceridemia and mixed hyperlipidemia (37). Hyperglycemia was an independent predictor of fatty liver, confirming the role of multiple metabolic disorders in the pathogenesis of steatosis.

### *Hypertension*

In hypertension, the prevalence of ultrasonographically-detected steatosis is more than doubled when compared to subjects with normal blood pressure (38). An association of hypertension with insulin resistance has long been documented (39). The higher prevalence of fatty liver in non-obese hypertensive patients with normal liver enzymes was related to increased insulin resistance and body weight (38).

### *Metabolic syndrome*

In a recent cross-sectional analysis of 3,405 South Korean adults (40), an association between ALT and the metabolic syndrome was found in both sexes, independently of age, BMI, smoking, alcohol drinking, and sedentary life style. The association was maintained even in the range below the current upper limits of ALT. Similarly, in 799 obese Italian patients entering a weight-reducing program, raised ALT levels were demonstrated in 21.0% of cases, with a lower prevalence of raised AST (8.6%) and GGT (13.7%). The median value of ALT increased slightly, but significantly, with obesity class ($P$ for trend: $=0.001$). Hyperglycemia ($\geqslant$110 mg/dl) and hypertriglyceridemia ($\geqslant$150 mg/dl) were the features of the metabolic syndrome most commonly associated with raised liver enzymes (41). In logistic regression analysis, after correction for age, gender, BMI and features of the metabolic syndrome, insulin resistance (homeostasis model assessment) maintained a highly predictive value for raised ALT, AST and GGT, suggesting that raised liver enzyme levels, indicative of subclinical liver disease, may be part of the insulin-resistance (metabolic) syndrome.

As previously outlined, the presence of multiple metabolic disorders is strictly associated with a higher prevalence of liver disease. Both in subjects cared for in liver units (6) and in patients observed in metabolic/diabetes units (42), the association of obesity with diabetes and/or altered lipid metabolism leads to a multiplicative effect on the final prevalence

of MS, and significantly increases the risk of more severe stages of liver disease (43–45).

## Association between diabetes and viral liver disease

### *Diabetes in HCV*

The prevalence of abnormal glucose regulation and diabetes is significantly elevated in HCV infected patients, after correction for the degree of liver cell failure. At population level (Third National Health and Nutrition Examination Survey), in persons older than 20 years of age, the presence of diabetes was over three times more prevalent in subjects with HCV infection than in those without HCV (46). In American-Indian women, increasing age, obesity, and positive HCV status were each independently related to the diagnosis of diabetes (47). Older age, obesity, severe liver fibrosis and family history of diabetes help identify HCV patients at risk for development of T2DM (48). A threefold increase in the prevalence of glucose abnormalities was observed in HCV positive patients with chronic hepatitis in comparison with HCV negative subjects (32% *versus* 12%; $P = 0.0003$) (49). In contrast, among patients with cirrhosis the difference was not statistically significant, although the prevalence of both diabetes and impaired fasting glucose (110–125 mg/dL) were more prevalent in HCV positive patients (40% *versus* 36% in HCV negative). This suggests that the connection between HCV infection and diabetes starts at early stages of hepatic disease.

### *Diabetogenic effect of HCV*

The mechanism accounting for the pro-diabetogenic effect of HCV is under investigation. In liver biopsies of HCV-infected subjects, defects in upstream insulin signaling pathways have been demonstrated at the level of insulin receptor substrate-1 (IRS-1) tyrosine phosphorylation, IRS-1/p85 phosphatidylinositol 3-kinase (PI3-kinase) and IRS-1-associated PI3-kinase enzymatic activity. They were not present in HCV negative biopsies, and might contribute to insulin resistance, which leads to progression to type 2 diabetes mellitus in patients with HCV infection (50). In a mouse model transgenic for the HCV core gene, plasma glucose levels during a glucose tolerance test were moderately higher than in control mice, in association with insulin resistance. The levels of tumor necrosis factor-$\alpha$ were also elevated, as commonly observed in human infection. The administration of an anti-tumor necrosis factor-$\alpha$ antibody restored insulin sensitivity, providing a direct experimental evidence for the contribution of HCV to the development T2DM (51).

### *Virus-induced steatosis*

The association of steatosis with HCV infection is larger than expected by chance. In patients with untreated chronic hepatitis C, steatosis is significantly associated with BMI ($P < 0.0001$) (52), namely with visceral adiposity (53), but is particularly common in genotype 3a infection (54). Steatosis also correlates with viral load (55); it disappears in subjects with sustained response to interferon and reoccurs after liver transplantation in subjects with HCV-genotype 3, whereas it does not change in patients with HCV genotype 1, irrespective of response (56), giving support for a causal association between genotype 3 infection and fat accumulation. The pathogenesis of HCV-related steatosis remains poorly understood. Serfaty et al. (57) found that cholesterol levels are lower in HCV-infected patients compared to a reference male population, as well as to patients with chronic hepatitis B or with non-alcoholic fatty liver. Hypobetalipoproteinemia is corrected by HCV eradication, suggesting a direct involvement of HCV in both hypobetalipoproteinemia and steatosis. Apoprotein AII and HCV core colocalize in human HCV-infected liver biopsies, and the hepatic overexpression of HCV core protein interferes with the hepatic assembly and secretion of triglyceride-rich very-low-density lipoproteins in a transgenic murine model (58).

### *Metabolic disorders and HCV-related disease severity*

Altered glucose regulation is significantly associated with insulin resistance and with the staging of liver fibrosis (48). In a large series of patients with chronic HCV-related hepatitis, age at infection, duration of infection, serum glucose and daily alcohol intake but not BMI were independently associated with significant fibrosis (59). Patients with high serum glucose had features of the metabolic syndrome, including a higher prevalence of steatosis, as well as faster fibrosis progression rates, suggesting that hyperglycemia is an independent co-factor of fibrosis, with a higher pro-fibrogenic impact than overweight (59). Finally, a BMI $\geqslant$30 kg/m$^2$ is a negative predictor of sustained response to interferon treatment (60). The reason(s) might be lower maximal interferon concentrations in obese patients, as well as a weaker biologic response to exogenous interferon-$\alpha$ (60,61).

Table I. Liver disease-related mortality associated with metabolic disorders.

| First author (ref) | Area | Population | Result | Comment |
|---|---|---|---|---|
| Sasaki (64) | Japan (Osaka prefecture) | Population-based study on 32,222 death certificates of patients with diabetes; observation period, 30 years | Death rate for liver cirrhosis increases from 2.1% in the period 1960–64 to 4.6% in 1985–89. | No data on other risk factors for liver disease in the population. |
| De Marco (65) | Italy (Verona Diabetes Study) | 7,158 patients with T2DM; follow-up, 5 years | SMR for cirrhosis as high as 2.5 (95% CI, 2.0–3.2), and higher than that observed for cardiovascular disease. | No correction for prevalence of hepatitis virus infection and cirrhosis prior to diabetes |
| El-Serag (66) | U S A. (Dept. of Veterans Admin. Affairs) | 173,643 patients with and 650,620 without DM; follow-up, 10–15 years. | Acute liver failure identified by ICD-9 570. The cumulative risk of ALF significantly higher among patients with diabetes (HR, 1.44; 95% CI, 1.26–1.63). This risk is maintained after excluding patients with liver disease or hepatitis during follow-up. | After excluding the first year of follow-up, diabetes increases the risk of ALF, independently of recognized underlying chronic liver disease or viral hepatitis. |
| Caldwell (67) | U S A (Liver disease registry) | 2,380 patients, 167 with NASH, 215 with CC; follow-up, 8 years. | Five middle-aged women, with no history of liver disease, died of subacute liver failure | Obesity-related liver disease may infrequently present as severe, subacute illness. |
| Jepsen (68) | Denmark (Danish National Registry) | 7,372 subjects with a diagnosis of fatty liver; follow-up, up to 16 years | All causes SMR for non-alcoholic or unspecified liver disease, 2.6 (95% CI, 2.4–2.9). In the same group, hepatobiliary SMR are as high as 19.7 (15.3–25.0). No differences in relation to T2DM. | Mortality remains increased after censoring patients upon diagnosis of liver cirrhosis. Cases with unspecified fatty liver might have biased the results |
| Ioannou (69) | U S A (NHANES I cohort) | 11,465 persons aged 25–74 years without evidence of cirrhosis at entry; mean follow-up, 12.9 years | Cirrhosis-related deaths or hospitalizations more common in obese persons (adjusted HR: 1.69; 95% CI, 1.0–3.0). The association is confined to persons with no or moderate alcohol consumption (adjusted HR: 4.1, 95% CI 1.4–11.4). | Obesity is a risk factor for cirrhosis-related death or hospitalization among persons who consume little or no alcohol. The use of a combined primary outcome is subject to criticism |
| Dam-Larsen (70) | Denmark | 215 patients with biopsy-proven pure fatty liver (109 non-alcoholic); median follow-up, 16.7 years | Survival estimates different between alcoholic and non-alcoholic fatty liver. Survival estimates in the non-alcoholic fatty liver group are not different from the Danish population. | The study supports the concept that pure fatty liver may have a benign clinical course. |

