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Mini Review

Diagnostic and Prognostic Value of Serum Biomarkers in Hepatocellular Carcinoma: Updated Review

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Abstract

Hepatocellular carcinoma (HCC) remains a major cause of mortality in patients with chronic liver disease worldwide. Early detection of HCC is critical to providing effective treatment and can have a significant impact on survival. Currently available serum tumor markers, such as alpha-fetoprotein (AFP), are characterized by low sensitivity in the detection of HCC. The development of highly sensitive and specific serum biomarkers for HCC may greatly enhance early detection rates and improve treatment success. Recent advances in proteomics and glyco-proteomics provided various types of novel tumor markers for HCC. While the clinical availability of these tumor markers is important, the molecular mechanisms underlying the production of tumor markers requires further clarification. This paper summarizes recent studies of specific biomarkers at early diagnosis or in monitoring metastasis or postoperative recurrence of HCC.

INTRODUCTION

Hepatocellular carcinoma (HCC) is the fifth most common tumor type and the third most common cause of cancer-related death worldwide. It carries a poor survival rate and has an increasing incidence worldwide. In most cases, HCC is diagnosed at a late stage. Therefore, the prognosis of patients with HCC is generally poor and has a less than 5% 5-year survival rate [1]. It is associated with multiple risk factors and is believed to arise from pre-neoplastic lesions, usually in the background of cirrhosis. However, the genetic and epigenetic events of hepatocarcinogenesis are relatively poorly understood.There has been marked progress in the treatment of HCC. However, effective treatments are limited to patients with less advanced HCC. The detection of HCC at an early stage is still a prerequisite for improved prognosis. Screening strategies including alpha-fetoprotein (AFP) and ultrasound (US) every 6 months in patients with liver cirrhosis have been recommended to detect HCC at earlier stages. AFP, however, is a marker with poor sensitivity and specificity and the ultrasound is highly dependent on the operator's experience [2,3]. Therefore, contrast-enhanced computerized tomography (CT) and magnetic resonance imaging (MRI) have superseded AFP and US, especially in early stages of HCC.

The most urgent needs are to find sensitive markers for early diagnosis or monitor postoperative recurrence and to give adequate treatment for HCC. The clinical value of serum AFP to detect early HCC has been questioned due to its low sensitivity and specificity. AFP, a 70-kD glycoprotein synthesized from the fetal yolk sac, liver, and intestines, has a half-life of 5-7 days. Total serum AFP level is a prognostic indicator of the response and survival of germ cell tumors [4]. However, when an AFP level is slightly elevated, it may be falsely elevated owing to nonneoplastic liver disease. The false negative rate with AFP level alone may be as high as 40% for patients with early stage HCC [5]. Even in patients with advanced HCC, the AFP levels may remain normal in 15~30% of the patients [5,6]. Other than AFP, several new serum biomarkers have been identified as useful HCC markers. Most widely studied ones among them are the circulating AFP isoform HCC specific (HS-AFP) AFP-L3, desgamma carboxy prothrombin (DCP), golgi protein 73 (GP-73), glypican-3 (GPC-3) and microRNA (miRNA) (Table 1). Other than AFP, these novel biomarkers have been found to improve the sensitivity, specificity, early detection, and prediction of prognosis. However, the overall results have been questioned [7-9]. Furthermore, recent developments in gene-expressing microarrays and proteomics promise even more potential diagnostic options.

Total AFP can be divided into three different glycoforms (L1, L2 and L3) according to their binding capacity for lens culinaris

Table 1: The biomarkers found to be useful in early detection and follow-up of HCC.

