Effect of metabolic dysfunction-associated fatty liver disease on the risk of hepatocellular carcinoma in patients with chronic hepatitis B: A systematic review and meta-analysis

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Abstract. Hepatocellular carcinoma (HCC) remains an important complication in patients with chronic hepatitis B (CHB). An association between the presence of metabolic dysfunction-associated fatty liver disease (MAFLD) and an increased HCC risk in patients with CHB may exist; however, the exact nature of this possible association remains unclear. The present study conducted a comprehensive meta-analysis by pooling data from 18 studies encompassing 23,927 participants. The odds ratios (ORs) were calculated using a random-effects inverse-variance model, and heterogeneity was assessed using Cochran's Q test and the I^2 statistic. In addition, subgroup analyses were performed on the basis of geographical region, study design and follow-up length. Publication bias and meta-regression were also assessed. The overall pooled OR for the association between MAFLD and HCC risk in patients with CHB was 1.053 (95% CI, 0.704-1.576), which suggested a lack of association. Heterogeneity was observed across studies. Subgroup analyses demonstrated a potentially protective effect for MAFLD on the risk of HCC in patients in Asian countries (OR, 0.783; 95% CI, 0.568-1.080) and the opposite effect in other regions (OR, 4.380; 95% CI, 2.440-7.864). Analysis of the prospective cohort studies suggested a significant protective effect for MAFLD (OR, 0.479; 95% CI, 0.365-0.629), while analysis of retrospective cohorts did not. The publication bias assessment was inconclusive and the meta-regression failed to identify heterogeneity sources. The association between MAFLD and HCC risk in patients with CHB appeared to be multifactorial and may vary on the basis of geographical region and study design. While the exact mechanisms remain elusive, the potential protective effect demonstrated in certain subgroups warrants further investigation.

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Introduction

Hepatocellular carcinoma (HCC) is a common primary malignancy of the liver and represents a major public health concern worldwide (1). Approximately 830,000 individuals succumb to HCC each year, making it the third leading cause of cancer-associated mortality globally. This is due to an insidious nature and late clinical presentation, which lead to a poor prognosis at the time of diagnosis (2). Identifying and addressing the underlying etiological factors of HCC is essential to promote early detection and develop effective preventive strategies.

Chronic hepatitis B (CHB) infection has been recognized as a primary driver in the progression to HCC (3). However, despite extensive vaccination campaigns and antiviral therapies, the global CHB burden remains high, with an estimated 296 million individuals affected worldwide as per the World Health Organization (4). Amongst individuals with CHB, the lifetime risk of developing HCC can be as high as 15-25% (5). The molecular and cellular pathophysiological mechanisms underlying this transition involve an interplay between viral replication, chronic inflammation and repeated hepatic injury, all of which can contribute to malignant transformation (6,7).

Metabolic dysfunction-associated fatty liver disease (MAFLD) has been previously studied in the context of liver pathologies. Formerly known as non-alcoholic fatty liver disease (NAFLD), MAFLD encompasses a spectrum of liver abnormalities ranging from simple steatosis to non-alcoholic steatohepatitis and it can progress to cirrhosis and even HCC (8). The prevalence of MAFLD has increased along with the global rise in obesity and type 2 diabetes, heralding an impending epidemic of MAFLD-associated complications, including HCC (9).

Consequently, the coexistence of MAFLD and CHB in a patient presents a complex clinical scenario (10). Preliminary evidence suggests that this convergence may have a multiplicative effect on the HCC risk (11). The metabolic derangements (insulin resistance and dyslipidemia) and inflammatory milieu of MAFLD (cytokine imbalance and oxidative stress) could

exacerbate the hepatocarcinogenic potential of CHB (12). However, the precise nature and magnitude of the effects of combining these conditions remains sparsely documented and unclear. Thus, evaluating the cumulative HCC risk that MAFLD may impart on patients with CHB is important. A clear understanding would help to elucidate the clinical prognosis of these patients and would facilitate stringent surveillance, early interventions and tailored management plans.

Systematic reviews and meta-analyses are powerful evidence synthesis tools, especially when the existing literature provides conflicting or inconclusive results (13). A rigorous, methodical consolidation of the available evidence may help clarify the scarce and heterogeneous data on the combined roles of MAFLD and CHB in HCC pathogenesis.

In the present systematic review and meta-analysis, the available literature on the topic was comprehensively evaluated and the risk of HCC in patients with CHB with concomitant MAFLD was assessed. The findings of the present study may potentially reduce the knowledge gap and pave the way for future focused research, refined clinical guidelines and targeted public health measures in this emergent field of hepatology.

Materials and methods

Study guidelines and registration. The methodology for the present systematic review and meta-analysis was planned according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses 2020 guidelines (13). The meta-analysis was registered at PROSPERO (registration no. CRD42023453979).

Eligibility criteria. The eligibility criteria for the present study were as follows: i) Population: Studies involving patients diagnosed with CHB were included and no restrictions were applied regarding age, sex, geographic location or ethnicity; ii) exposure and comparison: Studies on patients with CHB with or without MAFLD were included; iii) outcomes: The principal outcome of interest was the incidence of HCC; and iv) study design: All types of study designs that were published in English from the inception of the databases until July 2023 were included. To minimize publication bias, both published literature and grey literature were included in the literature search.

Information sources. Strategic searches were conducted across electronic databases including PubMed (https://pubmed.ncbi.nlm.nih.gov), Embase (https://www.embase.com/), Cochrane Central Register of Controlled Trials (https://www.cochranelibrary.com/central) and Cumulative Index to Nursing & Allied Health Literature (https://www.ebsco.com/products/research-databases/cinahl-database). In addition, manual searches were performed within references of pinpointed studies and pertinent reviews. To ensure exhaustive and comprehensive information retrieval, the authors of primary studies were contacted as needed to gather unpublished data or clarify study specifics. String searches were formulated using the following terms: 'Metabolic-associated fatty liver disease', 'MAFLD', 'non-alcoholic fatty liver disease', 'NAFLD', 'hepatic steatosis', 'viral hepatitis', 'chronic hepatitis B', 'hepatocellular carcinoma' and 'HCC' and both Medical Subject Headings and associated keywords were used. Appendix S1 delineates the detailed search algorithm used.

Study records

Data management. EndNote X9 (Clarivate) citation management software was used to systematically retrieve and manage studies. Duplicate entries were identified and excluded and the remaining articles were subjected to eligibility screening.

Selection process. A total of two independent individuals screened the titles and abstracts of the retrieved studies and then performed full-text evaluations to ensure relevance and fit for inclusion into the present study. Discrepancies between reviewers were reconciled through dialogue.