ALF=acute liver failure; BMI=body mass index; CC=cryptogenic cirrhosis; CI=confidence interval; DM=diabetes mellitus; HR=hazard ratio; ICD-9=International classification of diseases ($9^{th}$ edition); NASH=non-alcoholic steatohepatitis; SMR=standardized mortality rate; T2DM=type 2 diabetes mellitus.

## Liver disease and survival in metabolic patients

Whatever the origin of liver disease, the key question is whether liver disease may harm patients with metabolic disorders. These patients remain at very high risk of cardiovascular diseases, but evidence is emerging suggesting that liver disease needs to be tackled as well. The study of the natural history of NAFLD recently suggested that the presence of diabetes and high BMI are significantly associated with higher rates of liver fibrosis progression, also in the absence of raised aminotransferases (62). Metabolic liver disease is slowly progressive (63), and liver-related morbidity and mortality are expected to occur only in aged cohorts.

### *Liver-related mortality and cirrhosis*

The prognostic studies on liver disease-related mortality in metabolic disorders are reported in Table I

(64–70). In a registry-based cohort study, Jepsen et al. (68) studied overall and cause-specific standardized mortality ratios in Danish subjects discharged from hospital with a diagnosis of fatty liver. During follow-up, mortality was increased 2.6-fold (95% CI 2.4–2.9) in patients with non-alcoholic or unspecified fatty liver, independently of diabetes and after censoring patients upon diagnosis of liver cirrhosis. However, survival estimates in patients with pure fatty liver, without inflammation, were not different from those observed in the general Danish population (70), suggesting the need for additional factors favoring disease progression. In T2DM patients observed in the Verona cohort study, the standardized mortality ratio for cirrhosis during a 5-year follow-up was remarkably high (2.5; 95% CI, 2.0–3.2) (65), the ratio being even higher than that reported for cardiovascular disease. Less clear data had previously been reported in a Japanese cohort with a low prevalence of diabetes (64). Contrary to what is commonly believed, obesity is frequently associated with cirrhosis, even in subjects without significant alcohol intake. In a multicenter Italian study of 1402 patients with cirrhosis 29% of females and 18% of males appeared to be overnourished (71). In obesity-related cryptogenic cirrhosis, severe liver disease was as frequent as in HCV-related cirrhosis, and survival was even lower than in age- and sex-matched cirrhosis of viral origin (72). These data were not confirmed in a U S series of NASH patients, where prognosis was either similar or less severe than in HCV-cirrhosis (73). In a long-term follow-up of the NHANES I cohort, cirrhosis-related deaths or hospitalizations were more common in obese persons (adjusted hazard ratio, 1.7; 95% CI, 1.0–3.0), after exclusion of subjects with evidence of cirrhosis at entry or during the first 5 years of follow-up (69). The association was particularly strong among obese persons who did not consume alcohol (4.1; 1.4–11.4). In the presence of cirrhosis, diabetes is a risk factor for mortality (13), the larger mortality rate being related to an increased risk of hepatocellular failure, heralded by low platelet counts and high bilirubin (73), not to diabetes or diabetes-related complications. In subjects followed for NAFLD, overall mortality and mortality related to liver disease (relative risk, 22.8; $P = 0.003$) were both more common in subjects with diabetes (74). Obesity (67) and T2DM (66) also increase the risk of acute liver failure in a few cases. On the basis of the clinical and histological findings, Caldwell and Hespenheide (67) suggested that these patients had undiagnosed NASH with silent progression to cirrhosis followed by subacute liver failure.

*Hepatocellular carcinoma (HCC)*

The prognostic studies on progression to HCC and HCC-related mortality in metabolic disorders are reported in Table II (64,75–89). In a recent mortality study of the American Cancer Society, the risk of death from HCC during 16 years of follow-up was increased both in males ($P$ for trend $<0.001$) and in females ($P < 0.04$) with increasing BMI class (83). These data expand previous reports linking obesity to HCC (75,90).

An association of T2DM with HCC was first established by comparison with the prevalence of diabetes observed in subjects with other tumors (91). It has been confirmed in several recent reports (21,77,80,86,92). In a case-control study, El-Serag et al. (81) found that diabetes increases the risk of HCC in the presence of hepatitis B, hepatitis C, or alcoholic cirrhosis, whereas in chronic HCV-related hepatitis, steatosis *per se* was a risk for HCC (93). The temporal relationship between diabetes and HCC is important, due to the high prevalence of diabetes in cirrhosis. In a large cohort of subjects admitted to Veteran Administration hospitals (87), the incidence of HCC was over twice as high among patients with diabetes, and was higher in subjects with a longer follow-up. Diabetes was a risk factor independent of age, BMI, gender and race, with a hazard ratio of 2.16. A high BMI further increases the risk of HCC. In the Verona cohort, where 70% of cases were overweight or obese, the hazard ratio of HCC was 1.86 (95% CI, 1.43–2.38) (84). The majority of these studies have been carried out on referred cohorts, and are subject to referral biases. Only recently have population-based studies become available. In a Korean cohort with low average BMI (23.2 kg/m$^2$, only approximately 25% overweight or obese), the age-adjusted incidence rate of HCC increased progressively with increasing fasting glucose and BMI, reaching the top in men with diabetes (hazard ratio, 1.66; 95% CI, 1.53–1.79) (88). In this population with high HBV carrier state, neither NAFLD nor HBV were demonstrated as confounders, suggesting a primary role of hyperglycemia. In a large U S Medicare study in subjects aged 65 years or over, the risk of HCC was increased 2 to 3 times in the presence of diabetes, after adjustment for HBV, HCV, alcohol and hemocromatosis, and did not change in analyses restricted to subjects with diabetes detected between two and three years prior to HCC diagnosis (89).

