

# Simplified HCC-ART score for highly sensitive detection of small-sized and early-stage hepatocellular carcinoma in the widely used Okuda, CLIP, and BCLC staging systems

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## Abstract

**Background** Small-sized HCC can be effectively cured by surgery with good clinical outcomes. A highly sensitive HCC  $\alpha$ -fetoprotein routine test (HCC-ART) for HCC diagnosis as well as a simplified form of the HCC-ART were reported in the British Journal of Cancer. Here, we verified and studied the applicability of the HCC-ART to the detection of early-stage HCC.

**Methods** 341 cirrhotic patients and 318 HCC patients were included in this study. For each, the HCC-ART score was calculated, and then the sensitivity, specificity, and results of an ROC curve analysis were compared between the HCC-ART and AFP when these biomarkers were used to detect small-sized HCC.

**Results** Different HCC-ART cutoffs were set for the detection of different tumor sizes. The HCC-ART (AUC = 0.871, 70% sensitivity, 97% specificity) and the simplified HCC-ART (AUC = 0.934, 82% sensitivity, 100% specificity) were found to have high predictive power when attempting to separate cirrhotic patients from those with small-sized HCC. The simplified HCC-ART score was superior to AFP for determining stages according to the early Okuda (0.950 AUC, 84% sensitivity, 99% specificity),

CLIP (0.945 AUC, 84% sensitivity, 99% specificity), and BCLC (1.000 AUC, 100% sensitivity, 99% specificity) staging systems. The simplified HCC-ART score was more strongly correlated than AFP and other staging systems with HCC tumor size ( $P < 0.0001$ ;  $r = 0.8$ ).

**Conclusion** The HCC-ART is superior to AFP for diagnosing early-stage HCC. Due to its advantages of minimal variability and a wide continuous scale for assessing HCC severity, the simplified HCC-ART has the potential to be more widely used than the original HCC-ART.

**Keywords** Hepatocellular carcinoma · Early stage · Diagnosis · Blood biomarkers · HCC-ART score

## Introduction

Liver cancer is generally associated with less-developed regions. Indeed, 83% of the estimated 782,000 new liver cancer cases that occurred worldwide in 2012 were in less-developed regions [1]. Early diagnosis is important for successful treatment by means of ablation, resection, and/or transplant [2]. While effective HCC treatment is usually curative surgery, about 80% of patients are inoperable at presentation and die early due to late diagnosis [3]. Thus, identifying effective screening strategies to detect early-stage HCC is of the utmost importance as it could result in more effective treatment and extend patient survival [4].

Although  $\alpha$ -fetoprotein (AFP) is routinely used for screening and has been widely applied in HCC surveillance programs, it often gives normal or indeterminate results in early cancer cases. AFP is a low-sensitivity biomarker that is normal in up to 50% of patients with HCC, particularly during the early stages of the disease ( $HCC \leq 3$  cm) [5]. Other modalities such as ultrasonography (US) and

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conventional tumor markers such as  $\alpha$ -fetoprotein-L3 (AFP-L3) and des- $\gamma$ -carboxyprothrombin (DCP) are also widely used and important for HCC detection in clinical settings [6]. However, none of these modalities provides an entirely satisfactory means of detecting HCC in its early stages [7].

Recently, a clinically useful scoring system, the HCC  $\alpha$ -fetoprotein routine test (HCC-ART), was developed for the diagnosis of patients with HCC [8]. The area under the curve, sensitivity, specificity, and accuracy of the HCC-ART for differentiating HCC from non-malignant liver cirrhosis were found to be 0.99, 97, 96, and 96%, respectively. Moreover, the HCC-ART showed absolute sensitivity and specificity (100%) when discriminating patients with HCC from those with liver fibrosis and those without fibrosis. Subsequently, a simplified version of the HCC-ART was devised in which a numerical constant and coefficients were removed from the original HCC-ART, and this simplified HCC-ART was observed to yield similar promising results to the original HCC-ART.

Because there is an urgent need to identify novel biomarkers that can be used to detect early HCC, in the large-scale study reported in the present paper, we employed HCC-ART and its simplified form to detect HCC patients with tumors of various sizes, ranging from small ( $\leq 2$  cm) to very large ( $\geq 10$  cm), in order to evaluate the clinical applicability of these tests to the early diagnosis and differentiation of small-sized HCC from liver cirrhosis. The widely used Okuda, CLIP, and BCLC systems were used as standard HCC staging systems, and the HCC-ART score was identified as a potential biomarker for early-stage HCC.

