Research Square

Preprints are preliminary reports that have not undergone peer review.
They should not be considered conclusive, used to inform clinical practice,
or referenced by the media as validated information.

Effect of Positive Hepatitis B Surface Antigen on The Risk of Synchronous Liver Metastasis: A Retrospective Study of 868 Consecutive Patients of Newly Diagnosed Gastric Cancer

Tingya Wang

Department of oncology, The Affiliated Zhongda Hospital of Southeast University, Medical School of Southeast University, 87 Dingjia Bridge Road, Nanjing, China

Haijun Zhang (haijunzhang@seu.edu.cn)

Southeast University Zhongda Hospital

Research

Keywords: gastric cancer, liver metastasis, hepatitis B virus, cirrhosis, liver microenvironment

Posted Date: December 1st, 2020

DOI: https://doi.org/10.21203/rs.3.rs-113459/v1

License: This work is licensed under a Creative Commons Attribution 4.0 International License.
Read Full License

Abstract

Background. The study aimed to explore the influence of hepatitis B virus (HBV) infection on the risk of synchronous gastric cancer liver metastasis (synGCLM).

Methods. This was a retrospective study which enrolled 868 patients with newly diagnosed gastric cancer (GC). The study compared the prevalence of synGCLM between hepatitis B surface antigen (HBsAg)-positive ($HBsAg^+$) and -negative ($HBsAg^-$) patients. Logistic regression analysis was utilized to analyze the risk factors for synGCLM. Among patients with and without synGCLM, aspartate aminotransferase to platelet ratio index (APRI), liver fibrosis-4 index (FIB-4) and hepatitis B e antigen (HBeAg) status were further analyzed.

Results. The prevalence of synGCLM in the $HBsAg^+$ patients was higher than that in the $HBsAg^-$ patients, which was statistically significant ($P = 0.025$). Multivariate logistic regression analysis demonstrated that HBsAg, the elevated level of carcinoembryonic antigen (CEA), alpha-fetoprotein (AFP), γ-glutamyltransferase (GGT) and alkaline phosphatase (ALP) were risk factors for synGCLM. Among the $HBsAg^+$ patients, both ARPI and FIB-4 were significantly higher in the patients with synGCLM ($synGCLM^+$) than those without synGCLM ($synGCLM^-$) (ARPI: $P = 0.045$; FIB-4: $P = 0.047$); HBeAg positivity was detected in 20.0% of $synGCLM^+$ patients compared to 6.0% of $synGCLM^-$ patients, but the difference was of no significance ($P = 0.190$).

Conclusions. HBV infection significantly increases the risk of synGCLM, and elevated ARPI and FIB-4 may be pro-metastatic especially among the $HBsAg^+$ GC patients.

Background

Gastric cancer (GC) ranks the fifth most frequent cancer, and the third most common cause of cancer-related death over the world [1]. GC is more severe in China as it ranks the second commonly diagnosed cancer in males and the second leading cause of cancer-related death. Newly reported by National Cancer Center, there were approximately 679,100 new cases and 498,000 deaths from GC [2]. GC patients always have a poor prognosis due to presenting with distant metastases at initial diagnosis of the primary tumor. And the liver is the most common organ of GC hematogenous metastasis as a result of its unique anatomical structure [3]. Previous studies have shown that about 2.0%-9.9% of patients present synchronous gastric cancer liver metastases (synGCLM) at initial diagnosis of primary site, and up to 37% of GC patients develop metachronous liver metastasis after radical gastrectomy [4]. Unfortunately, the 5-year survival rate of synGCLM is less than 20% [5] due to losing the chance of undergoing curative resection and optimal treatments [6-10].

Distant metastasis is one of the most significant prognostic factors in advanced GC. The seed and soil hypothesis refers that the compatibility between tumor cells (seed) and a pre-metastatic niche (soil) is needed, which explains the mechanism of cancer metastasis [11, 12]. Previous clinical studies focused

on the risk factors for gastric cancer liver metastases (GCLM) from the side of ‘seed’, such as the aggressive behavior of tumor cells [13, 14]. However, our article tried to explore the impact of the liver-specific microenvironment (‘soil’) on GC metastasis, which rarely debated.

Hepatitis B virus (HBV) infection is prevalent in China as the prevalence of hepatitis B surface antigen (HBsAg) seropositivity is 5%-6% in the general population [15, 16], which plays a vital role in the progress of liver diseases including hepatocellular carcinoma (HCC) [17-19]. With the notable prevalence of HBV infection in China, we intented to explore the impact of HBV infection on premetastatic microenvironment formation. In the study, by conducting a large-sample, retrospective study of 868 cases with newly-diagnosed GC, we obtained some meaningful and unique results of the impact of HBV infection on synGCLM.

Methods

Study population

This is a retrospective, cross-sectional study of all consecutive newly-diagnosed GC patients who presented to the Affiliated Zhongda Hospital of Southeast University from May 2013 to May 2020. The inclusion criteria were as follows: 1) Newly diagnosis GC by pathological reports of gastroscopic biopsy or surgical specimen in our hospital; 2) No surgery, chemotherapy, radiotherapy or other treatments in other hospitals before admission; 3) Liver metastases confirmed by ultrasound, computed tomography (CT), magnetic resonance imaging (MRI), positron emission tomography/computed tomography (PET/CT) or liver biopsy, and complete imaging examination to detect other distant organ metastases; 4) Complete medical records including demographic information, serologic assay, tumor characteristics. We excluded the patients who conformed to the following standards: 1) Pathologically confirmed as gastric lymphoma, gastrointestinal stromal neoplasms, gastric leiomyoma; 2) Accompany with hepatitis virus infection other than HBV or with liver diseases such as fatty liver, alcohol liver and further apparent liver damage; 3) Lack of necessary clinical information; 4)History of primary tumors in other organs except for stomach.