Table II. Progression to hepatocellular carcinoma and HCC-related mortality in metabolic disorders.

| Author (ref) | Area | Population | Result | Comment |
|---|---|---|---|---|
| Sasaki (64) | Japan (Osaka prefecture) | Population-based study on 32,222 death certificates of patients with diabetes; observation period, 30 years | Death rate for HCC increases from 0.5% in the period 1960–64 to 3.2% in 1985–89. | No data on other risk factors for HCC in the population. The risk of ascertainment bias is elevated. |
| Moller (75) | Denmark | 37,957 obese persons, compared to the total Danish population; follow-up, 4.8 years. | Relative risk for HCC, 1.9 (95% CI, 1.5–2.5). Results are unchanged by restriction to a subcohort of 8,207 persons where obesity is the primary discharge diagnosis, and are also similar irrespective of first year of follow-up. | Overall, the incidence of cancer is increased by 16% in the cohort. Selection bias is not likely to have influenced the results. |
| Adami (76) | Sweden (Swedish in-patient and Cancer Registry) | 153,852 patients with diabetes compared to nationwide estimates; follow-up, 6–24 years | SIR for HCC in diabetes, 4.1 (95% CI, 3.8–4.5). The risk is higher in men (4.7, 4.2–52) than in women (3.4, 2.9–3.9). | After exclusion of subjects with alcoholism, cirrhosis and hepatitis, the risk continues to be three-fold increased. |
| La Vecchia (77) | Italy | Point-prevalence case-control study (428 with incident HCC and 1,502 controls); observation period, 13 years | In diabetes, OR for HCC, 2.1 (95% CI, 1.4–3.2) after adjustment for confounders, including history of hepatitis and liver cirrhosis, body mass index and history of liver cancer in first-degree relatives. | A history of diabetes mellitus can explain about 8% (95% CI 5–11) of cases of liver cancer in this population. |
| Wideroff (78) | Denmark | 109,581 hospitalized subjects with diabetes; follow-up, 4–16 years. | SIR for HCC, 4.0 (95%, 3.5–4.6) in males and 2.1 (95% CI=1.6–2.7) in females. SIR remains elevated after exclusion of patients with reported risk factors (e.g., cirrhosis and hepatitis) or patients whose cancers were diagnosed at autopsy. | Patients hospitalized with a diagnosis of diabetes are at higher risk of developing HCC. The elevated risk may be confounded by obesity. |
| Braga (79) | Italy (Northern Area) | Case-control of 320 HCC and 1,408 controls; observation period, 1984–93) | Population-ARs of HCC, 18% for cirrhosis, 16% for hepatitis, 8% for diabetes and 7% for heavy alcohol consumption. Compared with females, males had higher AR for cirrhosis (21% *versus* 11%), diabetes (10% *versus* 2%) and heavy alcohol intake (9% *versus* 1%). | Even if available information on dietary factors was limited, and the AR were based on arbitrary assumptions, diabetes control might significantly reduce the burden of HCC in Northern Italy. |
| Lagiou (80) | Greece | Point-prevalence case-control study (333 incident HCC and 360 controls); 4-year observation period. | History of diabetes associated with HCC (adjusted OR, 1.86; 95% CI, 0.99–3.51). The association is not confounded by any major risk factor (hepatitis B and C, alcohol consumption) | The authors speculate that diabetes may increase HCC risk either via hyperinsulinemia, or via NASH and non-alcoholic cirrhosis |
| El-Serag (81) | U S A (Dept. of Veterans Affairs) | Point-prevalence, case-control study (823 HCC, 3,459 controls); observation period, 3 years | In diabetes adjusted OR for HCC, 1.57(95% CI, 1.08–2.28) only in the presence of other etiologic factors (virus, alcohol). The combined presence of HCV and alcoholic cirrhosis further increases the risk (OR 79.2, 60.3–103.4). | Diabetes increases the risk of HCC only in the presence of other risk factors. Alcohol exerts a primary role in this population |
| Nair (82) | U S A (National transplant registry) | 19,271 patients with cirrhosis (28% obese; 19% alcoholic, 13% cryptogenic cirrhosis) | OR for HCC in obese patients with alcoholic cirrhosis (3.2; 95% CI, 1.5-6.6) and cryptogenic cirrhosis (11.1, 1.5-87.4) | No effect of obesity on HCC in cirrhosis of different etiology |

Table II. (*Continued.*)

| Author (ref) | Area | Population | Result | Comment |
|---|---|---|---|---|
| Calle (83) | U S A (American Cancer Society Study) | Prospective study of 900,053 adults; follow-up, 16 years | Death risk for HCC increased both in males (P for trend $<0.001$) and in females ($P<0.04$) with increasing BMI class. Compared to normal weight, a BMI in the range 30-35 kg/$m^2$ gives a RR of 1.90 (95% CI, 1.46-2.47) in males and 1.40 (0.97-2.00) in females. | Increased body weight is associated with increased death rates for all cancers combined and for cancers at multiple specific sites. The risk for HCC is particularly relevant in severely obese persons |
| Verlato (84) | Italy (Verona Diabetes Study) | 7,148 patients with T2DM; follow-up, 10 years | SMR for overall malignancies increased in females, but not in men. Excess mortality from hepatic cancer (SMR, 1.86, 95% CI, 1.44-2.38) observed in both men and women. | No adjustment for other etiologic factors (alcohol, virus) |
| Sorensen (85) | Denmark (Danish National Registry) | 7,326 subjects with fatty liver; follow-up, 1-16 years | SIR for HCC elevated in patients with alcoholic (9.5, 95% CI, 5.7-14.8) as well as non-alcoholic fatty liver (4.4; 1.2-11.4), compared with general Danish population. | Patients were censored upon diagnosis of cirrhosis; accordingly, the potential sequence of fatty liver to cirrhosis to HCC was missed. |
| Regimbeau (86) | France | 210 patients with liver resection for HCC; 18 (8.6%) with no underlying cause of liver disease | The prevalence of obesity (50% *vs.* 14%) and diabetes (56% *vs.* 11%) are higher in patients with cryptogenic liver disease than in patients chronic viral hepatitis ($P<0.0001$). | Obesity and diabetes may be important risk factors for cryptogenic chronic liver disease progressing to HCC |
| Coughlin (87) | U S A (Cancer Prevention Study II) | 467,922 men and 588,321 women; follow-up, 16 years | Diabetes significantly associated with fatal HCC in men (RR, 2.19; 95% CI, 1.76-2.72), but not in women (RR, 1.37; 0.94-2.00) | The association is not explained by high body mass index, suggesting that diabetes is an independent predictor of mortality from cancer |
| Jee (88) | Korea (National Health Insurance Co.) | 1,298,385 Koreans; follow-up, up to 10 years | Death rates from all cancers combined increased in subjects with fasting glucose $>$ 140 mg/dl (HR, 1.29; 95% CI, 1.22-1.37 in men and 1.23, 1.09-1.39 in women). By cancer site, the association is confirmed for HCC (Males: HR, 1.59; 1.45-1,74; Females: 1.28, 1.00-1.66) | In Koreans, a population with a low BMI, elevated fasting serum glucose levels and a diagnosis of diabetes are independent risk factors for HCC. |
| Davila (89) | U S A (Medicare database) | 2,061 HCC and 6,183 controls; population-based, case-control study; age$\geqslant$65 years; observation period, 1994-99 | After adjustment for HBV, HCV, alcohol and hemochromatosis, OR for diabetes, 2.87 (95% CI, 2.49-3.30). Similar data in analysis restricted to diabetes between 2-3 years from HCC | Diabetes is independently associated with HCC, regardless of other major risk factors in the elderly. Several features of MS also increase HCC risk. |

AR=attributable risk; BMI=body mass index; CI=confidence interval; HCC=hepatocellular carcinoma; HR=hazard ratio; MS=metabolic syndrome; NASH=non-alcoholic steatohepatitis; OR=odds ratio; RR=relative risk; SIR=standardized incidence ratio; SMR=standardized mortality rate; T2DM=type 2 diabetes mellitus.

Thus diabetes may be considered an additional risk factor for HCC, when other major risk factors are ruled out.

Finally, the association of NAFLD with HCC is well known, and is probably mediated by cryptogenic cirrhosis (4,72,85,93–95). Decreased surveillance (96) might be implicated in the poor risk of subjects with NAFLD-related HCC, compared with HCC occurring in cirrhosis of different etiology (72).