Data collection process. Data were extracted from the selected studies using a standardized extraction template by two reviewers. The harvested data included study attributes (authors, year of publication, design and setting), participant specifics (count, age, sex and MAFLD and CHB status), risk factor details and outcomes.

Risk of bias in individual studies. The risk of bias in observational studies was calculated using Newcastle Ottawa scale (NOS) (14). A score of ≥7 on NOS was classed as indicative of a high-quality study. A total of two individuals undertook the evaluations settling any disagreements via discussions.

Statistical analysis. STATA software (version 17; StataCorp LP) was used to consolidate the meta-analysis data. A random-effects model with the inverse variance technique was used to account for potential study variability. Heterogeneity variance was estimated using the DerSimonian-Laird method (15). Effect measures encompassed pooled hazard ratios (HRs; for studies reporting the estimates as HRs) and odds ratios (ORs; for dichotomous outcomes) (15). Forest plots were produced to visualize findings with 95% CIs. Subgroup analyses were conducted on geographical regions, study designs and follow-up lengths. Heterogeneity was assessed using the I^2 and τ^2 statistics and χ^2 tests (15). Funnel plot and Egger's regression test were used to detect publication bias. Sensitivity analysis was performed by excluding the included studies one-by-one and checking for the single study effects and the consistent nature of the effect size. The quality of evidence for every outcome was assessed by applying the Grading of Recommendations Assessment, Development and Evaluation (GRADE) approach, which considered bias risk, result consistency, evidence directness, estimate precision and publication bias susceptibility (16). $P<0.05$ was considered to indicate a statistically significant difference.

Results

Search results. Through primary screening, a total of 1,903 citations were identified across the databases. Following the removal of duplicates, 275 full-text articles were retrieved. After a secondary screening, 18 studies were included that fully satisfied the eligibility criteria (Fig. 1) (11,17-33).

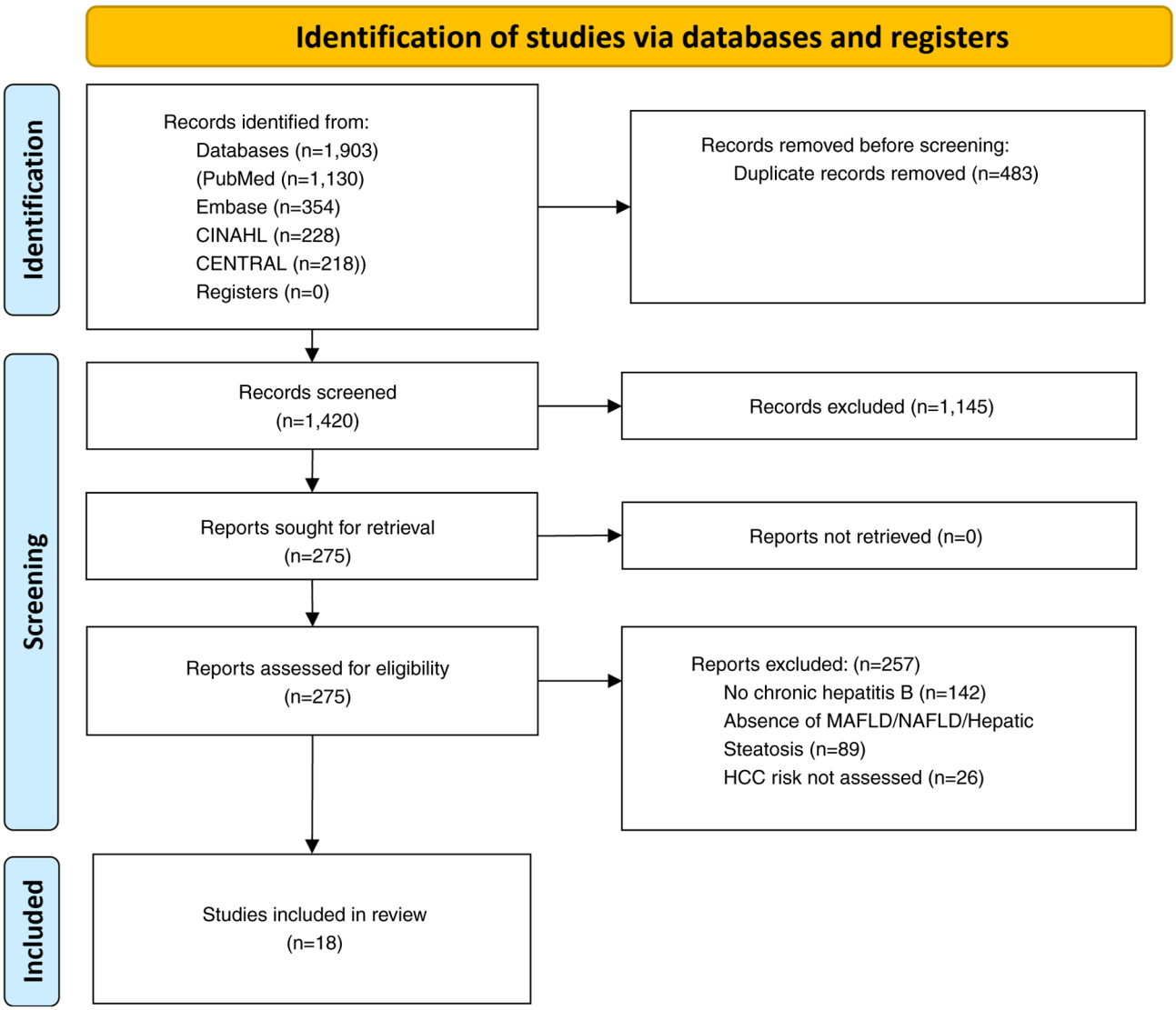

Figure 1. Search strategy for the present study. CINAHL, Cumulative Index to Nursing & Allied Health Literature; CENTRAL, Cochrane Central Register of Controlled Trials; MAFLD, metabolic dysfunction-associated fatty liver disease; NAFLD, non-alcoholic fatty liver disease; HCC, hepatocellular carcinoma.

Characteristics of the included studies. For the present meta-analysis, data were obtained from a diverse range of studies from across the globe (Hong Kong, Korea, Canada, Taiwan, Singapore, China, Israel and Thailand). Most studies had retrospective cohort designs, but three studies were based on prospective cohorts (22,29,31) and one on a nested case-control approach (25). The sample sizes varied from 270-63,273 participants. The follow-up periods lasted from 3.0-28.1 years. The participants' profiles also were varied, with some studies focusing on male participants only and others including participants of both sexes. A mixed risk of bias was found across studies, with 11 studies were designated as having a 'high' risk of bias (Table I).