Data characteristics

We collected information including primary data: gender, age, laboratory tests: HBsAg, aspartate aminotransferase (AST), alanine aminotransferase (ALT), γ-glutamyltransferase (GGT), alkaline phosphatase (ALP), total bilirubin (TBIL), platelet (PLT), fibrinogen (FIB) and tumor markers: carcinoembryonic antigen (CEA), carbohydrate antigen 19-9 (CA199), alpha-fetoprotein (AFP). And calculated ALT to platelet ratio index (APRI) and liver fibrosis-4 index (FIB-4) among the patients with HBsAg-positive ($HBsAg^+$) to assess the degree of HBV-related liver fibrosis. The upper limit of normal AST was 40 IU/L in our hospital. Pathological data of all patients were also collected, including primary tumor location, differentiation and histological type.

SynGCLM defined as liver metastasis detected before or at the meantime of diagnosing primary GC, including within six months after the diagnosis of the primary site, since that some suspicious liver lesions needed to follow up to clarify if they were liver metastases or not. HBV infection characterized by HBsAg seropositivity [20]. And $HBsAg^+$ was defined as detection of serum HBsAg more than 10 IU/mL by serologic assay in our hospital.

Statistical analysis

The study compared baseline clinicopathological features between $HBsAg^+$ and HBsAg-negative ($HBsAg^-$) patients. Continuous variables with skewed distribution were compared by Wilcoxon rank-sum test and expressed by median (range). Qualitative variables were compared by Chi-square test, Fisher's exact test and described in the form of a percentage. Possible risk factors for synGCLM were analyzed first by univariate logistic regression analysis, and then performed multivariate logistic regression analysis on the covariates which were of statistical significance in univariate analysis. All statistical analyses were performed by SPSS statistical software (version 25.0). And statistical significance was considered as two-sided P value < 0.05.

Results

The study enrolled a total of 868 patients with newly diagnosed GC between May 2013 and May 2020 in our hospital. There were 60 (6.9%) patients with HBsAg and 808 (93.1%) without HBsAg. According to the HBV infection status, these cases divided into two groups: $HBsAg^+$ and $HBsAg^-$. The clinicopathologic characteristics of these two groups summarized in **Table 1**. By Chi-square test, the prevalence of synGCLM in the $HBsAg^+$ group was 16.7%, which was significantly higher than that of 8.2% in the $HBsAg^-$ group (P = 0.025). No significant difference found in gender, age, tumor location, grade, histological type between $HBsAg^+$ and $HBsAg^-$ patients. **Figure 1** showed the typical CT, MRI and histopathological images of HBV infection leading to liver metastasis in GC patients, respectively.

For patients with synGCLM ($synGCLM^+$) and without synGCLM ($synGCLM^-$), there were no significant differences in terms of gender, tumor location, grade, PLT, TBIL, FIB (P > 0.05) in **Table 2**. Following univariate logistic regression, HBsAg, CEA, CA199, AFP, AST, ALT, GGT, ALP, histological type and age were statistically significant associated with synGCLM, which shown in **Table 3**. These ten risk factors were further chosen for the subsequent multivariate logistic regression analysis to find out the independent risk factors of synGCLM (**Table 4**). The results demonstrated that not only $HBsAg^+$ (vs $HBsAg^-$; OR, 2.849; 95% confidence interval, 1.130-7.186; P = 0.027), but also elevated levels of preoperative CEA, AFP, GGT, ALP were significant risk factors for synGCLM. Furthermore, receiver operating characteristic (ROC) curve analysis revealed that the combination of CEA and AFP show perfect sensitivity and specificity for synGCLM in **Figure 2**.

ARPI and FIB-4 were calculated among 60 $HBsAg^+$ patients. The median (25th-75th percentile) ARPI in the $synGCLM^+$ patients was 0.629 (0.270-1.147), was significantly higher than 0.278 (0.218-0.409) of

synGCLM$^-$ patients (Z = 2.003, *P* = 0.045, Mann-Whitney test). The median (25th-75th percentile) FIB-4 in synGCLM+ patients was 2.358 (1.775-4.489), which was also significantly higher compared to the median of 1.698 (1.098-2.403) in synGCLM- (Z = 1.984, *P* = 0.047, Mann-Whitney test).

HBeAg positivity was also detected in the 2 of 10 (20.0%) synGCLM+ patients and 3 of 50 (6.0%) synGCLM- patients. The prevalence synGCLM was 40.0% (2/5) in the patients with HBeAg (HBeAg+) compared to that of 14.5% (8/55) in the patients without HBeAg (HBeAg-), but the difference was not statistically significant (*P* = 0.190, χ^2 test) in **Table 5**.

Discussion

The liver is the most frequent site of hematogenous metastasis in GC, and the patients usually have dismal prognosis when diagnosed with GCLM [21]. Considering that liver metastasis commonly accompanies with simultaneous peritoneal and distant lymph node metastases, the guideline recommends chemotherapy instead of surgery as the first-line treatment for GCLM [3, 22]. The famous seed and soil hypothesis proposes that only the microenvironment that has acquired the capacities of engraftment, survival support and immune evasion could become a new metastatic target organ [23]. In the process of cancer metastasis, cancer cells first form micrometastases in distant metastatic organs and then proliferate into visible metastases. The specific organ microenvironment can promote the colonization, survival and proliferation of tumor cells. However, lack of research shed light on the liver microenvironment (soil) in GCLM. To early detect the high-risk individuals of synGCLM, the study intented to declare whether liver disease would remodel its microenvironment to become a fertile, premetastatic soil for GC cells (seed) to land and proliferate. Here we conducted a retrospective study to demonstrate the impact of HBV infection on the metastatic pattern of GC.

