Hepatoma Research

Review

Epidemiology of non-alcoholic fatty liver disease-related hepatocellular carcinoma: a western perspective

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Abstract

Non-alcoholic fatty liver disease (NAFLD) is now the most common cause of liver disease worldwide, and represents an increasingly important cause of hepatocellular carcinoma (HCC). As the prevalence of NAFLD has increased, the burden of NAFLD-related HCC has been rising in parallel. This is particularly evident in Western countries, where NAFLD is estimated to account for 10%-59% of all HCC. NAFLD-related HCC can occur in the presence or absence of cirrhosis, and, while those with cirrhosis remain at the greatest risk, factors such as steatohepatitis, age, genetic polymorphisms, type 2 diabetes mellitus and obesity also appear have an impact on the risk of developing HCC in NAFLD. In this review, we present the epidemiology of NAFLD-related HCC from a Western perspective, highlighting gaps in current knowledge and future directions for research in this field.

Keywords: Non-alcoholic fatty liver disease, steatohepatitis, cirrhosis, hepatocellular carcinoma, epidemiology

INTRODUCTION

Over the past two decades, non-alcoholic fatty liver disease (NAFLD) has rapidly become the most common cause of liver disease worldwide, affecting approximately a quarter of the global population[1]. It is considered to be a hepatic end organ effect of the metabolic syndrome and its rise to prominence has

coincided with the onset of the obesity and type 2 diabetes mellitus (T2DM) epidemics[2]. Previously, due to the strong association between NAFLD and the metabolic syndrome, NAFLD was considered a disease of affluent nations; however, with the epidemiological transition and increasing urbanisation in low and middle income countries, prevalence of the metabolic syndrome and NAFLD is now increasing rapidly across the globe[3-5]. This high prevalence and subsequent burden of progressive liver disease has resulted in NAFLD becoming an increasingly important cause of hepatocellular carcinoma (HCC)[6-8]. This review outlines the epidemiology of NAFLD-related HCC, with a focus on Western countries.

METHODS

We conducted a search of the Medline, PubMed and Cochrane databases to identify English language, original research articles conducted in adults over the age of 18, and published between 1 January 1990 and 1 September 2019 using the Medical Subject Headings "Nonalcoholic Fatty Liver Disease" and "Hepatocellular carcinoma". Duplicate results were removed using Endnote X9, before screening by title and abstract was performed to find research relating to HCC in patients with NAFLD (single reviewer - AF). Original research articles were eligible for selection, including meta-analyses, controlled trials, cohort studies and case-series, which clearly described their study population including their diagnostic criteria for NAFLD, and described the incidence or prevalence of NAFLD-related HCC.

NAFLD, nonalcoholic steatohepatitis and the natural history of disease

NAFLD is a broad term used to describe a spectrum of liver diseases characterised by excessive hepatic fat accumulation with associated insulin resistance, in the absence of a secondary cause or significant alcohol consumption[9,10]. More precisely, NAFLD is defined as the presence of steatosis in > 5% of hepatocytes histologically or by proton density fat fraction on magnetic resonance imaging (MRI-PDFF), and it can be subclassified into nonalcoholic fatty liver (NAFL) or nonalcoholic steatohepatitis (NASH)[9,10]. While NAFL refers to simple steatosis, NASH is characterised by hepatic lobular inflammation and is associated with an increased risk of progression to fibrosis, cirrhosis and hepatic decompensation[9]. NASH can only reliably be differentiated from NAFL histologically, and the prevalence of NASH in liver biopsies from NAFLD patients is estimated to be 59.10% (95%CI: 47.55%-69.73%) from pooled NASH prevalence data[1]. This estimate is higher than previous estimates, suggesting NASH occurred in approximately one third of NAFLD patients; however, retrospective biopsy studies are inherently affected by selection bias of patients in whom there is high clinical suspicion for steatohepatitis[11,12].

Fibrosis is an important prognostic factor in NAFLD. Fibrosis stage is independently associated with mortality, liver transplantation and liver-related events, with fibrosis occurring at twice the rate in patients with NASH compared with NAFL[13,14]. Furthermore, liver-related mortality increases significantly with increasing fibrosis stage[15]. In a meta-analysis of 411 patients from 11 cohort studies with biopsy proven NAFLD (2145.5 person-years of follow-up), the annual fibrosis progression rate in patients with NAFL and stage 0 fibrosis at baseline was 0.07 stages per year (95%CI: 0.02-0.11 stages/year), and 0.14 stages per year in NASH (95%CI: 0.07-0.21 stages per year)[16]. This is the equivalent of one stage of progression over 14.3 years (95%CI: 9.1-50 years) in NAFL *vs.* 7.1 years (95%CI: 4.8-14.3 years) for NASH. In patients with NASH, approximately 20% of those with advanced (F3) fibrosis will progress to cirrhosis, and 20% of compensated NASH cirrhotics will develop hepatic decompensation over a two-year period[17,18].