Alpha-fetoprotein (AFP)
Lens culinaris agglutinin reactive AFP (AFP-L3/HS-AFP)
Des-gamma carboxy prothrombin (DCP)
Protein induced by vitamin K absence or antagonist-II (PIVKA-II)
Glypican-3 (GPC-3)
Golgi protein-73 (GP-73)
Squamous cell carcinoma antigen-immunoglobulin M immune complexes (SCCA-IgM ICs)
Micro RNA (miRNA)
Tumor-associated glycoprotein 72 (TAG-72)
Alpha-l-fucosidase (AFU)
Hepatocyte growth factor (HGF)
Serum amyloid A (SAA)
Zinc-α2-glycoprotein (ZAG)

agglutinin (LCA) or their isoelectric point difference [10]. HS-AFP, as the LCA-bound fraction, is the major glycoform of AFP in HCC patients. Recent most studies have suggested that the percentage of HS-AFP may be a more specific marker than total AFP for early diagnosis and recurrence of HCC [11,12]. To increase the specificity of AFP, the AFP-L3 glycoform can be used as a measure of cancerous changes in the AFP composite carbohydrate moiety. The most frequently used cut-off value is 10% [13]. In a recent review, sensitivity of 18.8% and specificity of 99.4% has been reported for AFP-L3 [14]. Although AFP-L3 has high specificity, due to its low sensitivity, it is considered to be of limited use in screening. It is possible, however, to diagnose a marginally higher number of patients with advanced hepatocellular carcinoma. Results of pathological investigations conducted on patients who have undergone hepatic resection showed that those with infiltrative growth, capsular invasion, septum formation, portal vein infiltration, and hepatic vein infiltration were significantly more likely to have AFP-L3-positive (>10%) cancer [15]. From these studies we can conclude that AFP-L3, in particular its high sensitivity measurement, is extremely useful as an index of prognostication and for the degree of biological malignancy of hepatocellular carcinoma. Consequently, it is highly expected that AFP-L3 will become more popular worldwide. In Sassa et al., s recent study to evaluate the diagnostic efficacy of simultaneous measurements of high-sensitivity des-gamma-carboxy prothrombin (DCP) and AFP-L3 in small hepatocellular carcinoma (HCC), of 61 patients 44.3 % were positive for DCP and 23 % were positive for AFP-L3 [16]. There was no correlation between DCP and AFP-L3 %. 31.1% had positive H-DCP alone. 9.8% had positive AFP-L3 alone, and in 13.1% both markers were positive. There was a tendency for the AFP-L3 to be elevated in patients with moderately or poorly differentiated HCC and multiple HCC nodules, while the H-DCP showed no elevation related to the tumour type. The detection rate of small HCC was improved by combination assay with DCP and AFP-L3. These data indicate that the markers are complementary and useful for the diagnosis and evaluation of small HCC when measured simultaneously. In another study, Yuen and Lai tested the three most common markers (AFP, AFP-L3 and protein induced by vitamin K absence or antagonist-II, PIVKA-II) [17]. In their study, total AFP hah the sensitivity of 60% and specificity of 90% for the detection of HCC. Increase in the percentage of AFP-L3 over the total AFP (>10%) was very specific for small HCC. PIVKA-II was also shown to be more specific than total AFP in detecting HCC. AFP-L3 and PIVKA-II levels correlated well with tumour aggressiveness and prognosis. All three markers were found to be useful for monitoring treatment responsiveness and tumour recurrence. Since the levels of the three markers were independent of each other, combination of measurement of two or three markers has been suggested to increase the sensitivity and diagnostic accuracy.

The oncofetal antigen GPC3 is a glycosylphosphatidyl inositol-anchored membrane protein and has been shown to be present in sera from 40 to 50% of HCC patients, but was not detected in sera from patients with liver cirrhosis or chronic hepatitis, or in sera from healthy individuals [18]. In some other studies, the sensitivity and specificity of GPC3 in the diagnosis of HCC was found to be 77 and 96%, respectively [19]. On the strength of these results, GPC3 is a potential marker for HCC. Total positive rates of circulating GPC3 and its gene in combination with AFP could rise up to 94.3 % for HCC diagnosis [20]. The detecting GPC3 and GPC3 mRNA were found to be superior to AFP in sensitivity, specificity, positive predictive or negative predictive value, and accuracy for HCC. The detection of circulating GPC-3 or its gene transcription in HCC specificity was superior to serum AFP alone, with efficacious in HCC differentiating diagnosis or monitoring hematogenous metastasis. These data suggest that combining applications of GPC3 and AFP should rise up the HCC diagnostic sensitivity.