## Patients and methods

### Patient selection

659 consecutive patients (318 patients with HCC and 341 with liver cirrhosis) who were admitted to the Tropical Medicine Unit (Mansoura University Hospitals, Mansoura, Egypt) were enrolled in this study. All of the HCC patients had chronic hepatitis C or liver cirrhosis as the underlying liver disease. The diagnosis of HCC in those patients was made according to the American Association for the Study of Liver Diseases (AASLD) practice guidelines [9]. The tumors were detected by abdominal ultrasound (US) studies and/or AFP assays ( $>400$  U/L). Each focal lesion detected was further evaluated by multiphase spiral computed tomography (CT) or contrast-enhanced magnetic resonance imaging (MRI). The presence of enhancement in the arterial phase was considered diagnostic for HCC if the lesion was larger than 2 cm. The final diagnosis was,

whenever possible, confirmed by histopathologic analysis on US-assisted fine-needle biopsy. Informed consent was obtained from each patient and the study protocols followed the ethical guidelines of the 1975 Declaration of Helsinki.

### Differentiation of HCC stage

The tumor size was defined by contrast CT or MRI scan. HCC patients were divided according to tumor size into four groups: small tumor (size  $\leq 2$  cm,  $n = 25$ ); medium tumor (size 3–5 cm,  $n = 130$ ); large tumor (size 6–9 cm,  $n = 97$ ); and very large tumor (size  $\geq 10$  cm,  $n = 66$ ). The Okuda classification [10] is widely applied in HCC patients. It includes parameters related to liver functional status (albumin, ascites, bilirubin) and to the tumor stage (proportion of the area of the liver involved). All HCC patients were divided into three groups: early stage (0 points,  $n = 115$ ); intermediate stage (1 or 2 points,  $n = 169$ ); and advanced stage (3 or 4 points,  $n = 34$ ). The Cancer of the Liver Italian Program (CLIP) score [11] was also determined. This score combines four variables and provides a seven-stage classification system. The HCC patients studied were divided into three groups: early HCC (stages 0–1,  $n = 100$ ); intermediate HCC (stages 2–3,  $n = 175$ ); and advanced HCC (stages  $\geq 4$ ,  $n = 43$ ). Moreover, tumor stage was defined according to the Barcelona Clinic Liver Cancer (BCLC) staging system as either early (stage 0-A,  $n = 76$ ), intermediate (stage B,  $n = 86$ ), advanced (stage C,  $n = 76$ ), or end-stage (stage D,  $n = 80$ ).

### Sample collection and laboratory tests

Fasting blood samples were collected from all patients and divided into two parts; one was collected without an anticoagulant and the second was drawn into KEDTA tubes for a complete blood count. Sera were separated and tested fresh in liver function tests using an automated biochemistry analyzer (model A15, Biosystems S.A., Barcelona, Spain). The AFP level was determined by chemiluminescence with an Immulite AFP (1000) kit (Diagnostic Products Corporation, Los Angeles, CA, USA). In addition, serologic tests were carried out for HCV using commercial ELISA kits (ETI-AB-HCVK-3 kit, Sorine Biomedica, Suluggia, Italy). Complete blood counts were performed on a KX-21 automated hematology analyzer (Sysmex Corporation, Kobe, Japan).

### Statistical analysis

All statistical analyses were carried out using the Statistical Package for the Social Sciences (SPSS, SPSS Inc., Chicago, IL, USA) software, version 15.0, running in

**Table 1** Clinical characteristics of the patient groups investigated in the study, including liver cirrhosis and HCC tumor size stages