Current global trends in HCC

Liver cancer, of which HCC is the main subtype, has the sixth highest cancer incidence, and is the fourth leading cause of cancer-related death worldwide[19]. There are significant geographic variations in the incidence of HCC, reflecting the differences in aetiology of liver diseases in each region[20]. In Asian countries where the highest incidence rates of HCC are reported (20 per 100,000 in men and 6.9 in

women), viral hepatitis, particularly hepatitis B, accounts for the majority of cases with the population attributable fraction due to hepatitis B approximately 51%-57%[21,22]. Comparatively, in high income regions of the world such as Western Europe, North America and Australasia, where the predominant aetiologies are hepatitis C and alcohol, HCC incidence rates are much lower[8]. In Europe, the age-standardised incidence rate is estimated at 6.8 per 100,000 population in men and 2.2 in women, while, in the US, the overall incidence is 11.6 per 100,000 population in men and 4.3 per 100,000 in women[21,23]. This is on the background of an almost three-fold increase in HCC cases in the United States over the past two decades[24,25]. Australia and New Zealand have also demonstrated a significant increase in HCC with a 7.5-fold increase between 1982 and 2014[26]. In 2015, the Australian incidence of HCC was 7.6 per 100,000 persons, again with higher rates in men (12 per 100,000) than women (3.9 per 100,000), which is expected to have increased to 8.6 cases per 100,000 persons in 2019[27]. This increase is likely due in part to an aging population, with a high prevalence of hepatitis C; however, the rise of the obesity epidemic, and an associated increase in the prevalence of NAFLD-related HCC, may also be contributing[28,29]. Furthermore, within these Western countries, particularly in the US, NAFLD, fibrosis and HCC are more prevalent in some ethnic sub-populations. This has been seen in Hispanic, Pacific Islander and subcontinental Indian groups, whereas other sub-populations, such as African Americans, appear to have lower rates despite similar risk factors[30-32]. Whether this difference is due to disparities in socioeconomic status or diet remains to be seen; however, there may also be inherent biological differences with a higher prevalence of the PNPLA3 genetic polymorphism noted in the Hispanic population[33].

Prevalence of NAFLD-related HCC

NAFLD has become the most rapidly increasing cause of HCC in Western countries, with the proportion of HCC attributable to NAFLD increasing significantly over the past two decades[34,35]. This shift has mirrored the rise of obesity and the metabolic syndrome, while also coinciding with the development of effective treatments for viral hepatitis and greater coverage of hepatitis B vaccination programmes[6]. This increase in NAFLD-related HCC is most pronounced in Western nations: in the BRIDGE study, a multiregional, large-scale longitudinal cohort study of consecutive newly diagnosed HCC cases, NAFLD accounted for 10%-12% of HCC cases in North America and Europe, but only 1%-6% in Asian countries[7]. In North America, NAFLD is a common cause of HCC. In a large early cohort study of HCC patients identified from a healthcare claims database in the USA, NAFLD was found to be the most common aetiology, accounting for 59% of their 4406 cases[36]. However, subsequent similar US-based longitudinal cohort studies have reported a much lower proportion of NAFLD-related HCC. The Surveillance, Epidemiology, and End Results (SEER) cancer registry database found that NAFLD was the predominant aetiology in 14.1% of their 4929 HCC cases; this was even lower in the Veteran Affairs (VA) Hospitals cohort, where 8% of 1500 HCCs were attributable to NAFLD[8,37]. This may reflect differences in the diversity of the groups studied here, with the VA group enriched with older males with hepatitis C. NAFLD is now the second leading cause of liver transplantation for HCC in the United States after Hepatitis C virus (HCV), and is the most rapidly growing indication for HCC-related liver transplantation, having increased from 8.3% in 2002 to 13.5% in 2012[38]. Moreover, the number of people with NAFLD-related HCC on liver transplant waiting lists in the United States has also risen from 2.1% in 2002 to 17.9% in 2017[38,39].

European and Australasian studies have also described an increase in the prevalence of NAFLD-related HCC. In the UK, between 2000 and 2010 there was a 10-fold increase in NAFLD-related HCC, with histologically or radiologically proven NAFLD accounting for 34.8% of all HCC cases in 2010 in one cohort of 633 patients[34]. Data from the European Transplant Registry also show an increase in transplantation for NAFLD-related HCC from 0.2% in 2007 to 1.2% in 2017[40]. In Australia and New Zealand, NAFLD is the third leading cause of HCC in those who underwent transplantation, and an Australian cohort of 272 HCC patients found NAFLD to be the underlying aetiology with 14% of cases[41,42]. A summary of these cohorts is outlined in Table 1.