Association between MAFLD and HCC in patients with CHB. Data from 18 studies comprising 23,927 participants were included for the analysis of the number of events and participants. The pooled OR for the association between the presence of MAFLD and an increased risk of HCC in patients with CHB was 1.053 (95% CI, 0.704-1.576), with no statistical significance obtained from the test of overall effects (z=0.252; P=0.801; Fig. 2). A high degree of heterogeneity was found among the included studies, with a Cochran's Q value of 71.78 [degrees of freedom (df)=12; P<0.001]. The I^2 test result was 83.3% (95% CI, 37.5-92.4), which indicated that a substantial proportion of the total variation in effect estimates was due to between-study heterogeneity. The estimated heterogeneity variance using the DerSimonian-Laird method was 0.4076.

A random-effects inverse-variance model with the DerSimonian-Laird estimate of τ^2 was used to pool the HRs from individual studies. The HRs for the effect of MAFLD on the risk of HCC in patients with CHB from the included studies ranged from 0.420-7.270. The summary HR derived from the overall pooled data suggested that MAFLD was associated with a 1.253-fold increased risk of HCC in patients with CHB. However, this association was not statistically significant (95% CI, 0.895-1.754; z=1.313; P=0.189). In addition, significant heterogeneity was demonstrated among the included studies, as evidenced by a Cochran's Q value of 81.52 (P<0.001) and an I^2 statistic of 85.3%, which indicated substantial variations in

Table I. Characteristics of the 18 included studies.

First author, year	Geographical location	Study design	Sample size, n	Follow-up duration, months	Study participant description	Mean patient age, years	Male to female ratio, %	Risk of bias	(Refs.)
Chan *et al*, 2017	Hong Kong	Retrospective cohort	270	79.9	Patients with consecutive HBV infection undergoing liver biopsy between January 2006 and December 2009	43.6	75.2:24.8	High	(18)
Chang *et al*, 2021	Korea	Retrospective cohort	720	36.0	Treatment-naïve patients >18 years old with virologically (HBV DNA <2,000 IU/ml) and biochemically (alanine aminotransferase level <40 IU/l) quiescent chronic hepatitis B who underwent TE	52	58.2:41.8	High	(26)
Cho *et al*, 2020	Korea	Retrospective cohort	826	43.1	Patients whose serum HBV DNA levels were continuously suppressed <2,000 IU/ml by treatment	53.5	61.0:39.0	High	(30)
Choi *et al*, 2020	Canada	Retrospective cohort	1,089	120.0	Patients with CHB from electronic medical records who underwent a liver biopsy	38	65.9:34.1	Low	(27)
Hsueh *et al*, 2022	Taiwan	Prospective cohort	2,385	337.0	Male, HBsAg-positive civil servants aged ≥30 years at baseline who were recruited from the Government Employees Central Clinics during routine free physical examination	43.2	100.0:0.0	Low	(22)
Huang *et al*, 2023	Taiwan	Retrospective cohort	10,546	61.0	Patients aged ≥20 years with CHB	52	51.0:49.0	High	(21)
Kim *et al*, 2019	Korea	Retrospective cohort	334	60.0	Patients with treatment-naïve CHB with available TE results who were initiated on entecavir or tenofovir	51	62.9:37.1	High	(23)
Kim *et al*, 2023	Korea	Retrospective cohort	63,273	104.0	Patients with CHB	NR	NR	High	(20)
Lee *et al*, 2019	Korea	Retrospective cohort	321	63.0	Patients with consecutive CHB who underwent liver biopsy	41	61.1:38.9	High	(17)

Table I. Continued.

First author, year	Geographical location	Study design	Sample size, n	Follow-up duration, months	Study participant description	Mean patient age, years	Male to female ratio, %	Risk of bias	(Refs.)
Li *et al*, 2021	Taiwan	Retrospective cohort	2,158	132.0	Patients aged ≥18 years with CHB verified by individual chart review who were Asian	49.6	70.9:29.1	Low	(33)
Lim *et al*, 2020	Singapore	Retrospective cohort	289	111.1	Patients with CHB who underwent liver biopsy	46.4	72.3:27.7	High	(28)
Mak *et al*, 2021	China	Prospective cohort	2,403	46.4	Patients with CHB, defined as persistent seropositivity for HBsAg for ≥6 months, aged ≥18 years, who were treatment-naïve or not currently receiving treatment and were consecutively recruited for TE assessment	55.6	55.6:44.4	Low	(31)
Oh *et al*, 2021	Korea	Retrospective cohort	1,823	60.0	Adults aged ≥40 years with CHB who underwent Fibroscan evaluations	56	67.0:33.0	High	(19)
Peleg *et al*, 2019	Israel	Retrospective cohort	524	72.0	Patients with treatment-naïve CHB	50.5	60.1:38.9	High	(24)
Rugivarodom *et al*, 2023	Thailand	Prospective cohort	408	164.0	Patients with consecutive chronic HBV infection who underwent a liver biopsy to determine the need for antiviral treatment	44	65.4:34.6	Low	(29)
van Kleef *et al*, 2021	Canada	Retrospective cohort	,076	116.0	Patients with CHB who underwent 1liver biopsy	38.6	65.7:34.3	High	(11)
Wang *et al*, 2023	China	Retrospective cohort	332	47.0	Patients with hepatitis B-related cirrhosis	50.4	74.9:25.1	Low	(32)
Yu *et al*, 2022	Taiwan	Nested case control study	1,453	231.0	Male, HBsAg-positive civil servants aged >30 years	49.2	100.0:0.0	Low	(25)

NR, not reported; HBV, hepatitis B virus; CHB, chronic hepatitis B; TE, transient elastography; HBsAg, hepatitis B surface antigen.