Golgi protein 73 (GP73) is a type II Golgi-specific membrane protein and is significantly elevated in various types of cancer. Results of recent studies have shown that the serum GP73 is significantly elevated in primary hepatic carcinoma [21,22]. In their study, Mao et al demonstrated that GP73 in the serum of patients with HCC infected by HBV was significantly higher

compared with HBV carriers, patients without hepatic diseases and healthy adults [23]. The sensitivity of diagnosis of HCC (76.9%) was markedly elevated compared with AFP (48.6%), suggesting GP73 is a novel and effective serum biomarker for the diagnosis of HCC. Additional investigations identified fucosylated GP73 (FC-GP73). Compared with total GP73, FC-GP73 improves the sensitivity and specificity of diagnosis of HCC from 65–90 to 90–100%, respectively. For GP73-negative or low levels, detection of FC-GP73 is a viable option [24]. Although the study for GP73 is optimistic, there are limitations that should be considered such as the fact that the correlation between GP73 and tumor size, stage, recurrence and prognosis should be extensively investigated. Specifically, the mechanism for GP73 and HCC development remains to be elucidated. Thus, role of GP73 in the clinic remains to be determined.

Squamous cell carcinoma antigen (SCCA), a serine protease inhibitor isolated from cervical carcinoma, is typically expressed in epithelial tumors and protects tumor cells from apoptosis. Guido et al found that the expression of SCCA in HCC (93%) and dysplastic nodule (100%) is significantly higher than the regenerative nodule (29%), suggesting that the expression of SCCA increased in the early stages of HCC formation [25]. The high sensitivity and low specificity seems to be complementary with AFP. Thus, SCCA can be accepted as a valuable supplement marker for the diagnosis of HCC. SCCA-IgM IC is a circulating immune complex composed of SCCA and IgM. It was undetectable in the sera of a healthy control population. However, in chronic hepatitis, cirrhosis and HCC, the detection rates of SCCA-IgM IC were 18, 26 and 70%, respectively. No correlation was identified with AFP level [26]. Furthermore, in patients with liver cirrhosis progressing towards HCC, SCCA-IgM IC was consistently increased and had higher sensitivity compared with AFP [27]. Therefore, SCCA-IgM IC may be a novel valuable serum marker for HCC. A combination of SCCA-IgM IC and AFP can thus improve the diagnostic rate.

Tumor-associated glycoprotein 72 (TAG-72) is a macro-molecular glycoprotein complex similar to mucin-1 (MUC-1). It is overexpressed in the majority of human adenocarcinomas and is rarely expressed in normal tissues. Recent studies found that the expression of TAG-72 is significantly elevated in HCC tissues compared with normal liver tissues [28]. Its increased expression may promote tumor invasion and metastasis. Furthermore, overexpression of TAG-72 is closely correlated with poor survival in patients with HCC [28,29]. Thus, TAG-72 is a potential prognostic marker for HCC, which has important clinical implications.

MicroRNAs are small non-coding RNAs that effectively block translation by promoting the degradation of target mRNAs or binding to complementary sequences. In recent years, the association between microRNAs and tumors has become a point of debate. MiR-500 (miRNA) is a potential candidate biomarker for HCC, as proven by Yamamoto et al, using a global miRNA expression profile in mouse liver development [30]. In other studies, based on miRNA microarray, miR-29 and miR-122 were shown to be downregulated in HCC cells, suggesting their role as a prognostic marker for HCC therapy [31,32]. In addition, plasma miR-21 level in patients with HCC has been shown to be significantly higher than that in patients with chronic hepatitis and healthy individuals. The analysis revealed the sensitivity and specificity to be 87.3 and 92%, respectively, differentiating HCC patients from healthy adults. Thus, miR-21 is also a promising biomarker of HCC [32]. Moreover, miR-34a is determined to be involved in oncogenesis and progression of HCC. Cui et al have investigated the prognostic value of tissue miR-34a expression in patients with HCC treated with RFA [33]. Patients with early-stage single-nodule HCC treated with RFA were included, and tissue expression of miR-34a were assessed by quantitative reverse-transcription polymerase chain reaction. Main clinical endpoints were overall and early recurrence. The expression of miR-34a was also an independent predictive factor for early recurrence Taken together, this study suggested that the expression of miR-34a in HCC biopsy specimens has an independent predictive value of early recurrence after RFA.