Table 1. Overview of trials

Author	Country	Design/Method	Number of participants	Definition of NAFLD	Results	Proportion of NAFLD with cirrhosis (%)	Limitations/Bias
Marrero et al.[69]	USA	Single centre retrospective cohort of consecutive HCC cases	105	Histology	Cryptogenic cirrhosis 2nd leading cause of HCC (n = 30, 29%) and 6 of those cases had histological features of NASH, while 8 had clinical features of NAFLD	N/A	Retrospective study design
Sanyal et al.[47]	USA	Single centre prospective cohort of patients with NASH cirrhosis	152	Histologic features of steatohepatitis and cirrhosis	NASH had a lower risk of developing hepatocellular carcinoma compared to those with HCV cirrhosis (10/149 patients vs. 25/147 patients at risk; P < 0.01)	100%	Small cohort, with low numbers of HCC and comparator group was those with treatment naïve HCV or treatment non-responders in pre DAA therapy era
Paradis et al.[70]	France	Retrospective review of HCC liver resection specimens between 1995 and 2007	128	N/A	65.5% of cases with Met Sy associated HCC had F0-2 fibrosis	N/A	Surgical resection bias could account for high proportion of patients without significant fibrosis
Yatsuji et al.[46]	Japan	Prospective cohort study of patients with NASH and HCV cirrhosis	68	Histologic features of steatohepatitis and cirrhosis	5-year HCC rate was 11.3% for NASH and 30.5% for HCV	100%	Small cohort, with low numbers of HCC and comparator group was those with treatment naïve HCV or treatment non-responders in pre DAA therapy era
Ascha et al.[44]	USA	Prospective analysis of patients with NASH and HCV cirrhosis between 2003 and 2007	195	Histologic features of NASH or cryptogenic cirrhosis in the presence of metabolic syndrome and without a history of significant alcohol intake	NASH Cirrhotics had a yearly cumulative incidence of HCC 2.6% c/w 4% in HCV cirrhosis NASH patients were older at diagnosis of cirrhosis (56.6 years vs. 48.2 years)	100%	Small cohort, with low numbers of HCC and comparator group was those with treatment naïve HCV or treatment non-responders in pre DAA therapy era
Sanyal et al.[36]	USA	Retrospective review of HCC cases identified from a large healthcare claims database between 2002 and 2008	4406	Documentation of diagnosis based on ICD-9 coding	NAFLD (59%) most common aetiology of HCC followed by diabetes (36%) Of 729,018 patients with NAFLD/ NASH codes, 19,217 (2.7%) had HCC at one time (incident or prevalent case)	46%	Use of coding criteria likely underestimates NAFLD cases when estimating incidence or prevalence
Bhala et al.[45]	Australia, USA, UK, Italy	Prospective multicentre natural history study of NAFLD patients with advanced fibrosis or Child's Pugh A cirrhosis	247	Histologic features of advanced fibrotic NAFLD as well as elevated aminotransferases for at least 6 months	HCC occurred more commonly in patients with HCV cirrhosis than in NAFLD [n = 18 (6.8%) vs. 6 (2.4%); P = 0.03]	52.2%	Small cohort, with low numbers of HCC and comparator group was those with treatment naïve HCV or treatment non-responders in pre DAA therapy era
Ertle et al.[50]	Germany	Single centre review of new HCC cases within a 12-month period between 2007 and 2008	162	NAFLD/NASH was defined according to the histological features of NASH, when available or cryptogenic cirrhosis in the presence of MetSy and without a history of significant alcohol intake	NAFLD and NASH most common aetiology 24% followed by HCV 23.3% but cryptogenic made up 15.3%	58.3%	Small cohort. All NASH patients were cirrhotic with no non-cirrhotic NASH controls