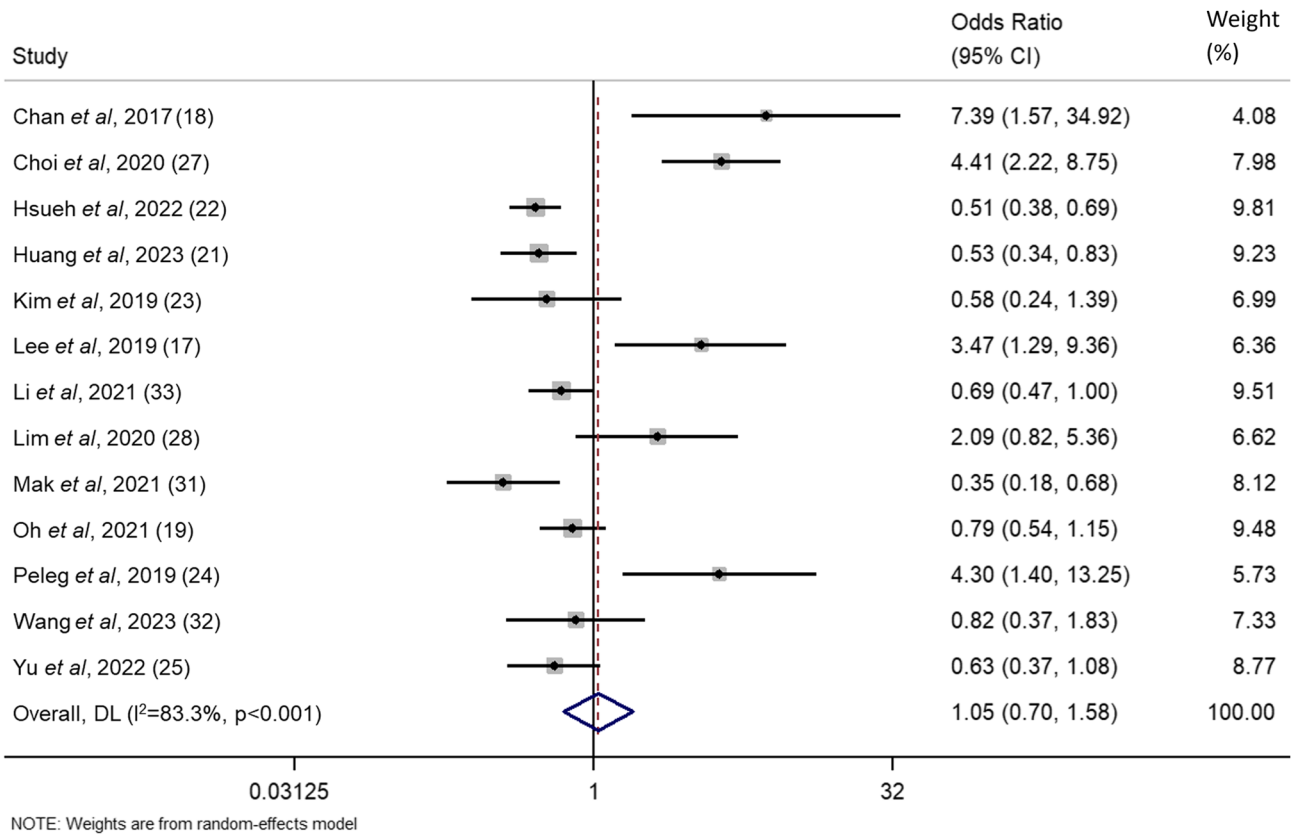

Figure 2. Forest plot showing the association between metabolic dysfunction-associated fatty liver disease and hepatocellular carcinoma using number of events and participants (n=13 studies). DL, DerSimonian and Laird approach.

study outcomes. The modified H^2 value was 5.793 and τ^2 was 0.2550 (Fig. 3).

Subgroup analyses. The association between MAFLD and the risk of HCC in patients with CHB was assessed based on the geographical location of the study (Fig. S1). Data from 11 of the studies included were from Asian countries (17-19,21-23,25,28,31-33). The pooled OR for the aforementioned studies was 0.783 (95% CI, 0.568-1.080), which accounted for 86.29% of the overall weight. The Cochran's Q value was 34.27 (df=10; P<0.001) with an I^2 of 70.8%, which indicated moderate heterogeneity. Data were also analysed from two studies from other geographical regions (Canada and Israel) (24,27). The pooled OR of the aforementioned studies was 4.380 (95% CI, 2.440-7.864), which represented 13.71% of the overall weight. No evidence of heterogeneity was found with a Cochran's Q value of 0.00 (df=1; P=0.971) and an I^2 of 0.0%. The test for the subgroup effect size demonstrated no significant difference for Asian countries (z=-1.490; P=0.136), while the subgroup effect of the other geographical location analysis was statistically significant (z=4.948; P<0.001). The heterogeneity between the subgroups was also statistically significant with a Q value of 25.54 (df=1; P<0.001).

Based on the study design, two prospective cohort studies were identified (Fig. S2) (22,31). The pooled OR of the aforementioned analysis was 0.479 (95% CI, 0.365-0.629), which contributed to 17.93% of the total weight. Heterogeneity measurement demonstrated no evidence of variation, with a Cochran's Q value of 1.00 (df=1; P=0.318) and an I^2 of 0.0%. A total of 11 retrospective cohort studies were identified (17-19,21,23-25,27,28,32,33). The combined OR of the aforementioned studies was 1.294 (95% CI, 0.813-2.059), which accounted for 82.07% of the total weight. Significant heterogeneity between these studies was demonstrated, as indicated by a Cochran's Q value of 55.95 (df=10, P<0.001) and an I^2 of 82.1%. The tests for subgroup effect sizes demonstrated a significant difference for the prospective cohort studies (z=-5.303; P<0.001), but not for the retrospective studies subgroup (z=1.086; P=0.277). Significant heterogeneity between the subgroups was also observed, with a Q value of 13.09 (df=1; P<0.001).

Data from nine studies were used for follow-up length (<10 years) subgroup analyses (Fig. S3) (17-19,21,23,24,28, 31,32). The combined OR was 1.147 (95% CI, 0.670-1.965), which contributed to 63.94% of the total weight. Significant heterogeneity among these studies was found with a Cochran's Q value of 38.58 (df=8; P<0.001) and an I^2 of 79.3%. A total of four studies had a follow-up period of ≥10 years (22,25,27,33). The pooled OR of the aforementioned studies was 0.944 (95% CI, 0.459-1.939), which accounted for 36.06% of the overall weight. These studies demonstrated substantial heterogeneity with a Cochran's Q value of 32.19 (df=3; P<0.001) and an I^2 of 90.7%. The tests for subgroup effect sizes did not reveal a statistically significant result for either follow-up group (<10 years, z=0.500; P=0.617; ≥10 years, z=-0.158; P=0.874). The heterogeneity between the two subgroups was not significant (Q=0.18; df=1; P=0.670).