Other then these most commonly studied and proved novel biomarkers, many other markers such as alpha-l-fucosidase (AFU), hepatocyte growth factor (HGF), nervous growth factor (NGF), serum amyloid A (SAA), zinc-α2-glycoprotein (ZAG), and etc. have been shown to be effective in both early diagnosis of HCC and follow-up of treatment (Table) [34-37]. However, these biomarkers are also overexpressed in many other diseases and inflammatory conditions. Therefore, their specificity is relatively poor and new studies should be undertaken before clinical application. Moreover, misleading false diagnoses of HCC can be possible carrying the patient to a major surgery, even to liver transplantation [38].

In conclusion, large numbers of HCC markers exist in the clinical setting, however, most single indicators lack specificity of the tissues and organs. Furthermore, the single indicator results in the varying degrees of false positivity in certain benign diseases. Therefore, effective test strategies should be considered to improve the early diagnostic rate of HCC including the combined detection of several serum markers that can complement each other in order to improve the early diagnostic rate. Combined detection with AFP can significantly improve the ability of identification and diagnosis for HCC. Despite the large number of studies devoted to the immunohistochemistry of HCC, at the present time, the absolute positive and negative markers for HCC are still lacking, and even those characterized by very high sensitivity and specificity do not have an universal diagnostic usefulness. Additional studies are likely to yield novel markers and adopt more effective combined detection methods to improve the sensitivity and specificity of diagnosis of HCC, resulting in improved treatment and prognosis.

REFERENCES

1. Masuda T, Miyoshi E. Cancer biomarkers for hepatocellular carcinomas: from traditional markers to recent topics. Clin Chem Lab Med. 2011; 49: 959-966.

2. Spangenberg HC, Thimme R, Blum HE. Serum markers of hepatocellular carcinoma. Semin Liver Dis. 2006; 26: 385-390.

3. Bertino G, Ardiri A, Malaguarnera M, Malaguarnera G, Bertino N,

Calvagno GS. Hepatocellualar carcinoma serum markers. Semin Oncol. 2012; 39: 410-433.

4. Wang L, Yao M, Dong Z, Zhang Y, Yao D. Circulating specific biomarkers in diagnosis of hepatocellular carcinoma and its metastasis monitoring. Tumour Biol. 2014; 35: 9-20.

5. Yoshida S, Kurokohchi K, Arima K, Masaki T, Hosomi N, Funaki T, et al. Clinical significance of lens culinaris agglutinin-reactive fraction of serum alpha-fetoprotein in patients with hepatocellular carcinoma. Int J Oncol. 2002; 20: 305-309.

6. El-Serag HB, Kramer JR, Chen GJ, Duan Z, Richardson PA, Davila JA. Effectiveness of AFP and ultrasound tests on hepatocellular carcinoma mortality in HCV-infected patients in the USA. Gut. 2011; 60: 992-997.

7. Miyaaki H, Nakashima O, Kurogi M, Eguchi K, Kojiro M. Lens culinaris agglutinin-reactive alpha-fetoprotein and protein induced by vitamin K absence II are potential indicators of a poor prognosis: a histopathological study of surgically resected hepatocellular carcinoma. J Gastroenterol. 2007; 42: 962-968.

8. Debruyne EN, Delanghe JR. Diagnosing and monitoring hepatocellular carcinoma with alpha-fetoprotein: new aspects and applications. Clin Chim Acta. 2008; 395: 19-26.

9. Malaguarnera G, Giordano M, Paladina I, Berretta M, Cappellani A, Malaguarnera M. Serum markers of hepatocellular carcinoma. Dig Dis Sci. 2010; 55: 2744-2755.