We found that the prevalence of HBsAg$^+$ was 6.9% among all the GC patients, in accordance with that in the general population in China [16]. The prevalence of synGCLM in our study was 8.8%, which was also consistent with that reported by previous studies [4]. Results illustrated that the prevalence of synGCLM in the HBsAg$^+$ patients was 16.7%, significantly higher than that of 8.2% in the HBsAg$^-$ patients (*P* = 0.025). As HBV infection affected the metastatic pattern of tumor, a study in Italy reported that the presence of HBsAg in patients with colorectal carcinoma significantly increased the incidence of liver metastasis recently [18]. Meanwhile, with the stimulation of HBsAg, interleukin (IL) -10, IL-12 and interferon (IFN) λ could be produced by dendritic cells, which played a vital role in the progression of HBV-related tumor [24]. Moreover, HBV-encoded X protein (HBx), as the only one protein encoded by the HBV genome and expressed by transformed hepatocytes, could affect differentiation and promote tumor progression metastasis, which proved by the positive cross-talk with the metastasis-associated 1 (MTA1) in stabilizing hypoxia-inducible factor (HIF)-1α [25, 26]. Experiments in vivo and vitro also proved that HBx could promote tumor invasion by up-regulating matrix metalloproteinases (MMPs) and cyclooxygenase (COX)-2, which provides new insights into its involvement in tumor metastasis and recurrence [27]. Also, HBx could up-regulate long non-coding RNA MALAT1 and further activate LTBP3, which resulted in the development and metastasis of tumor [28]. Furthermore, the transforming growth factor (TGF)-β

production stimulated by HBx could promote Treg cell recruitment, which maintained a tolerogenic liver microenvironment and finally induced the development of metastasis [29, 30]. These above mechanisms may explain how HBV infection adjust the liver microenvironment becoming a premetastatic niche (soil) for circulating tumor cells (seed) to colonize. Besides, a study conducted by Wei verified the objective evidence of the existence of HBV infection in GC tissues, it proposed that HBV infection was a possible significant predictor for the development of GC [31]. The influence of HBV infection on both GC tumor cells and liver microenvironment may mediate GC metastasis, and the interaction between tumor cells and liver microenvironment need to be further investigated.

Inspired by the classic hepatitis-cirrhosis-HCC pathway [32], we further investigated whether liver fibrosis/cirrhosis related to HBV infection influenced GCLM. As liver biopsy was seldom utilized in clinical work, we used the noninvasive liver fibrosis model to assess the degree of liver fibrosis in the 60 GC patients infected with HBV. Due to ARPI and FIB-4 were both efficient indexes used to evaluate the degree of HBV-related liver fibrosis, which took the factors like the age, platelet, AST, ALT of patients into consideration, and widely available in the clinical setting [33]. The study demonstrated that both the median of ARPI and FIB-4 in the $HBsAg^+$ patients with synGCLM were significantly higher than those among the $HBsAg^+$ patients without synGCLM. HBeAg positivity in $HBsAg^+$ patients with synGCLM was higher compared with that in $HBsAg^+$ patients without synGCLM, although the difference was of no significance. A study conducted by Chiou in Taiwan drew a similar conclusion that cancer patients with liver cirrhosis had a higher risk for liver metastasis in colorectal cancer [34]. Another study from China also reported that fibrosis niche might be a favourable microenvironment for the formation of hepatic metastasis based on noninvasive liver fibrosis scores [35]. The phenomenon may be resulted from local microenvironment change and immune reaction alteration. The continuous production of HBsAg can cause persistent HBV infection-associated inflammation, which accompanied by the transdifferentiation of hepatic stellate cells (HSC) into myofibroblasts. At the meantime, these fibroblasts will propagate, along with the deposition of the extracellular matrix, consequently lead to liver fibrosis even cirrhosis [36-38]. On the other hand, as the component of prometastatic liver microenvironment, HSC can promote tumor growth, facilitate tumor invasion and suppress the anti-tumor immune response [39]. Moreover, the angiogenic factors such as vascular endothlial growth factor (VEGF) and angiopoietin 1 in liver fibrosis environment could promote tumor angiogenesis [40]. In conclusion, more animal experiments and prospective clinical trials need to dig into the underlying mechanism of the cirrhotic microenvironment and active virus replication in the metastasis of GC.

There were several limitations in that it was a retrospective, cross-sectional study conducted in only one single center. The comorbidities like diabetes, obesity and the HBV-DNA level of all patients were insufficient. And liver fibrosis/cirrhosis was measured by ARPI/FIB-4, other than a liver biopsy. Despite these limitations, our study still enrolled a large number of newly diagnosed GC patients, and all with information about hepatitis B status. Moreover, our study investigated the effect of HBV infection only on synchronous liver metastasis, to avoid the interference of different anti-cancer treatments among GC patients and imprecision information about metachronous metastasis. Besides, our study was the first

study that demonstrated the impact of not only HBV infection but also HBV-related cirrhosis on liver metastasis formation and survival in GC. Overall, our study discovered that HBV infection and HBV-related cirrhosis might remodel the liver microenvironment, becoming a premetastatic niche for GC cells to proliferate. This unique and exciting information may provide novel anti-cancer treatments on blocking premetastatic niche formation (soil) to prevent liver metastasis and even cure gastric cancer metastasis in the future. More prospective studies with larger sample size and animal experiments are needed to validate our findings and further explore the cause-effect relationship between HBV infection and synGCLM.