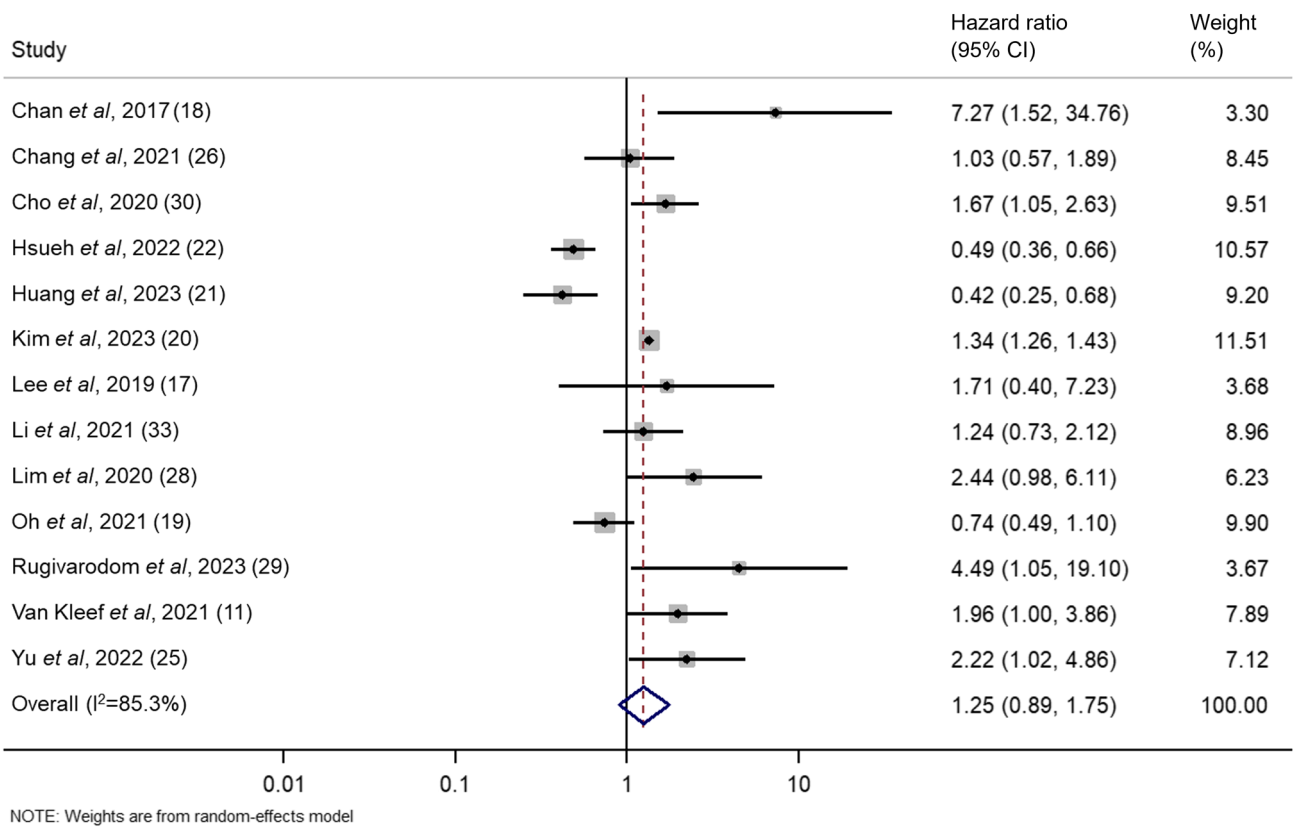

Figure 3. Forest plot showing the association between metabolic dysfunction-associated fatty liver disease and hepatocellular carcinoma using hazard ratios with CIs (n=13 studies).

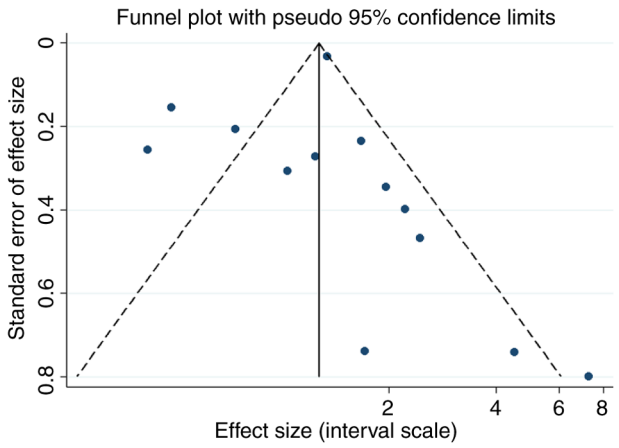

Figure 4. Funnel plot of publication bias assessment.

Publication bias assessment. Egger's regression asymmetry test demonstrated evidence of publication bias. The slope coefficient was 0.2585 (95% CI, 0.0396-0.4774; t=2.60; P=0.025). However, the intercept (bias) term was-0.3096 and was not statistically significant (95% CI, -2.3297-1.7105; t=-0.34; P=0.742), which suggested that the funnel plot asymmetry may be due to factors other than publication bias (Fig. 4).

Meta-regression analysis results. To identify potential sources of heterogeneity and evaluate the potential influence of study-level characteristics on the reported effect sizes, meta-regression analyses were conducted. The following covariates were considered: Mean age, follow-up duration, study design and geographical location of the study.

Mean age. The regression coefficient suggested that for every 1-year increase in the mean age, the effect size decreased by 0.0514; however, this association was not statistically significant (coefficient=-0.0514; P=0.253). A between-study variance (τ^2) of 0.4398 was demonstrated, which suggested that ~83.58% of the total variation in effect sizes was due to heterogeneity.

Follow-up duration. Similar effect sizes were found in the follow-up groups [coefficient for follow-up group 2 (≥10 years)=-0.0303; P=0.949]. The τ^2 value was 0.4117 with 82.20% of the total variation in effect sizes attributed to heterogeneity.

Study design. Similar effect sizes were demonstrated in the two study design groups [coefficient for design group 2 (≥10 years)=0.3617; P=0.557]. The between-study variance (τ^2) was 0.3627 and ~77.05% of the total variation in effect sizes resulted from heterogeneity.

Geographical location of study. The effect sizes were similar among the country groups [coefficient for country group 2 (countries outside Asia)=0.4470; P=0.574]. The τ^2 value was 0.3816 with 86.24% of the total variation in effect sizes attributed to heterogeneity.

Sensitivity analysis. Sensitivity analysis was performed to check the robustness of the estimates (Fig. S4). The findings of the present study were not unduly influenced by any single

study and the results remained consistent across the analysis, as there was no change in direction or magnitude of the overall pooled estimate after removal of any single study, which affirmed the reliability of the overall conclusions of the present study.

GRADE evidence. The quality of evidence was initially graded as low, because the review included observational studies. However, the presence of studies with high risk of bias, imprecision and non-significant associations between MAFLD and HCC risk caused a further downgrading in the quality of evidence rating to very low-quality.

