Acta Medica Okayama

Volume 56, *Issue* 6 2002 *Article* 7

DECEMBER 2002

Evaluation of tumor markers for the detection of hepatocellular carcinoma in Yangon General Hospital, Myanmar.

Kazuhisa Taketa* Shigeru Okada† Ne Win‡

Naomi Khaing Than Hlaing** Khin Maung Win††

*Ibara City Hospital,

†Okayama University,

‡Department of Medical Research Lower Myanmar,

**Yangon General Hospital,

††Yangon General Hospital,

Evaluation of tumor markers for the detection of hepatocellular carcinoma in Yangon General Hospital, Myanmar.*

Kazuhisa Taketa, Shigeru Okada, Ne Win, Naomi Khaing Than Hlaing, and Khin Maung Win

Abstract

Levels of alpha-fetoprotein (AFP), its glycoforms AFP-L3 and AFP-P4, and proteins induced by vitamin K absence or antagonist-II (PIVKA-II) were determined in sera obtained from patients in Yangon General Hospital (20 with hepatocellular carcinoma (HCC), 29 with chronic liver diseases, including 3 with chronic hepatitis and 26 with cirrhosis of the liver, and 9 with other hepatobiliary diseases). Forty-five percent of the patients with HCC had serum AFP levels above 10,000 ng/ml, indicating that nearly half of the HCC patients were at an advanced stage of the disease. Thus, the AFP sensitivity was as high as 70% with 100% specificity for a cutoff level of 200 ng/ml. The sensitivity of AFP-L3 was 75% and a specificity 90% for a cutoff level of 15%. AFP-P4 showed a higher sensitivity of 80% and a similar specificity of 86% for a cutoff level of 12%. Combined evaluation of AFP-L3 and/or AFP-P4 increased the sensitivity to 90% with the same specificity of 86%, indicating that AFP-L3 and AFP-P4 are useful as adjuncts for diagnosis of HCC in the present population. PIVKA-II had a high sensitivity of 90%, although the specificity was lower than 45%, probably due to the low cutoff level, as some cholestatic patients were included in the control group.

KEYWORDS: apoptosis, spontaneously hypertensive rat, osteonecrosis of the femoral head

---

*PMID: 12685861 [PubMed - indexed for MEDLINE]

Acta Med. Okayama, 2002
Vol. 56, No. 6, pp. 317-320

Acta Medica Okayama
http://www.lib.okayama-u.ac.jp/www/acta/

Short Communication

Evaluation of Tumor Markers for the Detection of Hepatocellular Carcinoma in Yangon General Hospital, Myanmar

Kazuhisa Taketa[a]*, Shigeru Okada[b], Ne Win[c], Naomi Khaing Than Hlaing[d], and Khin Maung Win[d]

[a]Department of Internal Medicine, Ibara City Hospital, Ibara 715-0019, [b]Department of Pathological Research, Okayama University Graduate School of Medicine and Dentistry, Okayama 700-8558, Japan, [c]Department of Pathology, Department of Medical Research Lower Myanmar, Yangon, and [d]Liver Unit, Yangon General Hospital, Yangon, Myanmar

Levels of alpha-fetoprotein (AFP), its glycoforms AFP-L3 and AFP-P4, and proteins induced by vitamin K absence or antagonist-II (PIVKA-II) were determined in sera obtained from patients in Yangon General Hospital (20 with hepatocellular carcinoma (HCC), 29 with chronic liver diseases, including 3 with chronic hepatitis and 26 with cirrhosis of the liver, and 9 with other hepatobiliary diseases). Forty-five percent of the patients with HCC had serum AFP levels above 10,000 ng/ml, indicating that nearly half of the HCC patients were at an advanced stage of the disease. Thus, the AFP sensitivity was as high as 70% with 100% specificity for a cutoff level of 200 ng/ml. The sensitivity of AFP-L3 was 75% and a specificity 90% for a cutoff level of 15%. AFP-P4 showed a higher sensitivity of 80% and a similar specificity of 86% for a cutoff level of 12%. Combined evaluation of AFP-L3 and/or AFP-P4 increased the sensitivity to 90% with the same specificity of 86%, indicating that AFP-L3 and AFP-P4 are useful as adjuncts for diagnosis of HCC in the present population. PIVKA-II had a high sensitivity of 90%, although the specificity was lower than 45%, probably due to the low cutoff level, as some cholestatic patients were included in the control group.