Variable	Liver cirrhosis	HCC	Differentiation of tumor size (cm)				P value
			Small ( $\leq 2$ )	Medium (3–5)	Large (6–9)	Very large ( $\geq 10$ )	
Number	341	318	25	130	97	66	–
Age (years)	49.1 $\pm$ 9.3	60 $\pm$ 10	58.2 $\pm$ 10.3	60.1 $\pm$ 9.8	59.7 $\pm$ 9.8	58.4 $\pm$ 9.4	<0.0001
AST (U/L)	69.4 $\pm$ 40.4	94.5 $\pm$ 100.3	97.2 $\pm$ 93.9	76.3 $\pm$ 48.9	102.4 $\pm$ 68.4	108.3 $\pm$ 161.3	<0.0001
ALT (U/L)	70.0 $\pm$ 50.7	61 $\pm$ 67	71.2 $\pm$ 94.9	56.1 $\pm$ 36.9	71.6 $\pm$ 62.5	67.5 $\pm$ 61.5	0.28
AAR	1.18 $\pm$ 0.55	1.65 $\pm$ 1.09	1.48 $\pm$ 0.46	1.50 $\pm$ 0.72	1.65 $\pm$ 0.81	1.87 $\pm$ 1.14	<0.0001
ALP (U/L)	98.9 $\pm$ 52.9	242 $\pm$ 271	154.9 $\pm$ 84.7	172 $\pm$ 124	185 $\pm$ 204	270 $\pm$ 288	<0.0001
Albumin (g/L)	39.1 $\pm$ 6.3	30.4 $\pm$ 7.4	34.7 $\pm$ 4.7	32.7 $\pm$ 5.3	33.6 $\pm$ 5.1	33.9 $\pm$ 5.7	<0.0001
Plt ( $\times 10^{-9}/L$ )	95.5 $\pm$ 49.6	185.3 $\pm$ 102.7	188.8 $\pm$ 138.3	141.1 $\pm$ 106.5	162.3 $\pm$ 97.8	224.3 $\pm$ 128.2	<0.0001
Ascites (no/yes)	321/20	246/72	20/5	93/37	79/18	54/12	–
AFP (U/L)	18.5 $\pm$ 34.8	9672 $\pm$ 60,273	469.2 $\pm$ 1146.4	5313.8 $\pm$ 25,569.1	4181.7 $\pm$ 9717.1	10,698.8 $\pm$ 15,852.3	<0.0001
Log AFP (U/L)	1.0 $\pm$ 0.4	2.3 $\pm$ 1.39	1.4 $\pm$ 1.0	2.1 $\pm$ 1.3	2.3 $\pm$ 1.3	2.9 $\pm$ 1.4	<0.0001

AAR AST/ALT ratio, ALP alkaline phosphatase, ALT alanine aminotransferase, AST aspartate aminotransferase, Plt platelet count, AFP  $\alpha$ -fetoprotein

ANOVA test or Mann–Whitney *U* test was used.  $P > 0.05$  is considered nonsignificant;  $P < 0.05$  is considered significant

Microsoft Windows XP, as well as the GraphPad Prism package; v.5.0 (GraphPad Software, San Diego, CA, USA). Continuous variables were expressed as the mean  $\pm$  SD or SEM, whereas categorical variables were expressed as numbers. ANOVA was used in cases where the Kolmogorov–Smirnov test results were not significant, while the Mann–Whitney *U* test was used in cases with nonparametric variables. The main endpoint was the identification of patients with hepatocellular carcinoma vs. liver cirrhosis. In secondary analyses, the accuracies of HCC-ART and AFP at discriminating HCC patients according to tumor size (small, medium, and large) were evaluated using the area under the ROC curve. The third step was to evaluate the predictive power of HCC-ART for the diagnosis of early-stage HCC in the Okuda, CLIP, and BCLC staging systems. The best cutoff values for the optimal prediction of HCC were determined from the ROC curves. In a fourth step, the correlations of HCC-ART and AFP with tumor size and different HCC classification systems were evaluated by Pearson's correlation coefficient. A  $P$  value of  $<0.05$  was considered statistically significant. The sensitivities and specificities of the HCC-ART and AFP were derived from a  $2 \times 2$  contingency table.

## Results

### Clinicopathological profiles of the patients

The clinicopathological findings (age, aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), albumin, platelet count, presence of ascites, and tumor size) were evaluated (Table 1). The HCC

patients were older than the cirrhotic patients. Univariate analysis of all tested variables except for ALT revealed that there was a significant difference ( $P < 0.05$ ) between the HCC and liver cirrhosis patients. The original and simplified HCC-ART [8] indices for discriminating patients with HCC were calculated using the following formulae: HCC-ART score =  $[2.17 + [\log(\text{AFP}-1) \times 10 \times 0.117] + \text{AST}/\text{ALT ratio} \times 0.025 + \text{age} \times 0.012 + \text{alkaline phosphatase (U/L)} \times 0.001] - [\text{albumin (g/L)} \times 0.015]$ ; simplified HCC-ART =  $[\text{age (years)} \times \log \text{AFP (U L}^{-1}) \text{AAR} \times \text{ALP (U L}^{-1})]/[\text{Alb (g}^{-1})]$ .