Discussion

The concomitant presence of MAFLD and HCC in patients with CHB has emerged as an area of notable clinical interest. The present comprehensive meta-analysis, which included data from 18 studies and 23,927 participants, aimed to explore a possible association between MAFLD and HCC with depth and rigor. The findings of the present study generated further questions and underscored the complexity of the topic.

The principal finding of the present meta-analysis was the non-significant association between MAFLD and the HCC risk in patients with CHB, with a pooled OR of 1.053. This finding diverges from several previous primary investigations which have proposed MAFLD as a significant risk factor for HCC (17,18,24,27). The wide CI value suggested that MAFLD may confer a modest risk, but it could also be protective against HCC. Thus, clinicians and researchers need to be cautious in their interpretation of the findings of the present study, considering the study's design and the populations analysed.

Further analysis of the potential mechanisms linking MAFLD to HCC in patients with CHB should clarify this issue and potentially reveal the processes involved in this association. Chronic inflammation is central to the progression of MAFLD (34). Hepatic steatosis, a hallmark of MAFLD, can activate Kupffer cells, the resident macrophages of the liver, which leads to the secretion of pro-inflammatory cytokines, such as TNF-α and IL-6 (35). Such inflammatory markers can promote hepatocarcinogenesis, especially if the liver is already compromised by CHB infection (36). Insulin resistance is a key feature of metabolic syndrome and MAFLD. Elevated insulin levels and a consequentially increased insulin-like growth factor can activate cellular pathways that stimulate hepatocyte proliferation and inhibit apoptosis, which fosters an environment conducive to neoplastic transformation (36).

Fatty acid accumulation in hepatocytes can cause mitochondrial dysfunction, which leads to elevated reactive oxygen species levels. Oxidative stress damages DNA and can initiate and promote carcinogenesis. In patients with CHB, the added viral-induced cellular stress may synergize with the oxidative stress from MAFLD and amplify the risk for malignant transformation (37). Adipose tissues, especially in the context of obesity and MAFLD, actively secrete adipokines, such as leptin and adiponectin. Leptin, which is increased in individuals with obesity, promotes cell proliferation and reduces apoptosis, whilst adiponectin serves an anti-inflammatory role. An imbalance in these adipokine contents, as observed in MAFLD, can alter hepatic homeostasis and promote oncogenesis (38). The role of the gut microbiota in liver diseases is being investigated, as dysbiosis, a disruption in the gut microbial equilibrium observed in MAFLD, can lead to increased gut permeability, which allows bacterial endotoxins to enter the liver via the portal circulation. These endotoxins can activate hepatic stellate and Kupffer cells, stimulating inflammation and fibrosis, which are both precursors for HCC, especially in the vulnerable milieu of a CHB-affected liver (39).

The geographical subgroup analysis performed in the present study provided some noteworthy observations. The studies from Asian countries, which represented a considerable proportion of the studies in the present meta-analysis, demonstrated a non-significant decreased risk of HCC in patients with MAFLD and CHB. By contrast, the pooled OR from studies from other geographical regions indicated a significantly higher risk of HCC in the patients with MAFLD and CHB. For hepatologists practicing in Asia, this information could be important for risk stratification and patient counselling.

Differing study designs also yielded varying results. Notably, prospective cohort studies demonstrated a significant protective effect of MAFLD on the HCC risk in patients with CHB, while retrospective cohort studies did not. This highlighted the inherent challenges of observational studies, in which confounding factors and biases can significantly impact study outcomes.

The apparent protective association between MAFLD and HCC in patients with CHB infection observed in certain studies, although seemingly counterintuitive, may occur due to a number of factors. The presence of MAFLD may modulate the immune response in a manner that could be protective against HCC. For example, certain immune cells that are prevalent in MAFLD, such as regulatory T cells, have previously been reported to suppress liver inflammation. This could potentially mitigate the inflammatory cascades that drive carcinogenesis in patients with CHB infection (40). A liver with MAFLD undergoes a high rate of hepatocyte turnover due to recurrent minor injury and repair. This constant cell renewal could prevent the long-term survival and accumulation of cells with oncogenic mutations induced by CHB infection (41). It has been suggested that lipid accumulation in hepatocytes, known as steatosis, may represent a cellular defense mechanism. Lipids could sequester harmful agents, such as viral proteins or other potential carcinogens, reducing their bioavailability and the harm they would otherwise cause to DNA and the cellular machinery (42). Genetic factors serve a significant role in the susceptibility to, and progression of, liver diseases. Certain genetic polymorphisms [*GCLC* promoter region polymorphism (c. c-129t, rs17883901, single nucleotide polymorphism rs4880)] associated with a higher risk of MAFLD may paradoxically confer a protective effect against HCC development in patients with CHB (43).

One of the major features of the present meta-analysis was the high degree of heterogeneity among the included studies. Several factors may be responsible for this heterogeneity. First, the definition and diagnostic criteria for MAFLD varied across studies, which led to potential misclassifications and introduced variability. Second, there are inherent challenges in collating data from studies spanning diverse populations conducted on the basis of diverse methodologies and time

frames. Meta-regression was used to attempt to identify the sources of the heterogeneity and the influence of certain factors, such as the mean age, follow-up length, study design and geographical location of the study, but none of these factors provided a satisfactory explanation for the observed heterogeneity.

The presence of publication bias, as suggested by Egger's regression asymmetry test results, was observed in the present study. This bias could imply a tendency towards publishing studies with significant findings, thereby possibly artificially enhancing the observed association. However, the non-significant intercept from Egger's test suggested that there may be other contributing factors to the funnel plot asymmetry such as methodological quality variations, artefacts or by chance. Moreover, the quality of evidence was downgraded to very low-quality according to the GRADE criteria, indicating that the certainty in the findings of the present study is limited.

The strengths of the present study lie in its comprehensive approach, rigorous statistical methodologies and subgroup analyses, which add depth to the findings. The inclusion of a diverse set of studies also adds to the generalizability of the present results. However, there were a number of limitations. First, the retrospective nature of most studies posed an inherent challenge with potential confounders. Second, individual patient-level data were unavailable, which restricted the ability to control for other potential confounders such as sociodemographic profile, behavioural risk factors and comorbidities. Moreover, the diagnosis of MAFLD and HCC was not uniform across studies and probably introduced a certain degree of bias. It is important to have uniformity in diagnostic criteria for producing consistent results. However, the criteria for MAFLD diagnosis have evolved over time and a number of the older studies included in the present meta-analysis used previous definitions (17,19), while newer studies adopted more recent criteria (20-22). Establishing a single standardized criterion would exclude a significant portion of available literature, potentially leading to loss of valuable insights. In addition, some studies directly reported the presence of MAFLD without explicitly detailing the diagnostic criteria used (19,30). Excluding these studies based on the absence of a specified criterion would further reduce the number of studies included, potentially compromising the comprehensiveness and depth of the present analysis. However, this heterogeneity was addressed by utilizing the random-effects model in the present meta-analysis, which takes into account the variability among studies. This provided a more conservative estimate of the association and reflected the diversity of included studies.