10. Kobayashi T, Aikata H, Honda F, Nakano N, Nakamura Y, Hatooka M, et al. Preoperative Fluorine 18 Fluorodeoxyglucose Positron Emission Tomography/Computed Tomography for Prediction of Microvascular Invasion in Small Hepatocellular Carcinoma. J Comput Assist Tomogr. 2016; 10.

11. Zhang Z, Zhang Y, Wang Y, Xu L, Xu W. Alpha-fetoprotein-L3 and Golgi protein 73 may serve as candidate biomarkers for diagnosing alpha-fetoprotein-negative hepatocellular carcinoma. Onco Targets Ther. 2015; 9: 123-129.

12. Stefaniuk P, Cianciara J, Wiercinska-Drapalo A. Present and future possibilities for early diagnosis of hepatocellular carcinoma. World J Gastroenterol. 2010; 16: 418-424.

13. Shimizu K, Katoh H, Yamashita F, Tanaka M, Tanikawa K, Taketa K, et al. Comparison of carbohydrate structures of serum alpha-fetoprotein by sequential glycosidase digestion and lectin affinity electrophoresis. Clin Chim Acta. 1996; 254: 23-40.

14. Kudo M. Alpha-fetoprotein-L3: Useful or Useless for Hepatocellular Carcinoma?. Liver Cancer. 2013; 2: 151-152.

15. Tada T, Kumada T, Toyoda H, Kiriyama S, Sone Y, Tanikawa M, et al. Relationship between Lens culinaris agglutinin-reactive alpha-fetoprotein and pathologic features of hepatocellular carcinoma. Liver Int. 2005; 25: 848-853.

16. Sassa T, Kumada T, Nakano S, Uematsu T. Clinical utility of simultaneous measurement of serum high-sensitivity des-gamma-carboxy prothrombin and Lens culinaris agglutinin A-reactive alpha-fetoprotein in patients with small hepatocellular carcinoma. Eur J Gastroenterol Hepatol. 1999; 11: 1387-1392.

17. Yuen MF, Lai CL. Serological markers of liver cancer. Best Pract Res Clin Gastroenterol. 2005; 19: 91-99.

18. Iwama T, Uchida T, Sawada Y, Tsuchiya N, Sugai S, Fujinami N, et al. Vaccination with liposome-coupled glypican-3-derived epitope peptide stimulates cytotoxic T lymphocytes and inhibits GPC3-expressing tumor growth in mice. Biochem Biophys Res Commun. 2016; 469: 138-143.

19. Libbrecht L, Severi T, Cassiman D, Vander Borght S, Pirenne J, Nevens F, et al. Glypican-3 expression distinguishes small hepatocellular carcinomas from cirrhosis, dysplastic nodules, and focal nodular hyperplasia-like nodules. Am J Surg Pathol. 2006; 30:1405-1411.

20. Nassar A, Cohen C, Siddiqui MT. Utility of glypican-3 and survivin in differentiating hepatocellular carcinoma from benign and preneoplastic hepatic lesions and metastatic carcinomas in liver fine-needle aspiration biopsies. Diagn Cytopathol. 2009; 37: 629-635.

21. Sai W, Wang L, Zheng W, Yang J, Pan L, Cai Y, et al. Abnormal Expression of Golgi Protein 73 in Clinical Values and Their Role in HBV-Related Hepatocellular Carcinoma Diagnosis and Prognosis. Hepat Mon. 2015; 15: 32918.

22. Shan SG, Gao YT, Xu YJ, Huang Y, Zhang Q, Zhai DK, et al. Gradually increased Golgi protein 73 expression in the progression of benign liver diseases to precancerous lesions and hepatocellular carcinoma correlates with prognosis of patients. Hepatol Res. 2013; 43: 1199-1210.

23. Mao YL, Yang HY, Xu HF, Sang XT, Lu X, Yang ZY, Zhang JC, Zhong SX, Huang JF, Zhang HB. Significance of Golgi glycoprotein 73, a new tumor marker in diagnosis of hepatocellular carcinoma: a primary study. Zhonghua Yi Xue Za Zhi. 2008; 88: 948-951.