*Proposed mechanism(s) for liver disease and disease progression*

The mechanism(s) implicated in fatty liver have been outlined in several recent reviews (16,97–99); they have insulin resistance as the key factor (Figure 1). The family clustering of diseases strongly supports a genetic inheritance of decreased insulin sensitivity, but the search for genes has so far been disappointing. Polygenic transmission is very likely in metabolic disorders, and determining the genetics

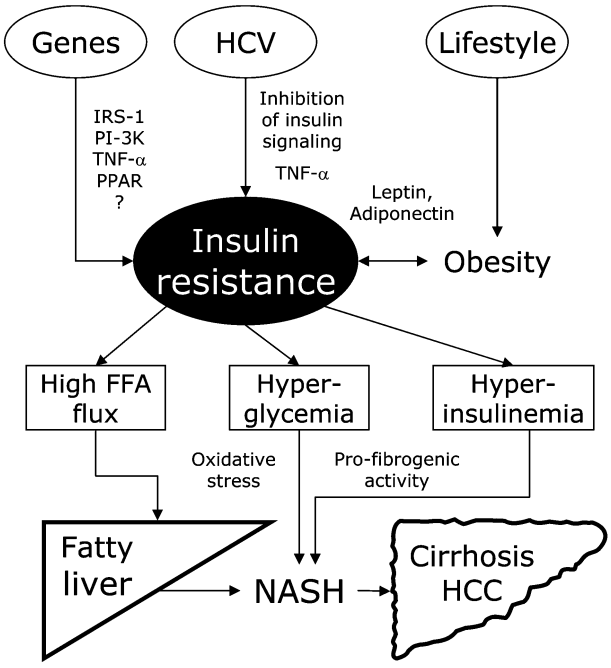


Figure 1. Interaction between genes, virus and lifestyle in the initiation and progression of metabolic liver disease. Insulin resistance is the core of the mechanism. Genes, through various and largely undefined polymorphisms, may cause or favor insulin resistance. Hepatitis-C virus, either by its viral genome, or via increased TNF-α production, may interfere with insulin signaling. Finally lifestyle, through obesity, adipokines and increased release of TNF-α, produces or enhances insulin resistance. Increased FFA flux promotes steatosis (possibly aggravated by HCV-dependent interference with the hepatic assembly and secretion of triglyceride-rich very-low-density lipoproteins); hyperglycemia induces oxidative stress, hyperinsulinemia favors fibrosis and hepatic hyperplasia, both leading from fatty liver to non-alcoholic steatohepatitis, fibrosis, cirrhosis and eventually to hepatocellular carcinoma. FFA=free fatty acids; HCV=hepatitis C virus; TNF=tumor necrosis factor; NASH=non-alcoholic steatohepatitis; HCC=hepatocellular carcinoma; IRS-1=insulin receptor substrate-1; PI-3K=phosphatidyl inosytol-3 kinase; PPAR=peroxisome-proliferator activated receptor.

of insulin resistance will not be easy (100,101). There are hundreds of gene interactions involved in fat storage (102) and in insulin action (103), and abnormalities in at least 23 genes have been detected in NAFLD (103). Further studies should also identify the site of insulin resistance (liver *versus* adipose tissue and skeletal muscle). Lifestyle maintains the primary role in fatty liver, via excessive food intake and low physical exercise, promoting obesity and further decreasing insulin sensitivity. Finally, the contribution of HCV in selected populations should not be forgotten. Disease progression and carcinogenesis may stem from hyperglycemia, either generating oxidative stress (104–106) or facilitating septal fibrosis (59). Also hyperinsulinemia and increased insulin-like growth factor-1 (107–109) have been associated with fibrogenesis and the generation of hepatic hyperplasia, through the up-regulation of connective-tissue growth factor in stellate cells demonstrated in human liver biopsies and in *in vitro* experiments (108). Finally, obesity *per se* may increase oxidative stress, causing a dysregulation of adipocytokines promoting the development of MS (110).

## Treatment of liver disease in metabolic patients

The liver disease of subjects cared for in metabolic/diabetic units is by no means less severe than that observed in hepatology units (42). Although the burden of cardiovascular disease largely outweighs that of liver failure, progressive liver disease is expected to reduce cardiovascular risk, by reducing several diabetes-related risk factors (arterial pressure, hyperlipidemia, platelets and clotting factors) (111). As a consequence, these subjects may ultimately be at risk because of their liver disease, and treatment may be worthwhile.

### *NAFLD treatment*

Several review articles are available on NAFLD treatment (112). Therapeutic options range from vitamins, antioxidants and cytoprotective agents to lipid-lowering drugs, phlebotomy and insulin-sensitizers (112,113). In all NAFLD subjects, weight-reducing programs and life-style interventions remain the first line of treatment (Table III) (114–120). They were reported as effective in reducing aminotransferase levels and improving liver histology in the short-term (116,120), but the results were not long-lasting. Similar approaches are being considered as background treatment in HCV-positive patients with obesity and/or T2DM before and during anti-viral therapy.

Insulin-sensitizing agents have proved effective as well (121–127), independently of glucose levels, with a single exception (126). Data on short-term treatment (121,124,128) have now been expanded to one year with both metformin (127) and glitazones (122,123); aminotransferase levels return within normal values, steatosis at histology is markedly reduced, apparently more with the use of glitazones (129), and also necroinflammation and fibrosis improve, but aminotransferase tends to return to pre-treatment levels after stopping treatment (121,124). We need long-term, placebo-controlled trials to validate therapeutic interventions on lifestyle and pharmacological treatment against

Table III. Clinical studies on treatment of non-alcoholic fatty liver disease with lifestyle/weight loss modifications and insulin-sensitizers, published in extension in peer-reviewed journals.

| Treatment First author (ref) | No. of cases | Type of study | Duration of treatment | Result |
|---|---|---|---|---|
| **Weight loss or lifestyle modifications** | | | | |
| Eriksson (114) | 3 | Observational | 6–12 months | Weight loss decreases ALT and improves histology. |
| Palmer (115) | 39 | Observational | Unspecified | In overweight adults, weight loss > 10% corrects hepatic abnormalities |
| Ueno (116) | 15 | Controlled against cases who refused treatment (n=10) | 3 months | Weight-losing diet and intensive physical activity. Biochemistry and histology (only steatosis) significantly better in the experimental arm |
| Franzese (117) | 38 | Observational | 6 months | In obese children, ALT and 'bright liver' improve with weight loss |
| Knobler (118) | 49 | Observational | 24 months | Improved biochemistry with dietary intervention and drugs (if indicated to treat associated diseases) |
| Kugelmans (119) | 16 | Observational, with/ without vitamin E | 3 months | Improved biochemistry with lifestyle changes (diet and physical exercise); no extra advantage of vitamin E |
| Hickman (120) | 14 | Observational | 15 months | Improved biochemistry, health-related quality of life, and histology with lifestyle modifications |
| **Insulin-sensitizers** | | | | |
| *Glitazones* | | | | |
| Caldwell (121) | 10 | Observational | 6 months | Biochemistry improves significantly: no significant differences in histology |
| Neuschwander – Tetri (122) | 25 | Observational | 12 months | Both biochemistry and histology (including fibrosis and necroinflammation) improve |
| Promrat (123) | 18 | Observational | 12 months | Both biochemistry and histology (including fibrosis and necroinflammation) improve |
| *Metformin* | | | | |
| Marchesini (124) | 14 | Controlled against cases who refused treatment (n=6) | 4 months | Metformin significantly improves ALT, reduces liver volume and decreases insulin resistance |
| Uygun (125) | 17 | RCT *versus* diet alone (n=17) | 6 months | Metformin significantly better on ALT. Liver histology similarly improves in both arms |
| Nair (126) | 15 | Observational | 12 months | No differences in the long term |
| Bugianesi (127) | 55 | RCT *versus* prescriptive diet or vitamin E (n=55) | 12 months | All subjects received nutritional counseling. Metformin significantly favors the general improvement in ALT . Histology (including fibrosis and necroinflammation) improves in a subgroup of subjects of the metformin arm |

ALT=Alanine aminotransferase; RCT=randomized controlled trial.

hard outcomes (disease progression to cirrhosis and ultimately death).