Potential variability introduced by different follow-up times was also demonstrated across the included articles. The duration of follow-up and interventional treatments received during this period may significantly influence outcomes and introduce heterogeneity among studies. To account for this, a subgroup analysis was conducted based on follow-up duration, segregating studies into categories, such as short-term, medium-term and long-term follow-up. This allowed the assessment to determine if the association between MAFLD and HCC risk in CHB patients was consistent across these subgroups or if duration-specific patterns emerged. Additionally, follow-up duration was included as a covariate in the present meta-regression analysis. This aided in quantifying the potential impact of varying follow-up durations on the observed effect sizes and ensured that the results of the present study considered this important aspect of study design.

There are several avenues for potential future research. Prospective cohort studies, with a standardized diagnostic criterion for MAFLD and adjusted for potential confounders, should provide a more definitive understanding of this possible association between MAFLD and HCC in patients with CHB. Moreover, molecular and genetic studies should elucidate any pathophysiological mechanisms linking MAFLD and HCC in patients with CHB to potentially reveal future therapeutic targets.

The present meta-analysis results were inconclusive for an association between MAFLD and the HCC risk in patients with CHB. These results highlighted the need for more rigorous studies on this complex topic. Currently, clinicians should keep in mind the nuanced nature of risk and the importance of individualized patient care. Continued research in required in this domain, given its profound clinical implications for a vast population of patients with CHB worldwide.

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Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Authors' contributions

SS conceived and designed the study. SS and LP collected the data and performed the literature search. SS wrote the manuscript. Both authors have read and approved the final manuscript. SS and LP confirm the authenticity of all the raw data.

Ethics approval and consent to participate

Not applicable.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1. Chidambaranathan-Reghupaty S, Fisher PB and Sarkar D: Hepatocellular carcinoma (HCC): Epidemiology, etiology and molecular classification. Adv Cancer Res 149: 1-61, 2021.
2. Kulik L and El-Serag HB: Epidemiology and management of hepatocellular carcinoma. Gastroenterology 156: 477-491.e1, 2019.