Conclusion

Our study found that HBV infection may increase the risk of liver metastasis in GC patients, and for hepatitis B surface antigen positive ($HBsAg^+$) GC patients, HBV-related liver fibrosis and cirrhosis may be prometastatic. This may provide novel anti-cancer treatments on blocking premetastatic niche formation to prevent liver metastasis. More investigations need to be taken to explore the mechanism of how HBV-related microenvironment promoting liver metastasis.

Abbreviations

AFP, alpha-fetoprotein; ALP, alkaline phosphatase; ALT, alanine aminotransferase; APRI, aspartate aminotransferase to platelet ratio index; AST, aspartate aminotransferase; CA199, carbohydrate antigen 19-9; CEA, carcinoembryonic antigen; COX, cyclooxygenase; CT, computed tomography; FIB, fibrinogen; FIB-4, liver fibrosis-4 index; GC, gastric cancer; GCLM, gastric cancer liver metastases; GGT, γ-glutamyltransferase; HBeAg, hepatitis B e antigen; HBsAg, hepatitis B surface antigen; HBV, hepatitis B virus; HBx, HBV-encoded X protein; HCC, hepatocellular carcinoma; HIF, hypoxia-inducible factor; HSC, hepatic stellate cells; IL, interleukin; IFN, interferon; MMPs, matrix metalloproteinases; MRI, magnetic resonance imaging; MTA1, metastasis-associated 1; PET/CT, positron emission tomography/computed tomography; PLT, platelet; ROC, receiver operating characteristic; synGCLM, synchronous gastric cancer liver metastasis; TBIL, total bilirubin; TGF, transforming growth factor; VEGF, vascular endothlial growth factor.

Declarations

Acknowledgments

Not applicable.

Authors' contributions

TYW complete data collection, data analysis, manuscript drafting. HJZ directed the arrangement of the study and supervised the whole writing of the manuscript and manuscript revision. All the authors contributed to the review and revison of the manuscript and have read and revised the final version.

Funding

Not applicable.

Availabilty of data

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

Ethics approval and consent to participate

The study was approved by the ethical committee of the Affiliated Zhongda Hospital of Southeast University.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Author details

[1] Department of oncology, The Affiliated Zhongda Hospital of Southeast University, Medical School of Southeast University, 87 Dingjia Bridge Road, Nanjing, China.

References

1. Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A: **Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries**. *CA Cancer J Clin* 2018, **68**(6):394-424.
2. Chen W, Zheng R, Baade PD, Zhang S, Zeng H, Bray F, Jemal A, Yu XQ, He J: **Cancer statistics in China, 2015**. *CA Cancer J Clin* 2016, **66**(2):115-132.
3. D'Angelica M, Gonen M, Brennan MF, Turnbull AD, Bains M, Karpeh MS: **Patterns of initial recurrence in completely resected gastric adenocarcinoma**. *Ann Surg* 2004, **240**(5):808-816.
4. Sakamoto Y, Sano T Fau - Shimada K, Shimada K Fau - Esaki M, Esaki M Fau - Saka M, Saka M Fau - Fukagawa T, Fukagawa T Fau - Katai H, Katai H Fau - Kosuge T, Kosuge T Fau - Sasako M, Sasako M: **Favorable indications for hepatectomy in patients with liver metastasis from gastric cancer**. (0022-4790 (Print)).
5. Xiao Y, Zhang B, Wu Y: **Prognostic analysis and liver metastases relevant factors after gastric and hepatic surgical treatment in gastric cancer patients with metachronous liver metastases: a population-based study**. *Irish journal of medical science* 2019, **188**(2):415-424.

6. Wang W, Liang H, Zhang H, Wang X, Xue Q, Zhang R: **Prognostic significance of radical surgical treatment for gastric cancer patients with synchronous liver metastases**. *Med Oncol* 2014, **31**(11):258.
7. Kataoka K, Kinoshita T, Moehler M, Mauer M, Shitara K, Wagner AD, Schrauwen S, Yoshikawa T, Roviello F, Tokunaga M *et al*: **Current management of liver metastases from gastric cancer: what is common practice? New challenge of EORTC and JCOG**. *Gastric Cancer* 2017, **20**(5):904-912.
8. Ministrini S, Solaini L, Cipollari C, Sofia S, Marino E, D'Ignazio A, Bencivenga M, Tiberio GAM: **Surgical treatment of hepatic metastases from gastric cancer**. *Updates Surg* 2018, **70**(2):273-278.
9. Oki E, Tokunaga S, Emi Y, Kusumoto T, Yamamoto M, Fukuzawa K, Takahashi I, Ishigami S, Tsuji A, Higashi H *et al*: **Surgical treatment of liver metastasis of gastric cancer: a retrospective multicenter cohort study (KSCC1302)**. *Gastric Cancer* 2016, **19**(3):968-976.
10. Tao F, Lv J, Wang W, Jin K: **Clinical modalities for management of gastric cancer hepatic metastasis**. *Int J Clin Exp Med* 2015, **8**(11):19850-19858.
11. Liu Q, Zhang H, Jiang X, Qian C, Liu Z, Luo D: **Factors involved in cancer metastasis: a better understanding to "seed and soil" hypothesis**. (1476-4598 (Electronic)).
12. Shimizu D, Kanda M, Kodera Y: **Emerging evidence of the molecular landscape specific for hematogenous metastasis from gastric cancer**. *World J Gastrointest Oncol* 2018, **10**(6):124-136.
13. Marrelli D, Roviello F, De Stefano A, Fotia G, Giliberto C, Garosi L, Pinto E: **Risk factors for liver metastases after curative surgical procedures for gastric cancer: a prospective study of 208 patients treated with surgical resection**. *J Am Coll Surg* 2004, **198**(1):51-58.
14. Sun Z, Zheng H, Yu J, Huang W, Li T, Chen H, Hu Y, Zhao M, Liu H, Jiang Y *et al*: **Liver Metastases in Newly Diagnosed Gastric Cancer: A Population-Based Study from SEER**. *J Cancer* 2019, **10**(13):2991-3005.
15. Liu Z, Yang Q, Shi O, Ye W, Chen X, Zhang T: **The epidemiology of hepatitis B and hepatitis C infections in China from 2004 to 2014: An observational population-based study**. *J Viral Hepat* 2018, **25**(12):1543-1554.
16. Seto WK, Lo YR, Pawlotsky JM, Yuen MF: **Chronic hepatitis B virus infection**. *Lancet* 2018, **392**(10161):2313-2324.
17. Huo T, Cao J, Tian Y, Shi X, Wu L, Zhang M, Wong LL, Zhao L: **Effect of Concomitant Positive Hepatitis B Surface Antigen on the Risk of Liver Metastasis: A Retrospective Clinical Study of 4033 Consecutive Cases of Newly Diagnosed Colorectal Cancer**. *Clin Infect Dis* 2018, **66**(12):1948-1952.
18. Schinzari V, Barnaba V, Piconese S: **Chronic hepatitis B virus and hepatitis C virus infections and cancer: synergy between viral and host factors**. *Clin Microbiol Infect* 2015, **21**(11):969-974.
19. Wei XL, Qiu Mz Fau - Chen W-w, Chen Ww Fau - Jin Y, Jin Y Fau - Ren C, Ren C Fau - Wang F, Wang F Fau - Luo H-y, Luo Hy Fau - Wang Z-q, Wang Zq Fau - Zhang D-s, Zhang Ds Fau - Wang F-h, Wang Fh Fau - Li Y-h *et al*: **The status of HBV infection influences metastatic pattern and survival in Chinese patients with pancreatic cancer**. (1479-5876 (Electronic)).