Study	Country	Study type	No. of patients	Diagnostic criterion	%	Findings	Limitations
Wong *et al.*[38]	USA	Retrospective cohort study of OLT recipients on organ registry for HCC in US from 2002 to 2012.	10,061	Documented diagnosis of NASH or obese patients with cryptogenic/ unknown aetiology of liver disease	N/A	NASH was 2nd leading cause of OLT for HCC (8.3% in 2002 *vs.* 10.3% in 2007 *vs.* 13.5% in 2012) behind HCV but the most rapidly growing indication (4× increase in NASH, 2× increase in HCV) Non-NASH-related HCC patients were more likely to be men (79.2% *vs.* 70.6%) and were younger at time of OLT (57.2 ± 7.6 *vs.* 59.3 ± 7.3) NASH-related patients had higher BMI (33.6 ± 4.3 *vs.* 27.3 ± 4.9) and higher rates of diabetes (42.8% *vs.* 20.8%)	Used a modified diagnostic criterion for NASH. Did not present proportion of cirrhotic patients. Transplant patients tend to have high adherence and engagement with medical care, which may account for the older and heavier patients undergoing transplant with NASH
Dyson *et al.*[34]	UK	Retrospective longitudinal cohort of consecutive HCC cases between 2000 and 2010	632	Evidence of a fatty liver on biopsy or imaging, with an otherwise negative liver screen, drinking < 21 or < 14 units of alcohol per week respectively for at least 5 years prior to their first presentation with liver disease	77.2%	NAFLD accounted for 34.8% of HCC cases NAFLD patients were older (71.3 *vs.* 67) Survival similar to other aetiologies but higher incidental presentation (38.2%) and lower prevalence of cirrhosis (77.2%)	Retrospective single centre study in a region which high rates of obesity and diabetes
Park *et al.*[7]	Multi-regional	The BRIDGE study. Observational longitudinal cohort in 42 countries between 2005 and 2012	18,301 (Asia 67% of patients, Europe 20% and North America 13%)	Medical chart review	N/A	Proportion of HCC attributable to NAFLD: North America 12%, Europe 10%, China 1%, Taiwan 5%, South Korea 6%	Multiple centres in different regions without a clear definition of NAFLD used
Younossi *et al.*[8]	USA	Longitudinal cohort study using SEER registries to identify patients with HCC between 2004 and 2009	4929	ICD codes NAFLD also defined by clinical diagnosis (cryptogenic cirrhosis, obese-diabetics with cryptogenic liver disease)	N/A	14.1% of HCC linked to NAFLD and a 9% annual increase in NAFLD-related HCC NAFLD HCC were older, had shorter survival time, more heart disease and more likely to die from primary liver cancer Only 5% who received transplant post HCC were for NAFLD	Coding information can underestimate NAFLD and obesity
Mittal *et al.*[37]	USA	Retrospective cohort analysis of patients with HCC in US Veterans Affairs hospitals between 2005 and 2010	1500	Histology or presence of the metabolic syndrome in the absence of other causes of liver disease	58.3%	NAFLD HCC accounted for 8% of HCC 1-year survival comparable between aetiologies	The vast majority of the cohort were male, which does not represent the general population
Perumpail *et al.*[71]	USA	Case series of HCC patients	44	Radiological or histological evidence of hepatic steatosis, with no causes for secondary hepatic fat accumulation such as significant alcohol consumption, use of steatogenic medication or hereditary disorders	N/A	9 cases of HCC in the absence of cirrhosis, and 6 were attributable to NAFLD/NASH	Very small case series

Study	Country	Methods	N	NAFLD definition	Results	%	Limitations
Piscaglia et al.[49]	Italy	Multicentre observational prospective cohort of patients with NAFLD-related HCC	145	NAFLD if all other known aetiologies of liver disease could be ruled out and if consistent present or past histological or ultra sonographic features of fatty liver and alcohol intake < 30 g/day	46.2% of NAFLD associated HCC were non-cirrhotic compared to 2.8% in those with HCV 73.1% of NAFLD-related HCC patients have diabetes mellitus compared with only 24.9% of HCV-related HCC patients NAFLD HCC detected at later stage, had larger volume and more infiltrative pattern compared to HCV-related HCC Non-significant survival difference NAFLD 28.5 months vs. 35 months in HCV	53.8%	Median follow up of 13 months so longer term survival results may not be truly reflective of the general population
Mohamad et al.[48]	USA	Retrospective analysis of patients diagnosed with HCC and NAFLD at the Cleveland Clinic between 2003 and 2012	83	Histologically	43.4% of NAFLD-related HCC occurred with a non-cirrhotic liver NAFLD HCC were older and more likely to present with a single nodule, larger tumour and more likely to undergo resection, but did have higher rates of recurrence	56.6%	Retrospective case series with selection bias for patients who underwent liver biopsy
Kanwal et al.[32]	USA	Prospective longitudinal cohort of NAFLD patients with matched controls	296,707	2 or more elevated ALT values (> 40 IU/mL for men and > 31 IU/mL for women) in the outpatient setting in the absence of other causes of liver disease or any ICD codes for alcohol use	Risk of HCC 0.21/1000 PYs in those with NAFLD HR: 7.62; 95%CI: 5.76-10.09 NAFLD with cirrhosis a/w higher risk 10.6/1000 PYs 20% of NAFLD associated HCC were not cirrhotic	80%	ALT is not always an accurate diagnostic marker for NAFLD and this method may be underestimate prevalence in both NAFLD and control group
Hong et al.[42]	Australia	Multicentre prospective cohort of newly diagnosed HCC between 2012 and 2013	272	Medical chart review	NAFLD associated HCC accounted for 14% of cases	N/A	No clear definition of NAFLD used
Bengtsson et al.[52]	Sweden	Single centre retrospective cohort study of all HCC patients at Karolinska University Hospital Stockholm from 2004 to 2017	225	NAFLD defined by no other cause of liver disease and one of: (1) Liver biopsy supporting NAFLD; (2) Radiological features supporting steatosis; (3) BMI ≥ 25 and T2DM or BMI ≥ 30	Non-cirrhotic NAFLD-HCC were older (74 years vs. 70 years), had a lower prevalence of T2DM (66% vs. 80%), larger tumours, less frequently underwent liver transplantation (0% vs. 11%) and more frequently underwent resection (35% vs. 8%) compared to those with cirrhosis Mortality was similar (aHR for non-cirrhotic NAFLD-HCC vs. cirrhotic NAFLD-HCC 0.93, 95%CI: 0.58-1.51)	63%	Retrospective design and cohort derived from a tertiary referral centre which may be affected by referral bias, with those not suitable for curative intervention not referred for review