3. Rapti I and Hadziyannis S: Risk for hepatocellular carcinoma in the course of chronic hepatitis B virus infection and the protective effect of therapy with nucleos(t)ide analogu. World J Hepatol 7: 1064-1073, 2015.
4. Brody H: Hepatitis B. Nature 603: S45, 2022.
5. El-Serag HB: Epidemiology of viral hepatitis and hepatocellular carcinoma. Gastroenterology 142: 1264-1273.e1, 2012.
6. Krump NA and You J: Molecular mechanisms of viral oncogenesis in humans. Nat Rev Microbiol 16: 684-698, 2018.
7. Mui UN, Haley CT and Tyring SK: Viral oncology: Molecular biology and pathogenesis. J Clin Med 6: 111, 2017.
8. Pouwels S, Sakran N, Graham Y, Leal A, Pintar T, Yang W, Kassir R, Singhal R, Mahawar K and Ramnarain D: Non-alcoholic fatty liver disease (NAFLD): A review of pathophysiology, clinical management and effects of weight loss. BMC Endocr Disord 22: 63, 2022.
9. Pipitone RM, Ciccioli C, Infantino G, La Mantia C, Parisi S, Tulone A, Pennisi G, Grimaudo S and Petta S: MAFLD: A multisystem disease. Ther Adv Endocrinol Metab 14: 20420188221145549, 2023.
10. Wang X and Xie Q: Metabolic dysfunction-associated fatty liver disease (MAFLD) and viral hepatitis. J Clin Transl Hepatol 10: 128-133, 2022.
11. van Kleef LA, Choi HSJ, Brouwer WP, Hansen BE, Patel K, de Man RA, Janssen HLA, de Knegt RJ and Sonneveld MJ: Metabolic dysfunction-associated fatty liver disease increases risk of adverse outcomes in patients with chronic hepatitis B. JHEP Rep 3: 100350, 2021.
12. Chen X, Zhou J, Wu L, Zhu X and Deng H: MAFLD is associated with the risk of liver fibrosis and inflammatory activity in HBeAg-negative CHB patients. Diabetes Metab Syndr Obes 15: 673-683, 2022.
13. Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, Shamseer L, Tetzlaff JM, Akl EA, Brennan SE, *et al*: The PRISMA 2020 statement: An updated guideline for reporting systematic reviews. BMJ 372: n71, 2021.
14. Lo CKL, Mertz D and Loeb M: Newcastle-Ottawa Scale: Comparing reviewers' to authors' assessments. BMC Med Res Methodol 14: 45, 2014.
15. Cumpston M, Li T, Page MJ, Chandler J, Welch VA, Higgins JP and Thomas J: Updated guidance for trusted systematic reviews: A new edition of the cochrane handbook for systematic reviews of interventions. Cochrane Database Syst Rev 10: ED000142, 2019.
16. Kirmayr M, Quilodrán C, Valente B, Loezar C, Garegnani L and Franco JVA: The GRADE approach, Part 1: How to assess the certainty of the evidence. Medwave 21: e8109, 2021.
17. Lee YB, Ha Y, Chon YE, Kim MN, Lee JH, Park H, Kim KI, Kim SH, Rim KS and Hwang SG: Association between hepatic steatosis and the development of hepatocellular carcinoma in patients with chronic hepatitis B. Clin Mol Hepatol 25: 52-64, 2019.
18. Chan AWH, Wong GLH, Chan HY, Tong JHM, Yu YH, Choi PCL, Chan HLY, To KF and Wong VWS: Concurrent fatty liver increases risk of hepatocellular carcinoma among patients with chronic hepatitis B. J Gastroenterol Hepatol 32: 667-676, 2017.
19. Oh JH, Lee HW, Sinn DH, Park JY, Kim BK, Kim SU, Kim DY, Ahn SH, Kang W, Gwak GY, *et al*: Controlled attenuation parameter value and the risk of hepatocellular carcinoma in chronic hepatitis B patients under antiviral therapy. Hepatol Int 15: 892-900, 2021.
20. Kim MN, Han K, Yoo J, Hwang SG, Zhang X and Ahn SH: Diabetic MAFLD is associated with increased risk of hepatocellular carcinoma and mortality in chronic viral hepatitis patients. Int J Cancer 153: 1448-1458, 2023.
21. Huang SC, Su TH, Tseng TC, Chen CL, Hsu SJ, Liao SH, Hong CM, Liu CH, Lan TY, Yang HC, *et al*: Distinct effects of hepatic steatosis and metabolic dysfunction on the risk of hepatocellular carcinoma in chronic hepatitis B. Hepatol Int 17: 1139-1149, 2023.
22. Hsueh RC, Wu WJ, Lin CL, Liu CJ, Huang YW, Hu JT, Wu CF, Sung FY, Liu WJ and Yu MW: Impact of PNPLA3 p.I148M and hepatic steatosis on long-term outcomes for hepatocellular carcinoma and HBsAg seroclearance in chronic hepatitis B. J Hepatocell Carcinoma 9: 301-313, 2022.
23. Kim DS, Jeon MY, Lee HW, Kim BK, Park JY, Kim DY, Ahn SH, Han KH and Kim SU: Influence of hepatic steatosis on the outcomes of patients with chronic hepatitis B treated with entecavir and tenofovir. Clin Mol Hepatol 25: 283-293, 2019.
24. Peleg N, Issachar A, Sneh Arbib O, Cohen-Naftaly M, Braun M, Leshno M, Barsheshet A and Shlomai A: Liver steatosis is a strong predictor of mortality and cancer in chronic hepatitis B regardless of viral load. JHEP Rep 1: 9-16, 2019.
25. Yu MW, Lin CL, Liu CJ, Wu WJ, Hu JT and Huang YW: Metabolic-associated fatty liver disease, hepatitis B surface antigen seroclearance, and long-term risk of hepatocellular carcinoma in chronic hepatitis B. Cancers (Basel) 14: 6012, 2022.
26. Chang JW, Lee JS, Lee HW, Kim BK, Park JY, Kim DY, Ahn SH and Kim SU: No influence of hepatic steatosis on the 3-year outcomes of patients with quiescent chronic hepatitis B. J Viral Hepat 28: 1545-1553, 2021.
27. Choi HSJ, Brouwer WP, Zanjir WMR, de Man RA, Feld JJ, Hansen BE, Janssen HLA and Patel K: Nonalcoholic steatohepatitis is associated with liver-related outcomes and all-cause mortality in chronic hepatitis B. Hepatology 71: 539-548, 2020.
28. Lim CT, Goh GBB, Li H, Lim TK, Leow WQ, Wan WK, Azhar R, Chow WC and Kumar R: Presence of hepatic steatosis does not increase the risk of hepatocellular carcinoma in patients with chronic hepatitis b over long follow-Up. Microbiol Insights 13: 1178636120918878, 2020.
29. Rugivarodom M, Pongpaibul A, Chainuvati S, Nimanong S, Chotiyaputta W, Tanwandee T and Charatcharoenwitthaya P: Prognostic relevance of metabolic dysfunction-associated steatohepatitis for patients with chronic hepatitis B. J Clin Transl Hepatol 11: 76-87, 2023.
30. Cho H, Chang Y, Lee JH, Cho YY, Nam JY, Lee YB, Lee DH, Cho EJ, Yu SJ, Kim YJ, *et al*: Radiologic nonalcoholic fatty liver disease increases the risk of hepatocellular carcinoma in patients with suppressed chronic hepatitis B. J Clin Gastroenterol 54: 633-641, 2020.
31. Mak LY, Hui RWH, Fung J, Liu F, Wong DK, Li B, Cheung KS, Yuen MF and Seto WK: Reduced hepatic steatosis is associated with higher risk of hepatocellular carcinoma in chronic hepatitis B infection. Hepatol Int 15: 901-911, 2021.
32. Wang X, Wei S, Wei Y, Wang X, Xiao F, Feng Y and Zhu Q: The impact of concomitant metabolic dysfunction-associated fatty liver disease on adverse outcomes in patients with hepatitis B cirrhosis: A propensity score matching study. Eur J Gastroenterol Hepatol 35: 889-898, 2023.
33. Li J, Yang HI, Yeh ML, Le MH, Le AK, Yeo YH, Dai CY, Barnett S, Zhang JQ, Huang JF, *et al*: Association between fatty liver and cirrhosis, hepatocellular carcinoma, and hepatitis b surface antigen seroclearance in chronic hepatitis B. J Infect Dis 224: 294-302, 2021.
34. Petrescu M, Vlaicu SI, Ciumărnean L, Milaciu MV, Mărginean C, Florea M, Vesa ŞC and Popa M: Chronic inflammation-A link between nonalcoholic fatty liver disease (NAFLD) and dysfunctional adipose tissue. Medicina (Kaunas) 58: 641, 2022.
35. Chen J, Deng X, Liu Y, Tan Q, Huang G, Che Q, Guo J and Su Z: Kupffer cells in non-alcoholic fatty liver disease: Friend or foe? Int J Biol Sci 16: 2367-2378, 2020.
36. Sakurai Y, Kubota N, Yamauchi T and Kadowaki T: Role of insulin resistance in MAFLD. Int J Mol Sci 22: 4156, 2021.
37. Ma Y, Lee G, Heo SY and Roh YS: Oxidative stress is a key modulator in the development of nonalcoholic fatty liver disease. Antioxidants (Basel) 11: 91, 2021.
38. Zorena K, Jachimowicz-Duda O, Ślęzak D, Robakowska M and Mrugacz M: Adipokines and obesity. Potential link to metabolic disorders and chronic complications. Int J Mol Sci 21: 3570, 2020.
39. Brenner DA, Paik YH and Schnabl B: Role of gut microbiota in liver disease. J Clin Gastroenterol 49 (Suppl 1): S25-S27, 2015.
40. Kountouras J, Kazakos E, Kyrailidi F, Polyzos SA, Zavos C, Arapoglou S, Boziki M, Mouratidou MC, Tzitiridou-Chatzopoulou M, Chatzopoulos D, *et al*: Innate immunity and nonalcoholic fatty liver disease. Ann Gastroenterol 36: 244-256, 2023.
41. Duncan AW, Dorrell C and Grompe M: Stem cells and liver regeneration. Gastroenterology 137: 466-481, 2009.
42. Ipsen DH, Lykkesfeldt J and Tveden-Nyborg P: Molecular mechanisms of hepatic lipid accumulation in non-alcoholic fatty liver disease. Cell Mol Life Sci 75: 3313-3327, 2018.
43. Severson TJ, Besur S and Bonkovsky HL: Genetic factors that affect nonalcoholic fatty liver disease: A systematic clinical review. World J Gastroenterol 22: 6742-6756, 2016.