24. Mao Y, Yang H, Xu H, Lu X, Sang X, Du S, et al. Golgi protein 73 (GOLPH2) is a valuable serum marker for hepatocellular carcinoma. Gut. 2010; 59: 1687-1693.

25. Yan-Jie Z, Qiang J, Guan-Cheng L. Tumor markers for hepatocellular carcinoma. Mol Clin Oncol. 2013; 1: 593-598.

26. Beneduce L, Castaldi F, Marino M, Quarta S, Ruvoletto M, Benvegnù L, et al. Squamous cell carcinoma antigen-immunoglobulin M complexes as novel biomarkers for hepatocellular carcinoma. Cancer. 2005; 103: 2558-2565.

27. Pontisso P, Quarta S, Caberlotto C, Beneduce L, Marino M, Bernardinello E, et al. Progressive increase of SCCA-IgM immune complexes in cirrhotic patients is associated with development of hepatocellular carcinoma. Int J Cancer. 2006; 119: 735-740.

28. Milenic DE, Brady ED, Garmestani K, Albert PS, Abdulla A, Brechbiel MW. Improved efficacy of alpha-particle-targeted radiation therapy: dual targeting of human epidermal growth factor receptor-2 and tumor-associated glycoprotein 72. Cancer. 2010; 116: 1059-1066.

29. Mracek T, Stephens NA, Gao D, Bao Y, Ross JA, Ryden M, et al. Enhanced ZAG production by subcutaneous adipose tissue is linked to weight loss in gastrointestinal cancer patients. Br J Cancer. 2011; 104: 441-447.

30. Yamamoto Y, Kosaka N, Tanaka M, Koizumi F, Kanai Y, Mizutani T, et al. MicroRNA-500 as a potential diagnostic marker for hepatocellular carcinoma. Biomarkers. 2009; 14: 529-538.

31. Augello C, Vaira V, Caruso L, Destro A, Maggioni M, Park YN, et al. MicroRNA profiling of hepatocarcinogenesis identifies C19MC cluster as a novel prognostic biomarker in hepatocellular carcinoma. Liver Int. 2012; 32:772-782.

32. Wang WY, Zhang HF, Wang L, Ma YP, Gao F, Zhang SJ, et al. miR-21 expression predicts prognosis in hepatocellular carcinoma. Clin Res Hepatol Gastroenterol. 2014; 38: 715-719.

33. Cui X, Wu Y, Wang Z, Liu X, Wang S, Qin C. MicroRNA-34a expression is predictive of recurrence after radiofrequency ablation in early hepatocellular carcinoma. Tumour Biol. 2015; 36: 3887-3893.

34. Gan Y, Liang Q, Song X. Diagnostic value of alpha-L-fucosidase for hepatocellular carcinoma: a meta-analysis. Tumour Biol. 2014; 35: 3953-3960.

35. Mizuguchi T, Nagayama M, Meguro M, Shibata T, Kaji S, Nobuoka T,

et al. Prognostic impact of surgical complications and preoperative serum hepatocyte growth factor in hepatocellular carcinoma patients after initial hepatectomy. J Gastrointest Surg. 2009; 13: 325-333.

36. Huang Y, Li LZ, Zhang CZ, Yi C, Liu LL, Zhou X, et al. Decreased expression of zinc-alpha2-glycoprotein in hepatocellular carcinoma associates with poor prognosis. J Transl Med. 2012; 10: 106.

37. Ni XC, Yi Y, Fu YP, He HW, Cai XY, Wang JX, et al. Serum amyloid A is a novel prognostic biomarker in hepatocellular carcinoma. Asian Pac J Cancer Prev. 2014; 15: 10713-10718.

38. Unal E, Francis F, Aquino A, Xu R, Morgan G, Teperman L. Liver transplant for mixed capillary-cavernous hemangioma masquerading as hepatocellular carcinoma in a patient with hepatocellular carcinoma. Exp Clin Transplant. 2011; 9: 344-348.

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