## Conclusions

Epidemiological studies suggest that 20% to 30% of adults in U S and other Western countries have excess fat accumulation in the liver, that about 10% of these individuals meet current diagnostic criteria for NASH, and that one third of them progress to fibrosis and cryptogenic cirrhosis (113). These estimates may become even higher in the next few years, considering the rapid spread of obesity and T2DM in the general population (5), thus increasing the global burden of theoretically-preventable metabolic diseases in Western countries (130).

Prevention and treatment strategies of metabolic liver disease are largely based on the same approaches used to tackle the metabolic syndrome, but we all know how difficult it is to implement lifestyle changes (131). Cognitive approaches to the treatment of patients with the metabolic syndrome need to include liver disease as an additional complication, preventable by a healthy lifestyle. More importantly, physicians need to consider the importance of surveillance for detection and early treatment of complications. The potential for disease prevention and health gain from tackling major known risk factors is substantial in metabolic disorders (132),

and also the risk of progressive liver disease should no longer be underestimated in these subjects.

## Conflict of interests

The authors declare that they have no conflict of interests in relation to this review.

## References


1. Tolman KG, Fonseca V, Tan MH, Dalpiaz A. Narrative review: hepatobiliary disease in type 2 diabetes mellitus. Ann Intern Med. 2004;141:946–56.
2. Ludwig J, Viaggiano TR, McGill DB, Oh BJ. Nonalcoholic steatohepatitis: Mayo Clinic experience with an hitherto unnamed disease. Mayo Clin Proc. 1980;55:434–8.
3. Falck-Ytter Y, Younossi ZM, Marchesini G, McCullough AJ. Clinical features and natural history of nonalcoholic steatosis syndromes. Semin Liver Dis. 2001;21:17–26.
4. Bugianesi E, Leone N, Vanni E, Marchesini G, Brunello F, Carucci P, et al. Expanding the natural history of nonalcoholic steatohepatitis: From cryptogenic cirrhosis to hepatocellular carcinoma. Gastroenterology. 2002;123:134–40.
5. Mokdad AH, Ford ES, Bowman BA, Dietz WH, Vinicor F, Bales VS, et al. Prevalence of obesity, diabetes, and obesity-related health risk factors, 2001. JAMA. 2003;289:76–9.
6. Marchesini G, Bugianesi E, Forlani G, Cerrelli F, Lenzi M, Manini R, et al. Nonalcoholic fatty liver, steatohepatitis, and the metabolic syndrome. Hepatology. 2003;37:917–23.
7. Mofrad P, Contos MJ, Haque M, Sargeant C, Fisher RA, Luketic VA, et al. Clinical and histologic spectrum of nonalcoholic fatty liver disease associated with normal ALT values. Hepatology. 2003;37:1286–92.
8. Marchesini G, Brizi M, Morselli-Labate AM, Bianchi G, Bugianesi E, McCullough AJ, et al. Association of nonalcoholic fatty liver disease with insulin resistance. Am J Med. 1999;107:450–5.
9. Cortez-Pinto H, Camilo ME, Baptista A, De Oliveira AG, De Moura MC. Non-alcoholic fatty liver: another feature of the metabolic syndrome? Clin Nutr. 1999;18:353–8.
10. Chitturi S, Abeygunasekera S, Farrell GC, Holmes-Walker J, Hui J, Fung C, et al. NASH and insulin resistance: insulin secretion and specific association with the insulin resistance syndrome. Hepatology. 2002;35:373–9.
11. Expert Panel on Detection Evaluation and Treatment of High Blood Cholesterol in Adults. Executive summary of the third report of the National Cholesterol Education Program (NCEP) expert panel on detection, evaluation, and treatment of high blood cholesterol in adults (Adult Treatment Panel III). JAMA. 2001;285:2486–97.
12. Kim HJ, Lee KE, Kim DJ, Kim SK, Ahn CW, Lim SK, et al. Metabolic significance of nonalcoholic fatty liver disease in nonobese, nondiabetic adults. Arch Intern Med. 2004;164:2169–75.
13. Bianchi G, Marchesini G, Zoli M, Bugianesi E, Fabbri A, Pisi E. Prognostic significance of diabetes in patients with cirrhosis. Hepatology. 1994;20:119–25.
14. Creutzfeldt W, Frerichs H, Sickinger K. Liver diseases and diabetes mellitus. Prog Liver Dis. 1970;3:371–407.
15. Kumar KS, Malet PF. Nonalcoholic steatohepatitis. Mayo Clin Proc. 2000;75:733–9.
16. Angulo P. Nonalcoholic fatty liver disease. N Engl J Med. 2002;346:1221–31.
17. Clark JM, Brancati FL, Diehl AM. Nonalcoholic fatty liver disease. Gastroenterology. 2002;122:1649–57.
18. Saverymuttu SH, Joseph AE, Maxwell JD. Ultrasound scanning in the detection of hepatic fibrosis and steatosis. BMJ. 1986;292:13–5.
19. Ricci C, Longo R, Gioulis E, Bosco M, Pollesello P, Masutti F, et al. Noninvasive in vivo quantitative assessment of fat content in human liver. J Hepatol. 1997;27:108–13.
20. Jick SS, Stender M, Myers MW. Frequency of liver disease in type 2 diabetic patients treated with oral antidiabetic agents. Diabetes Care. 1999;22:2067–71.
21. El-Serag HB, Tran T, Everhart JE. Diabetes increases the risk of chronic liver disease and hepatocellular carcinoma. Gastroenterology. 2004;126:460–8.
22. Ohlson LO, Larsson B, Bjorntorp P, Eriksson H, Svardsudd K, Welin L, et al. Risk factors for type 2 (non-insulin-dependent) diabetes mellitus. Thirteen and one-half years of follow-up of the participants in a study of Swedish men born in 1913. Diabetologia. 1988;31:798–805.
23. Vozarova B, Stefan N, Lindsay RS, Saremi A, Pratley RE, Bogardus C, et al. High alanine aminotransferase is associated with decreased hepatic insulin sensitivity and predicts the development of type 2 diabetes. Diabetes. 2002;51:1889–95.
24. Perry IJ, Wannamethee SG, Shaper AG. Prospective study of serum gamma-glutamyltransferase and risk of NIDDM. Diabetes Care. 1998;21:732–7.
25. Hanley AJ, Williams K, Festa A, Wagenknecht LE, D'Agostino RB, Jr., Kempf J, et al. Elevations in markers of liver injury and risk of type 2 diabetes: the insulin resistance atherosclerosis study. Diabetes. 2004;53: 2623–32.
26. Bellentani S, Saccoccio G, Masutti F, Croce LS, Brandi G, Sasso F, et al. Prevalence of and risk factors for hepatic steatosis in Northern Italy. Ann Intern Med. 2000; 132:112–7.
27. Marceau P, Biron S, Hould FS, Marceau S, Simard S, Thung SN, et al. Liver pathology and the metabolic syndrome X in severe obesity. J Clin Endocrinol Metab. 1999;84:1513–7.
28. Dixon JB, Bhathal PS, O'Brien PE. Nonalcoholic fatty liver disease: predictors of nonalcoholic steatohepatitis and liver fibrosis in the severely obese. Gastroenterology. 2001;121: 91–100.
29. Ratziu V, Giral P, Charlotte F, Bruckert E, Thibault V, Theodorou I, et al. Liver fibrosis in overweight patients. Gastroenterology. 2000;118:1117–23.
30. Silverman JF, Pories WJ, Caro JF. Liver pathology in diabetes mellitus and morbid obesity. Clinical, pathological, and biochemical considerations. Pathol Annu. 1989;24: 275–302.
31. Silverman JF, O'Brien KF, Long S, Leggett N, Khazanie PG, Pories WJ, et al. Liver pathology in morbidly obese patients with and without diabetes. Am J Gastroenterol. 1990;85:1349–55.
32. Clark JM, Brancati FL, Diehl AM. The prevalence and etiology of elevated aminotransferase levels in the United States. Am J Gastroenterol. 2003;98:960–7.
33. Ruhl CE, Everhart JE. Determinants of the association of overweight with elevated serum alanine aminotransferase activity in the United States. Gastroenterology. 2003;124: 71–9.
34. Hsiao TJ, Chen JC, Wang JD. Insulin resistance and ferritin as major determinants of nonalcoholic fatty liver disease in apparently healthy obese patients. Int J Obes Relat Metab Disord. 2004;28:167–72.