NASH: nonalcoholic steatohepatitis; HCC: hepatocellular carcinoma; NAFLD: non-alcoholic fatty liver disease; BMI: body mass index; HCV: hepatitis C virus; SEER: surveillance, epidemiology, and end results; HR: hazard ratio; aHR: adjusted hazard ratio; OLT: orthotopic liver transplantation; T2DM: type 2 diabetes mellitus; ALT: alanine aminotransferase; LT: in Wong USA 2012 please change the 'LT' in the methods column to OLT; DAA: direct-acting antiviral agents; ICD: international classification of diseases; N/A: not applicable; PYs: person years

Incidence of HCC in NAFLD

Although NAFLD represents an increasing proportion of HCC cases, the true risk of HCC in NAFLD patients with and without cirrhosis is not clear. As HCC is an infrequent outcome of chronic liver disease, there are limited studies with the adequate cohort size and duration of follow up required to accurately assess incidence rates. As a result, several meta-analyses have been performed to pool data and increase the sample size. A systematic review by White *et al.*[43] found the risk of HCC in NAFLD patients to be in the range of 0%-38% over a median of 5-10 years of follow up. More recently, Younossi *et al.*[1] performed a meta-analysis to evaluate the global prevalence and outcomes of NAFLD, and calculated an annual HCC incidence of 0.44 per 1000 person years (95%CI: 0.29-0.66) in patients with NAFLD, with much higher rates in those with NASH, 5.29 per 1000 person years (95%CI: 0.75-37.56). These meta-analyses are somewhat limited by the heterogeneity of the included studies, with small retrospective cohorts and inconsistent methods of defining NAFLD. As a result, a large United States longitudinal study led by Kanwal *et al.*[32] compared 296,707 patients with NAFLD to 296,707 matched healthy controls to determine the incidence of HCC in these cohorts. NAFLD was defined by persistently elevated transaminases (> 40 IU/mL for men and > 31 IU/mL for women, with a minimum of two results at least six months apart), in the absence of any significant alcohol intake or another cause of liver disease. The healthy controls had normal liver function tests, no other history of liver disease and minimal alcohol intake. The overall incidence of HCC in the NAFLD patients was low, 0.21 per 1000 patient years; however, HCC incidence was much higher in patients with NAFLD cirrhosis, 10.6 per 1000 patient years. Overall, cirrhosis in NAFLD appears to be the most significant risk factor for HCC [Figure 1], with approximately 6%-13% of patients with NAFLD cirrhosis developing a HCC during 3-10 years of follow up in a number of prospective observational cohorts[44-47].

NAFLD-related HCC in the absence of cirrhosis

NAFLD patients without liver cirrhosis are also at increased risk of HCC compared to those without liver disease[32,43]. This is particularly concerning given the high population prevalence of NAFLD, and the implications for HCC screening and increasing disease burden[1]. In the US, Sanyal *et al.*[36] found that, among patients with NAFLD-related HCC, as many as 54% were not known to be cirrhotic based on health care coding information. The proportion of non-cirrhotic patients was similarly high in the smaller Cleveland clinic cohort, which found that 43.4% of cases occurred in the absence of cirrhosis[48]. Those patients tended to be older (67.5 ± 12.3 years *vs.* 62.7 ± 8.1 years), and less likely to be obese (52% *vs.* 83%) or have type 2 diabetes (38% *vs.* 83%), than their cirrhotic counterparts. Similar findings have also been reported from European cohort studies, with a high proportion of NAFLD-related HCC occurring in non-cirrhotic patients: 46.2% in a multicentre Italian study, 41.7% in a single centre German study and 22.7% in a UK-based cohort[34,49,50]. In the cohort study by Kanwal *et al.*[32], the risk of HCC in non-cirrhotic NAFLD was higher than in the healthy controls, but the overall incidence rates were low (0.08 *vs.* 0.02 per 1000 person years). Despite this, due to the high global prevalence of NAFLD, the absolute burden of NAFLD-related HCC, including those with and without cirrhosis, remains significant.