Key words: alpha-fetoprotein (AFP), AFP-L3, AFP-P4, hepatocellular carcinoma, Yangon

The usefulness of tumor markers is determined by their sensitivity and specificity, which vary depending on the diagnostic level used as the gold standard in a disease [1], and also depending on the cutoff level of tumor markers. The sensitivities and specificities of alpha-fetoprotein (AFP), lentil lectin-reactive AFP-L3 [2], erythroagglutinating phytohemagglutinin-reactive AFP-P4 [2], and proteins induced by vitamin K absence or antagonist-II (PIVKA-II) [3] were studied to determine whether their cutoff levels are optimal for the diagnosis of patients with hepatocellular carcinoma (HCC) in Yangon General Hospital, Myanmar. The Liver Unit of this hospital is well-equipped, with the ability to use modern imaging techniques for HCC diagnosis. Because, however, HCC is detected at the first visit in many patients in this hospital, advanced cases of HCC are frequently encountered, as indicated by their high AFP levels.

Serum levels of HCC markers were determined in frozen sera from outpatients of and patients admitted to the Liver Unit, Yangon General Hospital. The patients studied consisted of 20 with HCC, 29 with chronic liver

Received May 10, 2002; accepted July 15, 2002.
*Corresponding author. Phone: +81-866-62-1133; Fax: +81-866-62-1275
E-mail: ibarahp@oka.urban.ne.jp (K. Taketa)

diseases (CLD), including 3 with chronic hepatitis (CH) and 26 with cirrhosis of the liver (COL), and 9 with other hepatobiliary diseases (mainly cholestasis resulting from acute hepatitis or obstruction of the biliary tract). Informed consent for determination of markers for HCC and hepatitis virus in their sera was obtained from the patients by their attending doctors.

AFP levels were determined by chemiluminescent immunoassay (CLIA) with Architect AFP (Dainabot Co., Ltd., Tokyo, Japan), AFP-L3 and AFP-P4 by affinity electrophoresis [2] with AFP Differentiation Kits L and P (Wako Pure Chemical Industries, Ltd., Osaka, Japan), and PIVKA-II by electro-chemiluminescence immunoassay with PICOLUMI PIVKA-II (Eisai Co., Ltd., Tokyo, Japan). HBsAg was analyzed by CLIA with Architect HBsAg (Dainabot), HBcAb by radioimmunoassay with HBcAb (recombinant) CORAB (Dainabot), and HCV Ab by immunoradiometric assay with Ortho HCV Ab IRMA Test III (Ortho-Clinical Diagnostics, Tokyo, Japan). The sensitivities of the tumor markers were calculated as the percentage of marker-positive cases among HCC patients, and the specificities were calculated as the percentage of marker-negative cases among patients with CLD (or other hepatobiliary diseases when specified) as a reference.

HCC was diagnosed by abdominal ultrasonography and computed tomography in most patients and by hepatic angiography and transarterial chemo-embolization with Lipiodol in patients with suspected early HCC to confirm or differentiate it from other space-occupying lesions. Tumor biopsy or ascitic fluid cytology was performed when necessary.

Of the 20 HCC patients, 18 were positive for HBsAg and/or HBcAb (hepatitis B virus (HBV)-related), 4 for HCV Ab (hepatitis C virus (HCV)-related), and 3 for both, while 1 was negative for both. In contrast, of the 29 CLD patients, 21 were related to HBV, 14 to HCV, and 10 to both, while 4 were negative for both. Of the 9 patients with other hepatobiliary diseases, 3 were related to HBV and the remainder were negative for both HBV and HCV. There were no apparent differences in the serum levels of AFP, AFP-L3, AFP-P4, and PIVKA-II between HBV-related HCC and HCV-related HCC.

Actual values for the HCC markers are shown in Fig. 1. Forty-five percent of the patients with HCC had AFP levels above 10,000 ng/ml, indicating that nearly half were at advanced stages of the disease. In our previous study [2], 47% of the patients with HCC showed AFP levels greater than 10,000 ng/ml. Thus, the populations