35. Prati D, Taioli E, Zanella A, Della Torre E, Butelli S, Del Vecchio E, et al. Updated definitions of healthy ranges for serum alanine aminotransferase levels. Ann Intern Med. 2002;137:1–10.
36. Stranges S, Dorn JM, Muti P, Freudenheim JL, Farinaro E, Russell M, et al. Body fat distribution, relative weight, and liver enzyme levels: A population-based study. Hepatology. 2004;39:754–63.
37. Assy N, Kaita K, Mymin D, Levy C, Rosser B, Minuk G. Fatty infiltration of liver in hyperlipidemic patients. Dig Dis Sci. 2000;45:1929–34.
38. Donati G, Stagni B, Piscaglia F, Venturoli N, Morselli-Labate AM, Rasciti L, et al. Increased prevalence of fatty liver in arterial hypertensive patients. Role of insulin resistance. Gut. 2004;53:1020–3.
39. Ferrannini E, Buzzigoli G, Bonadonna R, Giorico MA, Oleggini M, Graziadei L, et al. Insulin resistance in essential hypertension. N Engl J Med. 1987;317:350–7.
40. Jeong SK, Nam HS, Rhee JA, Shin JH, Kim JM, Cho KH. Metabolic syndrome and ALT: a community study in adult Koreans. Int J Obes Relat Metab Disord. 2004;28:1033–8.
41. Marchesini G, Avagnina S, Barantani EG, Ciccarone AM, Corica F, Dall'Aglio E, et al. Aminotransferase and gamma-glutamyltranspeptidase levels in obesity are associated with insulin resistance and the metabolic syndrome. J Endocrinol Invest. 2005;28:333–9.
42. Marchesini G, Bugianesi E, Forlani G, Marzocchi R, Zannoni C, Vanni E, et al. Non-alcoholic steatohepatitis in patients cared in metabolic units. Diabetes Res Clin Pract. 2004;63:143–51.
43. Bugianesi E, Manzini P, D'Antico S, Vanni E, Longo F, Leone N, et al. Relative contribution of iron burden, HFE mutations and insulin resistance to fibrosis in nonalcoholic fatty liver. Hepatology. 2004;39:179–87.
44. Papadia FS, Marinari GM, Camerini G, Murelli F, Carlini F, Stabilini C, et al. Liver damage in severely obese patients: a clinical-biochemical-morphologic study on 1,000 liver biopsies. Obes Surg. 2004;14:952–8.
45. Angulo P, Keach JC, Batts KP, Lindor KD. Independent predictors of liver fibrosis in patients with nonalcoholic steatohepatitis. Hepatology. 1999;30:1356–62.
46. Mehta SH, Brancati FL, Sulkowski MS, Strathdee SA, Szklo M, Thomas DL. Prevalence of type 2 diabetes mellitus among persons with hepatitis C virus infection in the United States. Hepatology. 2001;33:1554.
47. Wilson C. Hepatitis C infection and type 2 diabetes in American-Indian women. Diabetes Care. 2004;27:2116–9.
48. Petit JM, Bour JB, Galland-Jos C, Minello A, Verges B, Guiguet M, et al. Risk factors for diabetes mellitus and early insulin resistance in chronic hepatitis C. J Hepatol. 2001;35:279–83.
49. Lecube A, Hernandez C, Genesca J, Esteban JI, Jardi R, Simo R. High prevalence of glucose abnormalities in patients with hepatitis C virus infection: a multivariate analysis considering the liver injury. Diabetes Care. 2004; 27:1171–5.
50. Aytug S, Reich D, Sapiro LE, Bernstein D, Begum N. Impaired IRS-1/PI3-kinase signaling in patients with HCV: a mechanism for increased prevalence of type 2 diabetes. Hepatology. 2003;38:1384–92.
51. Shintani Y, Fujie H, Miyoshi H, Tsutsumi T, Tsukamoto K, Kimura S, et al. Hepatitis C virus infection and diabetes: direct involvement of the virus in the development of insulin resistance. Gastroenterology. 2004;126:840–8.
52. Hourigan LF, Macdonald GA, Purdie D, Whitehall VH, Shorthouse C, Clouston A, et al. Fibrosis in chronic hepatitis C correlates significantly with body mass index and steatosis. Hepatology. 1999;29:1215–9.
53. Adinolfi LE, Gambardella M, Andreana A, Tripodi MF, Utili R, Ruggiero G. Steatosis accelerates the progression of liver damage of chronic hepatitis C patients and correlates with specific HCV genotype and visceral obesity. Hepatology. 2001;33:1358–64.
54. Monto A, Alonzo J, Watson JJ, Grunfeld C, Wright TL. Steatosis in chronic hepatitis C: relative contributions of obesity, diabetes mellitus, and alcohol. Hepatology. 2002;36:729–36.
55. Rubbia-Brandt L, Giostra E, Mentha G, Quadri R, Negro F. Expression of liver steatosis in hepatitis C virus infection and pattern of response to alpha-interferon. J Hepatol. 2001;35: 307.
56. Kumar D, Farrell GC, Fung C, George J. Hepatitis C virus genotype 3 is cytopathic to hepatocytes: Reversal of hepatic steatosis after sustained therapeutic response. Hepatology. 2002;36:1266–72.
57. Serfaty L, Andreani T, Giral P, Carbonell N, Chazouilleres O, Poupon R. Hepatitis C virus induced hypobetalipoproteinemia: a possible mechanism for steatosis in chronic hepatitis C. J Hepatol. 2001;34:428–34.
58. Perlemuter G, Sabile A, Letteron P, Vona G, Topilco A, Chretien Y, et al. Hepatitis C virus core protein inhibits microsomal triglyceride transfer protein activity and very low density lipoprotein secretion: a model of viral-related steatosis. FASEB J. 2002;16:185–94.
59. Ratziu V, Munteanu M, Charlotte F, Bonyhay L, Poynard T. Fibrogenic impact of high serum glucose in chronic hepatitis C. J Hepatol. 2003;39:1049–55.
60. Bressler BL, Guindi M, Tomlinson G, Heathcote J. High body mass index is an independent risk factor for non-response to antiviral treatment in chronic hepatitis C. Hepatology. 2003;38:639–44.
61. Lam NP, Pitrak D, Speralakis R, Lau AH, Wiley TE, Layden TJ. Effect of obesity on pharmacokinetics and biologic effect of interferon-alpha in hepatitis C. Dig Dis Sci. 1997;42:178–85.
62. Adams LA, Sanderson S, Lindor KD, Angulo P. The histological course of nonalcoholic fatty liver disease: a longitudinal study of 103 patients with sequential liver biopsies. J Hepatol. 2005;42:132–8.
63. Poynard T, Mathurin P, Lai CL, Guyader D, Poupon R, Tainturier MH, et al. A comparison of fibrosis progression in chronic liver diseases. J Hepatol. 2003;38:257–65.
64. Sasaki A. Mortality and causes of death in patients with diabetes mellitus in Japan. Diabetes Res Clin Pract. 1994;24 Suppl:S299–306.
65. de Marco R, Locatelli F, Zoppini G, Verlato G, Bonora E, Muggeo M. Cause-specific mortality in type 2 diabetes. The Verona Diabetes Study. Diabetes Care. 1999;22:756–61.
66. El-Serag HB, Everhart JE. Diabetes increases the risk of acute hepatic failure. Gastroenterology. 2002;122:1822–8.
67. Caldwell SH, Hespenheide EE. Subacute liver failure in obese women. Am J Gastroenterol. 2002;97:2058–62.
68. Jepsen P, Vilstrup H, Mellemkjaer L, Thulstrup AM, Olsen JH, Baron J, et al. Prognosis of patients with a diagnosis of fatty liver - a registry-based cohort study. Hepatogastroenterology. 2003;50:2101–4.
69. Ioannou GN, Weiss NS, Kowdley KV, Dominitz JA. Is obesity a risk factor for cirrhosis-related death or hospitalization? a population-based cohort study. Gastroenterology. 2003;125:1053–9.
70. Dam-Larsen S, Franzmann M, Andersen IB, Christoffersen P, Jensen LB, Sorensen TI, et al. Long term