Clinical outcomes in NAFLD-related HCC

Patients with NAFLD-related HCC tend to present at an older age compared to patients with HCC due to other aetiologies[8,28,31]. In the SEER cohort, NAFLD-related HCC patients were older (73 ± 8 years *vs.* 66 ± 11 years), had more heart disease (35.1% *vs.* 7%-27%) and had a shorter mean survival time (14.17 ± 17.14 months *vs.* 17.85 ± 21.47 months), compared to other aetiologies; and 84.3% died of their primary liver cancer[8]. This group found higher odds of one-year mortality (OR: 1.21, 95%CI: 1.01-1.45) in those with NAFLD-related HCC, although other large HCC cohorts have found no difference in overall survival[34,37,49]. Furthermore, a Canadian study of 929 patients who underwent a transplant for HCC, including 60 with NAFLD-related HCC, also found that these patients had similar one-, three- and five-year survival outcomes (98%, 96% and 80%, respectively) compared to non-NAFLD HCC (95%, 84% and 78%, respectively), with no difference in tumour recurrence (13.3% *vs.* 14%)[51]. As patients with NAFLD-related
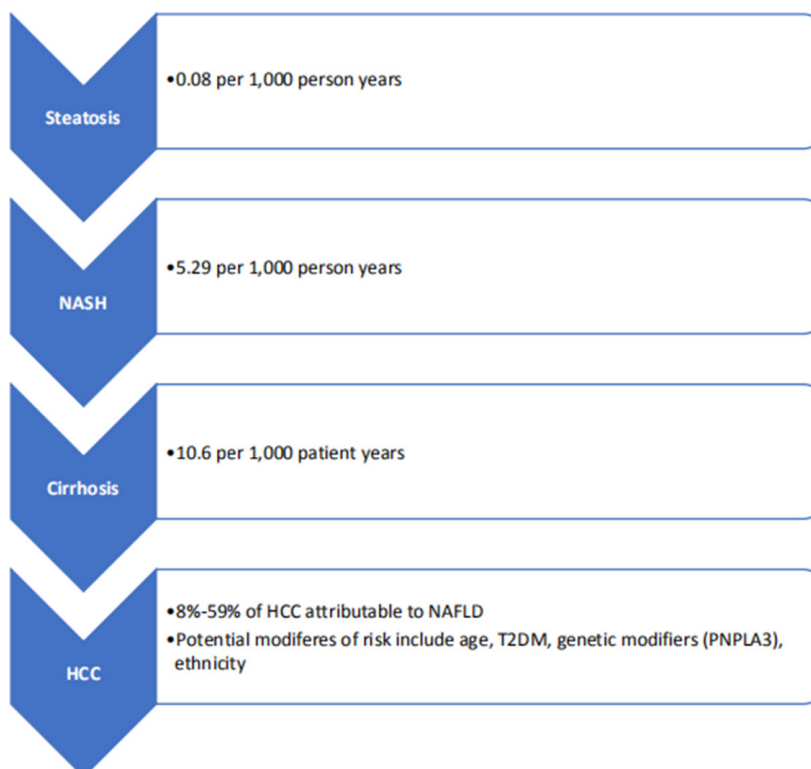
Figure 1. Increasing risk of HCC with stage of disease[1,7,32,36-38]. NASH: nonalcoholic steatohepatitis; HCC: hepatocellular carcinoma; NAFLD: non-alcoholic fatty liver disease; T2DM: type 2 diabetes mellitus

HCC tend to be older, with greater metabolic and vascular comorbidities than those presenting with HCC from other causes, the relative equivalence in survival may be due to the proportion of patients without cirrhosis in some of these cohorts (22.8% and 41.7% in the studies by Dyson and Mittal, respectively)[8,34,37].

Although non-cirrhotic and cirrhotic NAFLD-related HCC may present differently, overall survival rates between these two groups appear to be the same. A review of NAFLD-related HCC cases at the Cleveland clinic found that HCC in non-cirrhotic patients was more likely to present as a single nodule (80.6% *vs.* 52.2%) and with a larger nodule size (> 5 cm) (77.8% *vs.* 10.6%)[48]. From a treatment perspective, they were more likely to receive hepatic resection (66.7% *vs.* 17%) but less likely to receive loco-regional therapy (22.3% *vs.* 61.7%) or orthotopic liver transplantation (OLT) (0% *vs.* 72.3%). They also found that HCC in non-cirrhotic NAFLD had a higher rate of recurrence than in those with cirrhosis (86% *vs.* 14%); however, after adjusting for age or OLT, there was no difference in overall survival between the two groups. This is consistent with data from a large European cohort of NAFLD-related HCC patients in Sweden, which found non-cirrhotic patients to be older (median 74 years *vs.* 70 years), with larger tumours[52]. Similarly, in this group, those without cirrhosis were more likely to have a resection (35% *vs.* 8%), and less frequently underwent OLT (0% *vs.* 11%), but no significant difference was found in the overall mortality rates [adjusted hazard ratio (aHR): 0.93, 95%CI: 0.58-1.51, *P* = 0.78]. On further analysis, parameters independently associated with a higher risk for overall mortality in non-cirrhotic patients included; Barcelona Clinic Liver Cancer class C or D (aHR compared with class 0:7.03, 95%CI: 2.89-17.11, *P* < 0.001), the presence of four or more tumours (aHR compared with one tumour: 9.26, 95%CI: 3.20-26.81, *P* < 0.001), higher serum albumin (aHR 0.29 per increased g/dL, 95%CI: 0.15-0.56, *P* < 0.001) and the presence of T2DM (aHR: 3.65, 95%CI: 1.69-7.89, *P* = 0.001).