20. Yuen MF, Chen DS, Dusheiko GM, Janssen HLA, Lau DTY, Locarnini SA, Peters MG, Lai CL: **Hepatitis B virus infection**. *Nat Rev Dis Primers* 2018, **4**:18035.
21. Kataoka KA-O, Kinoshita T, Moehler M, Mauer M, Shitara K, Wagner AD, Schrauwen S, Yoshikawa T, Roviello F, Tokunaga M *et al*: **Current management of liver metastases from gastric cancer: what is common practice? New challenge of EORTC and JCOG**. (1436-3305 (Electronic)).
22. Oguro S, Imamura H, Yoshimoto J, Ishizaki Y, Kawasaki S: **Liver metastases from gastric cancer represent systemic disease in comparison with those from colorectal cancer**. *J Hepatobiliary Pancreat Sci* 2016, **23**(6):324-332.
23. Zhong J, Chen Y, Wang LJ: **Emerging molecular basis of hematogenous metastasis in gastric cancer**. *World J Gastroenterol* 2016, **22**(8):2434-2440.
24. Yang P, Markowitz GJ, Wang XF: **The hepatitis B virus-associated tumor microenvironment in hepatocellular carcinoma**. *Natl Sci Rev* 2014, **1**(3):396-412.
25. Kordestani R, Mirshafiee H, Hosseini SM, Sharifi Z: **Effect of Hepatitis B Virus X Gene on the Expression Level of p53 Gene using Hep G2 Cell Line**. *Avicenna J Med Biotechnol* 2014, **6**(1):3-9.
26. Wang H, Chen L: **Tumor microenviroment and hepatocellular carcinoma metastasis**. (1440-1746 (Electronic)).
27. Lara-Pezzi E, Gomez-Gaviro MV, Galvez BG, Mira E, Iniguez MA, Fresno M, Martinez AC, Arroyo AG, Lopez-Cabrera M: **The hepatitis B virus X protein promotes tumor cell invasion by inducing membrane-type matrix metalloproteinase-1 and cyclooxygenase-2 expression**. *J Clin Invest* 2002, **110**(12):1831-1838.
28. Hou Z, Xu X, Fu X, Tao S, Zhou J, Liu S, Tan D: **HBx-related long non-coding RNA MALAT1 promotes cell metastasis via up-regulating LTBP3 in hepatocellular carcinoma**. *Am J Cancer Res* 2017, **7**(4):845-856.
29. Peng G, Li S Fau - Wu W, Wu W Fau - Sun Z, Sun Z Fau - Chen Y, Chen Y Fau - Chen Z, Chen Z: **Circulating CD4+ CD25+ regulatory T cells correlate with chronic hepatitis B infection**. (1365-2567 (Electronic)).
30. Yang P, Li Qj Fau - Feng Y, Feng Y Fau - Zhang Y, Zhang Y Fau - Markowitz GJ, Markowitz Gj Fau - Ning S, Ning S Fau - Deng Y, Deng Y Fau - Zhao J, Zhao J Fau - Jiang S, Jiang S Fau - Yuan Y, Yuan Y Fau - Wang H-Y *et al*: **TGF-β-miR-34a-CCL22 signaling-induced Treg cell recruitment promotes venous metastases of HBV-positive hepatocellular carcinoma**. (1878-3686 (Electronic)).
31. Wei XL, Qiu MZ, Jin Y, Huang YX, Wang RY, Chen WW, Wang DS, Wang F, Luo HY, Zhang DS *et al*: **Hepatitis B virus infection is associated with gastric cancer in China: an endemic area of both diseases**. (1532-1827 (Electronic)).
32. Yuen MF, Chen DS, Dusheiko GM, Janssen HLA, Lau DTY, Locarnini SA, Peters MG, Lai CL: **Hepatitis B virus infection**. (2056-676X (Electronic)).
33. Kim WR, Berg T, Asselah T, Flisiak R, Fung S, Gordon SC, Janssen HL, Lampertico P, Lau D, Bornstein JD *et al*: **Evaluation of APRI and FIB-4 scoring systems for non-invasive assessment of hepatic fibrosis in chronic hepatitis B patients**. *J Hepatol* 2016, **64**(4):773-780.