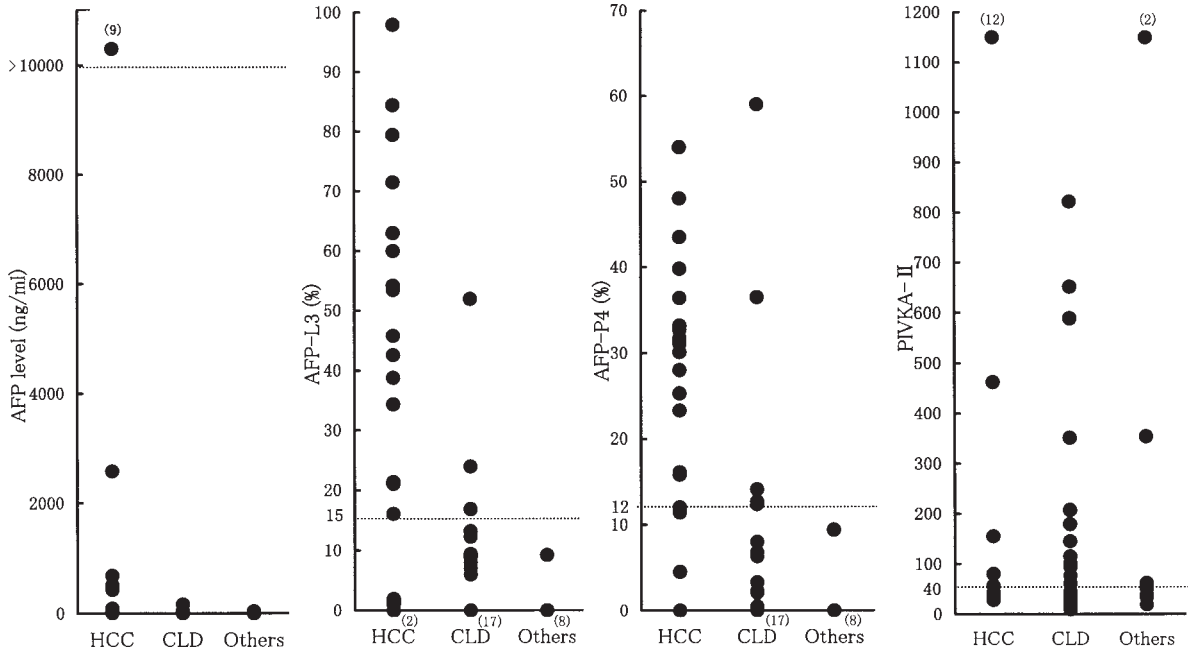

Fig. 1 Serum levels of HCC markers in HCC, CLD, and other hepatobiliary diseases. Horizontal dotted lines indicate cutoff levels of HCC markers. The numbers in parentheses indicate the number of overlapping closed circles.

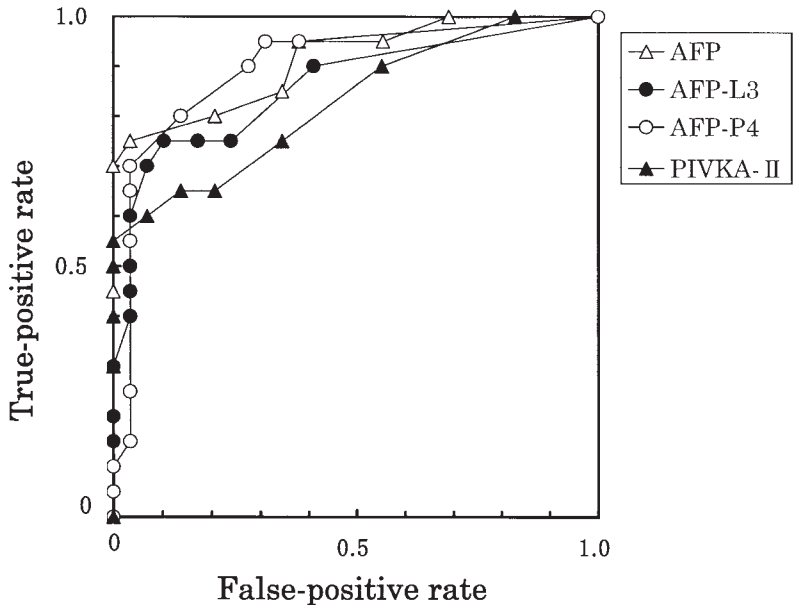

Fig. 2 ROC curves for AFP, AFP-L3, AFP-P4, and PIVKA-II.

of these two studies were similar with respect to the distribution of HCC staging.

The sensitivities and specificities of the HCC markers are summarized in Fig. 2 as receiver-operating characteristic (ROC) curves. With a convenient cutoff level of 200 ng/ml for AFP, the sensitivity was 70% with a specificity of 100%, indicating that AFP is a sensitive and specific marker of advanced HCC. When the cutoff level was lowered to 10 ng/ml, which is the cutoff level for healthy subjects [3], the sensitivity increased to 95 %, although the specificity decreased to 66% and there was a false-positive rate of 11% among patients with other hepatobiliary diseases (see Fig. 1).