prognosis of fatty liver: risk of chronic liver disease and death. Gut. 2004;53:750–5.
71. Italian Multicentre Cooperative Project on Nutrition in Liver Cirrhosis. Nutritional status in cirrhosis. J Hepatol. 1994;21:317–25.
72. Ratziu V, Bonyhay L, Di Martino V, Charlotte F, Cavallaro L, Sayegh-Tainturier MH, et al. Survival, liver failure, and hepatocellular carcinoma in obesity-related cryptogenic cirrhosis. Hepatology. 2002;35:1485–93.
73. Hui JM, Kench JG, Chitturi S, Sud A, Farrell GC, Byth K, et al. Long-term outcomes of cirrhosis in nonalcoholic steatohepatitis compared with hepatitis C. Hepatology. 2003;38:420–7.
74. Younossi ZM, Gramlich T, Matteoni CA, Boparai N, McCullough AJ. Nonalcoholic fatty liver disease in patients with type 2 diabetes. Clin Gastroenterol Hepatol. 2004;2:262–5.
75. Moller H, Mellemgaard A, Lindvig K, Olsen JH. Obesity and cancer risk: a Danish record-linkage study. Eur J Cancer. 1994;30A:344–50.
76. Adami HO, Chow WH, Nyren O, Berne C, Linet MS, Ekbom A, et al. Excess risk of primary liver cancer in patients with diabetes mellitus. J Natl Cancer Inst. 1996;88:1472–7.
77. La Vecchia C, Negri E, Decarli A, Franceschi S. Diabetes mellitus and the risk of primary liver cancer. Int J Cancer. 1997;73:204–7.
78. Wideroff L, Gridley G, Mellemkjaer L, Chow WH, Linet M, Keehn S, et al. Cancer incidence in a population-based cohort of patients hospitalized with diabetes mellitus in Denmark. J Natl Cancer Inst. 1997;89:1360–5.
79. Braga C, La Vecchia C, Negri E, Franceschi S. Attributable risks for hepatocellular carcinoma in northern Italy. Eur J Cancer. 1997;33:629–34.
80. Lagiou P, Kuper H, Stuver SO, Tzonou A, Trichopoulos D, Adami HO. Role of diabetes mellitus in the etiology of hepatocellular carcinoma. J Natl Cancer Inst. 2000;92:1096–9.
81. El-Serag HB, Richardson PA, Everhart JE. The role of diabetes in hepatocellular carcinoma: a case-control study among United States Veterans. Am J Gastroenterol. 2001;96:2462–7.
82. Nair S, Mason A, Eason J, Loss G, Perrillo RP. Is obesity an independent risk factor for hepatocellular carcinoma in cirrhosis? Hepatology. 2002;36:150–5.
83. Calle EE, Rodriguez C, Walker-Thurmond K, Thun MJ. Overweight, obesity, and mortality from cancer in a prospectively studied cohort of U.S. adults. N Engl J Med. 2003;348:1625–38.
84. Verlato G, Zoppini G, Bonora E, Muggeo M. Mortality from site-specific malignancies in type 2 diabetic patients from Verona. Diabetes Care. 2003;26:1047–51.
85. Sorensen HT, Mellemkjaer L, Jepsen P, Thulstrup AM, Baron J, Olsen JH, et al. Risk of cancer in patients hospitalized with fatty liver: a Danish cohort study. J Clin Gastroenterol. 2003;36:356–9.
86. Regimbeau JM, Colombat M, Mognol P, Durand F, Abdalla E, Degott C, et al. Obesity and diabetes as a risk factor for hepatocellular carcinoma. Liver Transpl. 2004;10:S69–73.
87. Coughlin SS, Calle EE, Teras LR, Petrelli J, Thun MJ. Diabetes mellitus as a predictor of cancer mortality in a large cohort of US adults. Am J Epidemiol. 2004;159:1160–7.
88. Jee SH, Ohrr H, Sull JW, Yun JE, Ji M, Samet JM. Fasting serum glucose level and cancer risk in Korean men and women. JAMA. 2005;293:194–202.
89. Davila JA, Morgan RO, Shaib Y, McGlynn KA, El-Serag HB. Diabetes increases the risk of hepatocellular carcinoma in the United States: a population based case control study. Gut. 2005;54:533–9.
90. Wolk A, Gridley G, Svensson M, Nyren O, McLaughlin JK, Fraumeni JF, et al. A prospective study of obesity and cancer risk (Sweden). Cancer Causes Control. 2001;12:13–21.
91. Lawson DH, Gray JM, McKillop C, Clarke J, Lee FD, Patrick RS. Diabetes mellitus and primary hepatocellular carcinoma. QJM. 1986;61:945–55.
92. Huo TI, Lui WY, Huang YH, Chau GY, Wu JC, Lee PC, et al. Diabetes mellitus is a risk factor for hepatic decompensation in patients with hepatocellular carcinoma undergoing resection: a longitudinal study. Am J Gastroenterol. 2003;98:2293–8.
93. Ohata K, Hamasaki K, Toriyama K, Matsumoto K, Saeki A, Yanagi K, et al. Hepatic steatosis is a risk factor for hepatocellular carcinoma in patients with chronic hepatitis C virus infection. Cancer. 2003;97:3036–43.
94. Yang S, Lin HZ, Hwang J, Chacko VP, Diehl AM. Hepatic hyperplasia in noncirrhotic fatty livers: is obesity-related hepatic steatosis a premalignant condition? Cancer Res. 2001;61:5016–23.
95. Zen Y, Katayanagi K, Tsuneyama K, Harada K, Araki I, Nakanuma Y. Hepatocellular carcinoma arising in non-alcoholic steatohepatitis. Pathol Int. 2001;51:127–31.
96. Marrero JA, Fontana RJ, Su GL, Conjeevaram HS, Emick DM, Lok AS. NAFLD may be a common underlying liver disease in patients with hepatocellular carcinoma in the United States. Hepatology. 2002;36:1349–54.
97. Medina J, Fernandez-Salazar LI, Garcia-Buey L, Moreno-Otero R. Approach to the pathogenesis and treatment of nonalcoholic steatohepatitis. Diabetes Care. 2004;27:2057–66.
98. Bugianesi E, Zannoni C, Vanni E, Marzocchi R, Marchesini G. Non-alcoholic fatty liver and insulin resistance: a cause-effect relationship? Dig Liver Dis. 2004;36:165–73.
99. Browning JD, Horton JD. Molecular mediators of hepatic steatosis and liver injury. J Clin Invest. 2004;114:147–52.
100. Groop L. Genetics of the metabolic syndrome. Br J Nutr. 2000;83 Suppl 1:S39–48.
101. Rich SS, Concannon P. Challenges and strategies for investigating the genetic complexity of common human diseases. Diabetes. 2002;51 Suppl 3:S288–94.
102. Ashrafi K, Chang FY, Watts JL, Fraser AG, Kamath RS, Ahringer J, et al. Genome-wide RNAi analysis of Caenorhabditis elegans fat regulatory genes. Nature. 2003;421:268–72.
103. Sreekumar R, Rosado B, Rasmussen D, Charlton M. Hepatic gene expression in histologically progressive non-alcoholic steatohepatitis. Hepatology. 2003;38:244–51.
104. Marfella R, Quagliaro L, Nappo F, Ceriello A, Giugliano D. Acute hyperglycemia induces an oxidative stress in healthy subjects. J Clin Invest. 2001;108:635–6.
105. Esposito K, Nappo F, Marfella R, Giugliano G, Giugliano F, Ciotola M, et al. Inflammatory cytokine concentrations are acutely increased by hyperglycemia in humans: role of oxidative stress. Circulation. 2002;106:2067–72.
106. Guha M, Bai W, Nadler JL, Natarajan R. Molecular mechanisms of tumor necrosis factor alpha gene expression in monocytic cells via hyperglycemia-induced oxidant stress-dependent and -independent pathways. J Biol Chem. 2000;275:17728–39.
107. Svegliati-Baroni G, Ridolfi F, Di Sario A, Casini A, Marucci L, Gaggiotti G, et al. Insulin and insulin-like