34. Chiou WY, Chang CM, Tseng KC, Hung SK, Lin HY, Chen YC, Su YC, Tseng CW, Tsai SJ, Lee MS *et al*: **Effect of liver cirrhosis on metastasis in colorectal cancer patients: a nationwide population-based cohort study**. *Jpn J Clin Oncol* 2015, **45**(2):160-168.
35. Hu X, Marietta A, Dai WX, Li YQ, Ma XJ, Zhang L, Cai SJ, Peng JJ: **Prediction of hepatic metastasis and relapse in colorectal cancers based on concordance analyses with liver fibrosis scores**. (2001-1326 (Print)).
36. Suhail M, Abdel-Hafiz H, Ali A, Fatima K, Damanhouri GA, Azhar E, Chaudhary AG, Qadri I: **Potential mechanisms of hepatitis B virus induced liver injury**. (2219-2840 (Electronic)).
37. Ryder SD, Irving Wl Fau - Jones DA, Jones Da Fau - Neal KR, Neal Kr Fau - Underwood JC, Underwood JC: **Progression of hepatic fibrosis in patients with hepatitis C: a prospective repeat liver biopsy study**. (0017-5749 (Print)).
38. Yao QY, Feng YD, Han P, Yang F, Song GQ: **Hepatic microenvironment underlies fibrosis in chronic hepatitis B patients**. (2219-2840 (Electronic)).
39. Kang N, Gores Gj Fau - Shah VH, Shah VH: **Hepatic stellate cells: partners in crime for liver metastases?** (1527-3350 (Electronic)).
40. Taura K, De Minicis S Fau - Seki E, Seki E Fau - Hatano E, Hatano E Fau - Iwaisako K, Iwaisako K Fau - Osterreicher CH, Osterreicher Ch Fau - Kodama Y, Kodama Y Fau - Miura K, Miura K Fau - Ikai I, Ikai I Fau - Uemoto S, Uemoto S Fau - Brenner DA *et al*: **Hepatic stellate cells secrete angiopoietin 1 that induces angiogenesis in liver fibrosis**. (1528-0012 (Electronic)).

Tables

Table 1 Baseline Characteristics of Patients With Gastric Cancer in Hepatitis B Surface Antigen-Positive and -Negative Cohorts

Factor	$HBsAg^+$ (n =60)	$HBsAg^-$ (n = 808)	Z/χ^2 Value	*P* Value
synGCLM(yes/no)	10/50	66/742	5.049	0.025
Gender(male/female)	42/18	564/244	0.001	0.974
Age, y, median(range)	63(31-87)	65(25-94)	-1.624	0.104
Primary GC				
Tumor location			0.215	0.643
Cardia	13(21.7)	164(20.3)		
Others	47(78.3)	644(79.7)		
Grade			0.090	0.764
Well or moderate	19(31.7)	241(29.8)		
Poor or undifferentiated	41(68.3)	567(70.2)		
Histological classification				0.513
Adenocarcinoma	59(98.3)	799(98.9)		
Non-adenocarcinoma	1(1.7)	9(1.1)		

Data are prentented as No. (%) unless otherwise indicated.

Abbreviations: HBsAg, hepatitis B surface antigen; synGCLM, synchronous gastric cancer liver metastasis.

Table 2 Clinicopathological Parameters of Gastric Cancer in synGCLM-Positive and -Negative Cohorts

Factor	synGCLM^{+} (n = 76)	synGCLM^{-} (n = 792)	Z/χ^2 Value	*P* Value
HBsAg(yes/no)	10/66	50/742	5.049	0.025
Gender(male/female)	57/19	549/243	1.062	0.303
Age, y, median(range)	67(25-94)	65(25-90)	2.069	0.039
Primary GC				
Tumor location			0.022	0.881
Cardia	16(21.1)	161(20.3)		
Others	60(78.9)	631(79.7)		
Grade			0.105	0.746
Well or moderate	24(31.6)	236(29.8)		
Poor or undifferentiated	52(68.4)	556(70.2)		
Histological classification				0.049
Adenocarcinoma	73(96.1)	785(99.1)		
Non-adenocarcinoma	3(3.9)	7(0.9)		
CEA, ng/mL			54.483	<0.001
>5	46(60.5)	174(22.0)		
≤5	30(39.5)	618(78.0)		
CA199, U/mL			26.237	<0.001
>39	29(38.2)	119(15.0)		
≤39	47(61.8)	673(85.0)		
AFP, ng/mL			68.988	<0.001
>7	26(34.2)	49(6.2)		
≤7	50(65.8)	743(93.8)		
PLT, 10^9/L			0.434	0.510
≥125	74(97.4)	752(94.9)		
<125	2(2.6)	40(5.1)		
AST, U/L			54.665	<0.001
>40	19(25.0)	30(3.8)		
≤40	57(75.0)	762(96.2)		

ALT, U/L			9.778	0.002
>50	9(11.8)	28(3.5)		
≤50	67(88.2)	764(96.5)		
GGT, U/L			99.234	<0.001
>60	31(40.8)	49(6.2)		
≤60	45(59.2)	743(93.8)		
ALP, U/L			105.984	<0.001
>125	27(35.5)	33(4.2)		
≤125	49(64.5)	759(95.8)		
TBIL, umol/L			2.914	0.088
>20	10(13.2)	60(7.6)		
≤20	66(86.8)	732(92.4)		
FIB, g/L			3.581	0.058
>4	41(53.9)	338(42.7)		
≤4	35(46.1)	454(57.3)		

Data are prentented as No. (%) unless otherwise indicated.