In contrast, AFP-L3 had a sensitivity of 75% at a similarly high specificity of 90% for a cutoff level of 15 % and AFP-P4 had an even higher sensitivity of 80% at a slightly reduced specificity of 86% for a cutoff level of 12%. In combined evaluation of AFP-L3 and/or AFP-P4, the sensitivity increased to 90% at the same specificity of 86%. The increase in sensitivity was due to the fact that AFP-L3 and AFP-P4 were expressed independently, representing tumor heterogeneity [2]. These results indicate that AFP-L3 and AFP-P4 can be useful adjuncts for detection of HCC in a population of patients such as those in Yangon General Hospital. The slightly lower specificity of AFP-P4 compared with AFP-L3 in this study may have resulted from the use of a cutoff level of 15% for AFP-P4 + 5 instead of the 12 % level used for AFP-P4 in 6 patients with poor electrophoretic resolution of AFP-P4 and AFP-P5. AFP-P5 is less specific for HCC than AFP-P4, as can readily be seen when comparing the results obtained with AFP-P4 alone [2] with those obtained with AFP-P4 + AFP-P5 [6], where not only unresolved AFP-P4 + 5, but also resolved AFP-P4 and AFP-P5, were combined. There were no false-positives among patients with other hepatobiliary diseases for AFP-L3 and AFP-P4 (see Fig. 1). These results are comparable to those of our previous study [2], which was carried out with nearly the same proportion of patients with advanced HCC except that the AFP-L3 and AFP-P4 specificities were slightly lower. This lower specificity was probably due to the CLD patients in our previous study having been followed-up for one year to exclude the patients with early HCC that could not be detected by imaging, while the patients in the present study were not followed-up for such a long period of time.

PIVKA-II had a similarly high sensitivity of 90%, although the specificity was low, 45% for CLD and 33 % for other hepatobiliary diseases, when the cutoff level was 40 mAU/ml [4]. When a higher cutoff level of 100 mAU/ml was employed, which was previously used as the cutoff level for PIVKA-II [5], the results for PIVKA-II showed a reasonably high sensitivity of 70%

as well as a higher specificity of 68% without changing the results in patients with other hepatobiliary diseases. The low specificity of PIVKA-II, particularly in cholestatic diseases, at the currently used cutoff level of 40 mAU/ml suggests that vitamin K deficiency occurs under such conditions. In addition, it suggests that the current cutoff level is set too low to detect smaller nodules of HCC.

Acknowledgements. We wish to thank Wako Pure Chemical Industries, Ltd., Mitsubishi Kagaku Biochemical Laboratories, Inc. and Eisai Co., Ltd. for their active cooperation in this study.

References

1. Taketa K and Ikeda S: Meta-analysis of cutoff levels of lentil lectin-reactive AFP-L3 and erythroagglutinating phytohemagglutinin-reactive AFP-P4 for diagnosis of hepatocellular carcinoma. Ibara City Hosp Med Bull (2001) **2**, 5-12.
2. Taketa K, Sekiya C, Namiki M, Akamatsu K, Ohota Y, Endo Y and Kosaka K: Lectin-reactive profiles of alpha-fetoprotein characterizing hepatocellular carcinoma and related conditions. Gastroenterology (1990) **99**, 508-518.
3. Matsui H, Rimal N, Kamakura K, Uesugi S, Yamamoto H, Ikeda S and Taketa K: Serum α-fetoprotein levels in healthy Japanese adults. Acta Med Okayama (1998) **52**, 149-154.
4. Tanaka Y, Kashiwagi T, Tsutsumi H, Nagasawa M, Toyama T, Ozaki S, Naito M, Ishibashi K and Azuma M: Sensitive measurement of serum abnormal prothrombin (PIVKA-II) as a marker of hepatocellular carcinoma. Hepato-gastroenterology (1999) **46**, 2464-2468.
5. Fujiyama S, Izuno K, Yamasaki K, Sato T and Taketa K: Determination of optimum cutoff levels of plasma des-γ-carboxy prothrombin and serum α-fetoprotein for the diagnosis of hepatocellular carcinoma using receiver operating characteristic curves. Tumour Biol (1992) **13**, 316-323.
6. Taketa K, Endo Y, Sekiya C, Tanikawa K, Koji T, Taga H, Satomura S, Matsuura S, Kawai T and Hirai H: A collaborative study for the evaluation of lectin-reactive α-fetoproteins in early detection of hepatocellular carcinoma. Cancer Res (1993) **53**, 5419-5423.