Obesity and type 2 diabetes and HCC risk

Obesity and T2DM are metabolic comorbidities closely associated with NAFLD, which have also been independently linked to the development of HCC[53,54]. T2DM has been found to be associated with increased HCC risk, particularly in patients with non-viral liver disease. A 2012 meta-analysis including 17 case-control studies and 32 cohort studies showed a statistically significant increased risk of HCC prevalence among diabetic individuals (RR: 2.31, 95%CI: 1.87-2.84)[53]. The pooled risk estimate from 17 case-control studies (OR: 2.40, 95%CI: 1.85-3.11) was slightly higher than that from the 25 cohort studies included (RR: 2.23, 95%CI: 1.68-2.96). This analysis was limited by lower patient numbers, which prevented adequate subgroup analysis, and, while there was some adjustment for the presence of viral hepatitis and cirrhosis, the possible presence of underlying NAFLD was not accounted for. In a study from the Mayo Clinic, the hazard ratio of HCC in cirrhotic patients without HCV infection was found to be two times higher in patients with T2DM compared to those without [Hazard Ratio (HR): 2.1, 95%CI: 1.1-4.1][55]. Additionally, T2DM has also been found to be an independent risk factor for HCC in patients with cryptogenic cirrhosis; however, these patients also demonstrated features of NAFLD based on markers of insulin resistance and lipid profiles[56]. Overall, while T2DM does appear to be associated with an increased risk of HCC in those with or without known liver disease, current studies are limited by confounding and the difficulty of excluding underlying NAFLD as a contributing factor.

Obesity is associated with an increased risk of many types of malignancy, and several epidemiological studies suggest a modest increase in the relative risk of HCC in obese populations[57]. A 2012 meta-analysis of 26 prospective cohort studies, including 25,337 individuals, found excess body weight [body mass index (BMI) $\geq$ 25 kg/m^2] and obesity (BMI $\geq$ 30 kg/m^2) to be associated with an increased risk of primary liver cancer [Summary Relative Risks (SRR) 1.48, 95%CI: 1.31-1.67; and SRR 1.83, 95%CI: 1.59-2.11, respectively][54]. While this evidence is somewhat limited by heterogeneity, on subgroup analysis, there was an increased risk in males, those with HCV and cirrhosis from any aetiology compared with the general population, suggesting that obesity may be an important cofactor in the development of HCC.

Genetic markers in NAFLD-related HCC

Several host genetic polymorphisms have been linked to the presence of NAFLD, risk of fibrosis progression and the development of NAFLD-related HCC[58]. These are particularly relevant as they may present novel biomarkers to help triage those at most risk of HCC and therefore in need of screening, particularly among those with NAFLD and no cirrhosis. The most studied of these is a single-nucleotide polymorphism (SNP) in PNPLA3. The association between PNPLA3 (rs738409) and hepatic steatosis was first identified in the genome-wide survey performed on the > 2000 participants of the Dallas Heart Study who underwent Magnetic Resonance Spectroscopy of the liver for quantification of liver fat[59]. The presence of this SNP has since been found to be associated with an increased risk of advanced fibrosis in all patients with liver disease, and a 2014 meta-analysis confirmed that it was also associated with an increased risk of HCC in patients with NASH and alcohol-related cirrhosis (OR: 1.67, 95%CI: 1.27-2.21), but not in other aetiologies (OR: 1.33, 95%CI: 0.96-1.82)[60]. Moreover, in a large well-characterised Northern European cohort of patients with histologically proven NAFLD patients, 100 of whom had NAFLD-related HCC, it was shown that PNPLA3 rs738409, especially the C > G polymorphism, was associated with an increased risk of HCC independent of other traditional risk factors including age, gender, BMI, presence of T2DM or the presence of advanced fibrosis or cirrhosis[61]. Despite this evidence, the potential use of this SNP as a predictive biomarker to identify high-risk individuals for HCC screening has not yet been validated in larger cohorts, and it is not widely used.