Abbreviations:HBsAg, hepatitis B surface antigen; synGCLM, synchronous gastric cancer liver metastasis; CEA, carcinoembryonic ntigen; CA199, carbohydrate antigen 199; AFP, alpha fetoprotein; PLT, platelet; AST, aspartate aminotransferase; ALT, alanine aminotransferase; GGT, γ-glutamyltransferase; ALP, alkaline phosphatase; TBIL, total bilirubin; FIB, fibrinogen.

Table 3 Univariate Logistic Regression Analysis of the Predictors for Synchronous Gastric Cancer Liver Metastasis

Variable	Coefficient	SE	Wald χ^2	*P* Value	Odds Ratio	(95%CI)
HBsAg	0.810	0.369	4.810	0.028	2.248	(1.090-4.638)
CEA	1.695	0.250	46.008	<0.001	5.446	(3.337-8.887)
CA199	1.250	0.256	23.792	<0.001	3.490	(2.112-5.766)
AFP	2.065	0.283	53.157	<0.001	7.885	(4.526-13.737)
AST	2.136	0.324	43.532	<0.001	8.467	(4.489-15.970)
ALT	1.299	0.404	10.347	0.001	3.665	(1.661-8.088)
GGT	2.346	0.276	72.207	<0.001	10.446	(6.080-17.946)
ALP	2.540	0.298	72.408	<0.001	12.673	(7.061-22.747)
Histological classification	-1.528	0.701	4.753	0.029	0.217	(0.055-0.857)
Age	0.021	0.011	4.026	0.045	1.021	(1.000-1.043)

Abbreviations: CI, confidence interval; HBsAg, hepatitis B surface antigen; CEA, carcinoembryonic ntigen; CA199, carbohydrate antigen 199; AFP, alpha fetoprotein; AST, aspartate aminotransferase; ALT, alanine aminotransferase; GGT, γ-glutamyltransferase; ALP, alkaline phosphatase; SE, standard error.

Table 4 Multivariate Logistic Regression Analysis of the Significant Predictors for Synchronous Gastric Cancer Liver Metastasis

Variable	Coefficient	SE	Wald χ^2	*P* Value	Odds Ratio	(95%CI)
HBsAg	1.047	0.472	4.920	0.027	2.849	(1.130-7.186)
CEA	1.315	0.307	18.391	<0.001	3.724	(2.042-6.793)
CA199	0.446	0.333	1.793	0.181	1.562	(0.813-3.000)
AFP	1.759	0.349	25.444	<0.001	5.806	(2.931-11.500)
AST	0.996	0.695	1.932	0.164	2.628	(0.673-10.264)
ALT	-0.956	0.794	1.450	0.229	0.384	(0.081-1.823)
GGT	1.695	0.399	18.056	<0.001	5.444	(2.492-11.895)
ALP	1.389	0.416	11.163	0.001	4.010	(1.776-9.058)
Histological classification	-1.312	0.882	2.213	0.137	0.269	(0.048-1.516)
Age	0.024	0.012	3.634	0.057	1.024	(0.999-1.049)
Constant	-4.210	1.213	12.045	0.001	0.015	

Abbreviations: CI, confidence interval; HBsAg, hepatitis B surface antigen; CEA, carcinoembryonic ntigen; CA199, carbohydrate antigen 199; AFP, alpha fetoprotein; AST, aspartate aminotransferase; ALT, alanine aminotransferase; GGT, γ-glutamyltransferase; ALP, alkaline phosphatase;SE, standard error.

Table 5 Liver Metastasis in Hepatitis B Surface Antigen-Positive Patients With or Without Hepatitis B e Antigen

Group	synGCLM+	synGCLM-	*P* Value
HBeAg+(n=5)	2(40.0%)	3(60.0%)	0.190
HBeAg-(n=55)	8(14.5%)	47(85.5%)	

Abbreviations: HBeAg, hepatitis B e antigen; synGCLM, synchronous gastric cancer liver metastasis.