growth factor-1 stimulate proliferation and type I collagen accumulation by human hepatic stellate cells: differential effects on signal transduction pathways. Hepatology. 1999;29:1743–51.
108. Paradis V, Perlemuter G, Bonvoust F, Dargere D, Parfait B, Vidaud M, et al. High glucose and hyperinsulinemia stimulate connective tissue growth factor expression: a potential mechanism involved in progression to fibrosis in nonalcoholic steatohepatitis. Hepatology. 2001;34:738–44.
109. Hickman IJ, Powell EE, Prins JB, Clouston AD, Ash S, Purdie DM, et al. In overweight patients with chronic hepatitis C, circulating insulin is associated with hepatic fibrosis: implications for therapy. J Hepatol. 2003;39: 1042–8.
110. Furukawa S, Fujita T, Shimabukuro M, Iwaki M, Yamada Y, Nakajima Y, et al. Increased oxidative stress in obesity and its impact on metabolic syndrome. J Clin Invest. 2004;114:1752–61.
111. Marchesini G, Ronchi M, Forlani G, Bugianesi E, Bianchi G, Fabbri A, et al. Cardiovascular disease in cirrhosis–a point-prevalence study in relation to glucose tolerance. Am J Gastroenterol. 1999;94:655–62.
112. Bugianesi E, Marzocchi R, Villanova N, Marchesini G. Non-alcoholic fatty liver disease/non-alcoholic steatohepatitis (NAFLD/NASH): treatment. Best Pract Res Clin Gastroenterol. 2004;18:1105–16.
113. Neuschwander-Tetri BA, Caldwell SH. Nonalcoholic steatohepatitis: Summary of an AASLD Single Topic Conference. Hepatology. 2003;37:1202–19.
114. Eriksson S, Eriksson KF, Bondesson L. Nonalcoholic steatohepatitis in obesity: a reversible condition. Acta Med Scand. 1986;220:83–8.
115. Palmer M, Schaffner F. Effect of weight reduction on hepatic abnormalities in overweight patients. Gastroenterology. 1990;99:1408–13.
116. Ueno T, Sugawara H, Sujaku K, Hashimoto O, Tsuji R, Tamaki S, et al. Therapeutic effects of restricted diet and exercise in obese patients with fatty liver. J Hepatol. 1997;27:103–7.
117. Franzese A, Vajro P, Argenziano A, Puzziello A, Iannucci MP, Saviano MC, et al. Liver involvement in obese children. Ultrasonography and liver enzyme levels at diagnosis and during follow-up in an Italian population. Dig Dis Sci. 1997;42:1428–32.
118. Knobler H, Schattner A, Zhornicki T, Malnick SD, Keter D, Sokolovskaya N, et al. Fatty liver–an additional and treatable feature of the insulin resistance syndrome. QJM. 1999;92:73–9.
119. Kugelmas M, Hill DB, Vivian B, Marsano L, McClain CJ. Cytokines and NASH: a pilot study of the effects of lifestyle modification and vitamin E. Hepatology. 2003;38:413–9.
120. Hickman IE, Jonsson JR, Prins JB, Ash S, Purdie DM, Clouston AD, et al. Modest weight loss and physical activity in overweight patients with chronic liver disease results in sustained improvements in alanine aminotransferase, fasting insulin, and quality of life. Gut. 2004;53:413–9.
121. Caldwell SH, Hespenheide EE, Redick JA, Iezzoni JC, Battle EH, Sheppard BL. A pilot study of a thiazolidinedione, troglitazone, in nonalcoholic steatohepatitis. Am J Gastroenterol. 2001;96:519–25.
122. Neuschwander-Tetri BA, Brunt EM, Wehmeier KR, Oliver D, Bacon BR. Improved nonalcoholic steatohepatitis after 48 weeks of treatment with the PPAR-gamma ligand rosiglitazone. Hepatology. 2003;38:1008–17.
123. Promrat K, Lutchman G, Uwaifo GI, Freedman RJ, Soza A, Heller T, et al. A pilot study of pioglitazone treatment for nonalcoholic steatohepatitis. Hepatology. 2004;39:188–96.
124. Marchesini G, Brizi M, Bianchi G, Tomassetti S, Zoli M, Melchionda N. Metformin in non-alcoholic steatohepatitis. Lancet. 2001;358:893–4.
125. Uygun A, Kadayifci A, Isik AT, Ozgurtas T, Deveci S, Tuzun A, et al. Metformin in the treatment of patients with non-alcoholic steatohepatitis. Aliment Pharmacol Ther. 2004;19:537–44.
126. Nair S, Diehl AM, Wiseman M, Farr GH, Jr., Perrillo RP. Metformin in the treatment of non-alcoholic steatohepatitis: a pilot open label trial. Aliment Pharmacol Ther. 2004;20:23–8.
127. Bugianesi E, Gentilcore E, Manini R, Natale S, Vanni E, Villanova N, et al. A randomized controlled trial of metformin versus vitamin E or prescriptive diet in non-alcoholic fatty liver disease. Am J Gastroenterol. 2005; 100:1082–90.
128. Neuschwander-Tetri BA, Brunt EM, Wehmeier KR, Sponseller CA, Hampton K, Bacon BR. Interim results of a pilot study demonstrating the early effects of the PPAR-gamma ligand rosiglitazone on insulin sensitivity, aminotransferases, hepatic steatosis and body weight in patients with non-alcoholic steatohepatitis. J Hepatol. 2003;38:434–40.
129. Tiikkainen M, Hakkinen AM, Korsheninnikova E, Nyman T, Makimattila S, Yki-Jarvinen H. Effects of rosiglitazone and metformin on liver fat content, hepatic insulin resistance, insulin clearance, and gene expression in adipose tissue in patients with type 2 diabetes. Diabetes. 2004;53:2169–76.
130. Ezzati M, Lopez AD, Rodgers A, Vander Hoorn S, Murray CJ. Selected major risk factors and global and regional burden of disease. Lancet. 2002;360:1347–60.
131. Marchesini G, Trovati M. Type 2 diabetes and the Naaman syndrome. Diabetes Care. 2003;26:3195.
132. Ezzati M, Hoorn SV, Rodgers A, Lopez AD, Mathers CD, Murray CJ. Estimates of global and regional potential health gains from reducing multiple major risk factors. Lancet. 2003;362:271–80.