Other potential genetic risk modifiers of interest include a variant in the *MBOAT7* gene, which was associated with an increased risk of NAFLD-related HCC in an Italian cohort of NAFLD patients (OR: 1.65, 95%CI: 1.08-2.55; *n* = 765), particularly in those without advanced fibrosis (*P* < 0.001), and TERT

promoter mutations that can be found in low and high grade dysplastic lesions, and in increased frequency in early HCC[62-64]. The *TM6SF2* genetic variant on chromosome 19 has also been shown to be associated with an increased risk of NAFLD and NAFLD-related hepatic fibrosis and cirrhosis, independent of other known traditional risk factors including PNPLA3 status[65]. In one cohort of 99 biopsy-confirmed HCC patients, homozygous carriage of the *TM6SF2* rs58542926 minor allele was associated with an increased risk of NAFLD-related HCC. However, this was not shown to be significant on multivariate analysis when adjusting for traditional risk factors such as age, gender or cirrhosis[66]. Further investigation is required to determine the clinical significance of these associations and establish whether they may be of prognostic or diagnostic significance in the future. Consideration of cost, access to and acceptability of genetic sequencing as a screening or prognostic tool for patients with NAFLD must also be explored.

Should patients with NAFLD undergo HCC surveillance?

Routine HCC surveillance is recommended for all patients with cirrhosis regardless of the aetiology of their liver disease[9,10], and as such all patients with NAFLD cirrhosis should be enrolled in a screening program. While HCCs that are detected by surveillance programs are found at an earlier stage than those found incidentally, patients with NAFLD-related HCC are less likely to have recognised liver disease, and as such have a lower proportion of HCCs detected through screening compared with other causes of liver disease[67]. In the UK Newcastle cohort and the HCC NAFLD Italian groups, only 22.8%-47.6% of NAFLD-related HCC cases were detected during surveillance compared with 46.2%-63.3% in HCV[34,49]. Additionally, even in high risk patients who have NAFLD cirrhosis, screening is less likely to detect early stage HCCs compared with other aetiologies[34,37,49]. This is likely attributable to the current recommended screening modality of ultrasound, which can be technically challenging in obese patients with excess visceral adiposity, and is highly operator dependant[68]. Cross-sectional imaging such as MRI may be used as an alternative in patients in whom adequate views of the liver are unable to be obtained via ultrasound, however the suitability and cost-effectiveness of this approach has yet to be determined. Concern remains regarding whether NAFLD patients without cirrhosis should also be screened, considering that 23%-54% of NAFLD-related HCC occurs in non-cirrhotic patients. However, due to the large volume of NAFLD patients and low annual HCC incidence of less than 2% in this cohort, routine screening is not currently recommended[34,36,37,49]. The development of risk stratification tools may be of use in identifying high-risk patients who would benefit from screening, and may include the use of genetic markers.

Limitations in current estimates and Future directions

While these observational studies are informative in reviewing trends, there are several limitations to this method of determining the true burden of disease. The largest cohorts have typically identified NAFLD using coded registry data, or blood tests, which can be inconsistent and will not always reflect whether there is underlying fibrosis or steatohepatitis. Additionally, HCC registry information tends to be more representative of tertiary care and patients who have been referred for active management. As NAFLD-related HCC patients tend to be older and more comorbid, it is possible that not all cases are referred to these specialist centres, and they may have remained in community care settings. Similarly, as the SEER cohort demonstrated, patients with NAFLD-related HCC are less likely to be transplanted compared to HCC due to other causes, thus transplant registry information is also likely to underestimate the true burden of disease. Another limitation of cross-sectional retrospective studies is the evolving nature of NAFLD and NASH. These are not static disease states, but can progress or regress over time with changes in weight, which is often difficult to assess. While these studies have examined the risk associated with NAFLD alone, the increasing prevalence of obesity and diabetes in HCC of all aetiologies suggests that these metabolic factors including NAFLD could be an important co-factor in hepatic carcinogenesis[34]. In the future, the development of a simple, cost-effective population-based diagnostic test would be of benefit to improve the detection rate of NAFLD and trigger screening for underlying fibrosis and cirrhosis. Additionally, the development of prognostic markers or algorithms to identify high-risk patients for HCC surveillance is also an area of ongoing research. Further, as treatment for NAFLD progresses, and weight

management becomes a greater focus of therapy, the ongoing risk of HCC in individuals with NAFLD who have subsequently lost weight or undergone therapy will need to be evaluated.

CONCLUSION

NAFLD is an important cause of liver disease worldwide and is associated with an increased risk of developing HCC, particularly in the presence of liver cirrhosis. While the absolute risk appears to be low, prevalence of NAFLD-related HCC is increasing in parallel with the prevalence of NAFLD worldwide, particularly in the West. The future global burden of NAFLD-related HCC represents a major public health threat and further research to identify cost-effective prevention and treatment strategies are urgently required.

DECLARATIONS

Authors' contributions
Performed the literature review and generated the manuscript: Farrell A
Assisted in developing the outline for the manuscript: Howell J
Responsible for review of the manuscript: Ryan M, Howell J

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