Figures

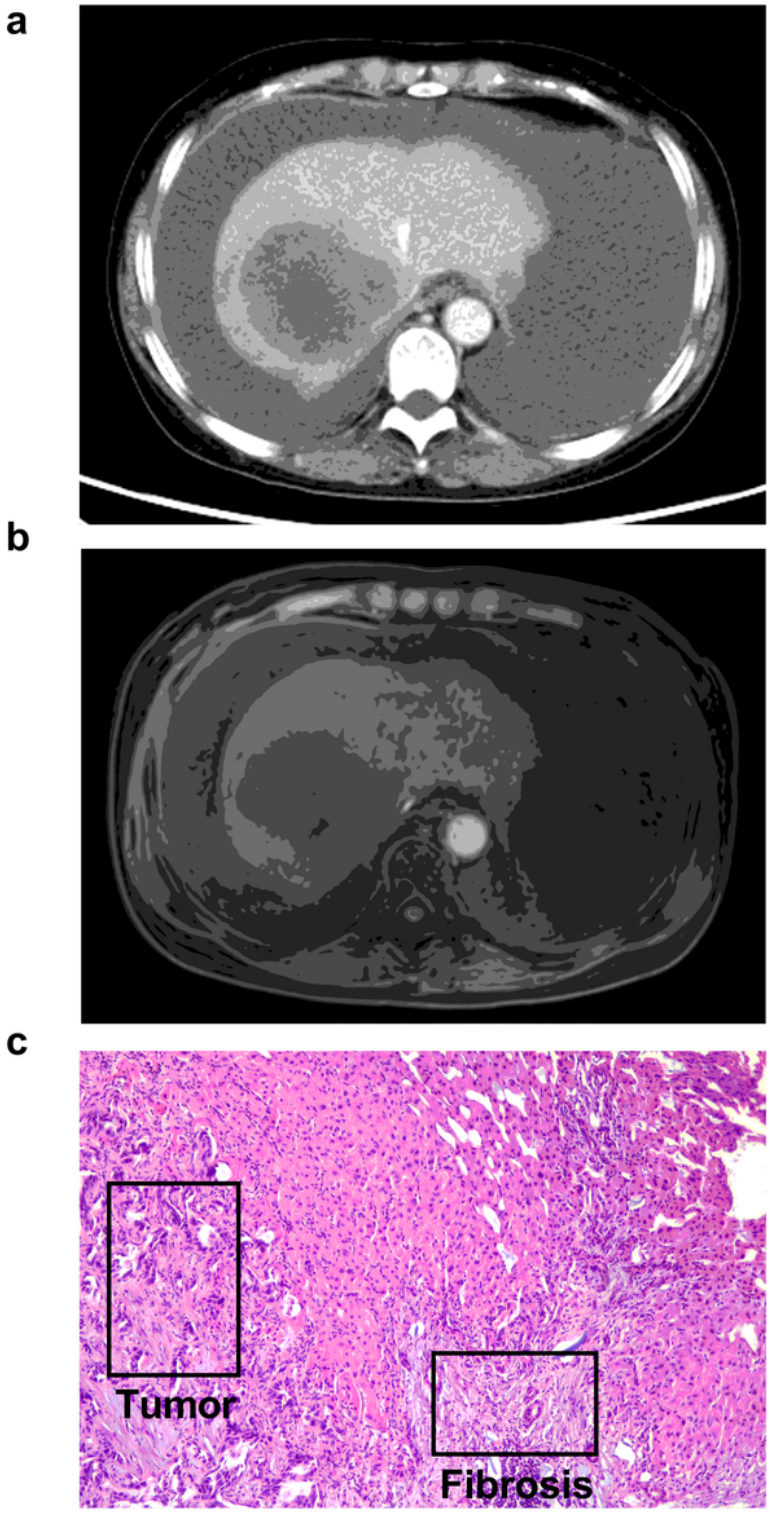

Figure 1

CT, MRI and histopathological images of HBV infection leading to liver metastasis in GC patients (a) The liver had a regular shape, and a mass of low density shadow with ring-like enhancement could be seen in the right lobe of the liver, which had an unclear boundary. A large amount of liquid density shadow was seen in the abdominal cavity. (b) Multiple mass and nodular abnormal signals were seen in the liver parenchyma with progressive ring-like enhancement, and the diffusion was obviously limited. Massive

ascites could also be seen around the liver. (c) Magnified 100 times, hematoxylin-eosin staining: Fibrotic changes and inflammatory cell infiltration were observed in tumor-adjacent tissues. There were punctate and focal necrosis of cancer cells, yperplasia of connective tissue in metastases. The liver metastasis was in accordance with the origin of gastric cancer.

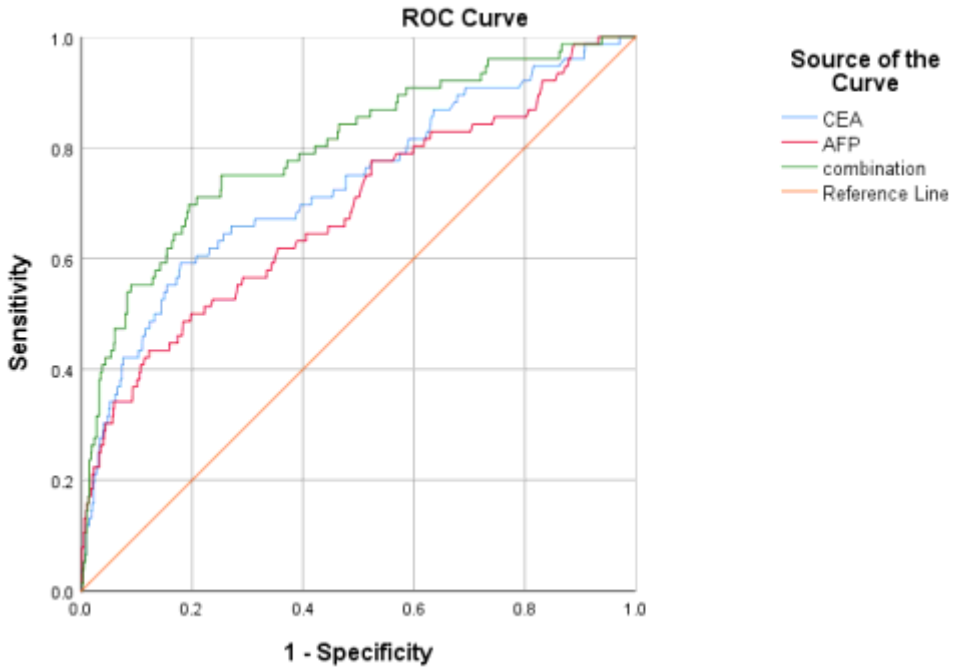

Figure 2

Receiver operating characteristic analysis for the prediction of synchronous liver metastasis in gastric cancer patients