### HCC-ART is superior to serum AFP for the detection of small-sized HCCs

We compared HCC-ART with AFP in terms of their ability to differentiate different tumor sizes. The values of both HCC-ART and its simplified form gradually increased as the tumor size expanded from small ( $\leq 2$  cm) to very large tumors ( $\geq 10$  cm) ( $P = 0.001$  for HCC-ART,  $P = 0.01$  for simplified HCC-ART). In contrast, AFP levels fluctuated in a nonsignificant manner as the tumor increased ( $P = 0.10$ ). HCC-ART at a cutoff level of  $\geq 2.5$  detected small HCCs ( $\leq 2$  cm) with a sensitivity of 70%. At the same cutoff value, HCC-ART has a specificity of 97% for separating non-neoplastic liver cirrhotic subjects from those with small HCCs. Notably, the sensitivity increased from 70% for small HCCs to 94% for medium-sized HCCs, 90% for large HCCs, and 93% for very large HCCs (Table 2).

Interestingly, the simplified HCC-ART gave superior results to the original HCC-ART when a cutoff of  $\geq 280$  was used to differentiate non-neoplastic liver cirrhotic subjects from those with small tumors (Table 2). In the same

**Table 2** Diagnostic performance of HCC-ART and of AFP in discriminating HCC patients according to tumor size

Index	Cutoff	AUC	Sensitivity (%)	Specificity (%)
To detect very large HCC <sup>a</sup>				
HCC-ART	2.8	0.935	93	87
Simplified HCC-ART	560	0.945	93	83
AFP	400 U/L	0.833	70	99
To detect large HCC <sup>b</sup>				
HCC-ART	2.65	0.945	90	88
Simplified HCC-ART	360	0.946	93	84
AFP	400 U/L	0.790	53	99
To detect medium-sized HCC <sup>c</sup>				
HCC-ART	2.55	0.939	94	97
Simplified HCC-ART	320	0.945	97	97
AFP	400 U/L	0.819	33	100
To detect small HCC <sup>d</sup>				
HCC-ART	2.5	0.871	70	97
Simplified HCC-ART	280	0.934	82	100
AFP	400 U/L	0.560	12	100

AUC area under the curve

<sup>a</sup> Very large HCC ( $\geq 10$  cm) vs. cirrhosis/small/medium-sized/large HCC combined

<sup>b</sup> Large HCC (6–9 cm) vs. cirrhosis/small/medium-sized HCC combined

<sup>c</sup> Medium-sized HCC (3–5 cm) vs. cirrhosis/small HCC combined

<sup>d</sup> Small HCC ( $\leq 2$  cm) vs. cirrhosis

cohort of patients, AFP was less sensitive (12%) for detecting small HCC tumors (cutoff  $\geq 400$  U/L), and its expression increased with increasing tumor size. When we used a cutoff of  $\geq 200$  U/L, the sensitivity rose to 18% (not significantly different from the sensitivity obtained with a cutoff of  $\geq 400$  U/L). ROC analysis also showed that HCC-ART (AUC: 0.871) and its simplified form (AUC: 0.934) were superior to serum AFP (AUC: 0.560) for separating non-neoplastic individuals from those with small HCCs (Table 2). These results are consistent with previous reports that AFP is insensitive to small HCCs [8].

### Predictive power of HCC-ART in the diagnosis of early-stage HCC according to common staging systems

As a consequence of these favorable characteristics, we further compared the diagnostic performance of HCC-ART with that of serum AFP for the diagnosis of early-stage HCC according to three commonly employed staging systems.

### Okuda

The Okuda score is used as a standard classification of HCC. In contrast to AFP, the HCC-ART score (mean  $\pm$  SEM) significantly increased from stage 1 ( $2.7 \pm 0.01$ ) to stage 3 ( $3.3 \pm 0.2$ ). Similarly, the simplified HCC-ART score gradually increased ( $P = 0.04$ ) with Okuda stage progression (Fig. 1a–c), although the range of simplified HCC-ART scores was wider than the corresponding range of scores for the original HCC-ART. Sensitivities to Okuda early-stage tumors were 90, 84, 42, and 52 for HCC-ART, simplified HCC-ART, AFP ( $\geq 400$  U/L), and AFP ( $\geq 200$  U/L), respectively.

### CLIP (Cancer of the Liver Italian Program score)

When CLIP was used as the standard score to define different HCC stages, HCC-ART and its simplified form increased ( $P < 0.0001$  for HCC-ART and  $P = 0.02$  for simplified HCC-ART) with CLIP score progression (from 0–1 to  $\geq 4$ ). In contrast, AFP levels fluctuated with CLIP stage progression in a nonsignificant manner ( $P = 0.10$ ) (Fig. 1d–f). Sensitivities to early-stage CLIP early were 93, 84, 10, and 11 for HCC-ART, simplified HCC-ART, AFP ( $\geq 400$  U/L), and AFP ( $\geq 200$  U/L), respectively.

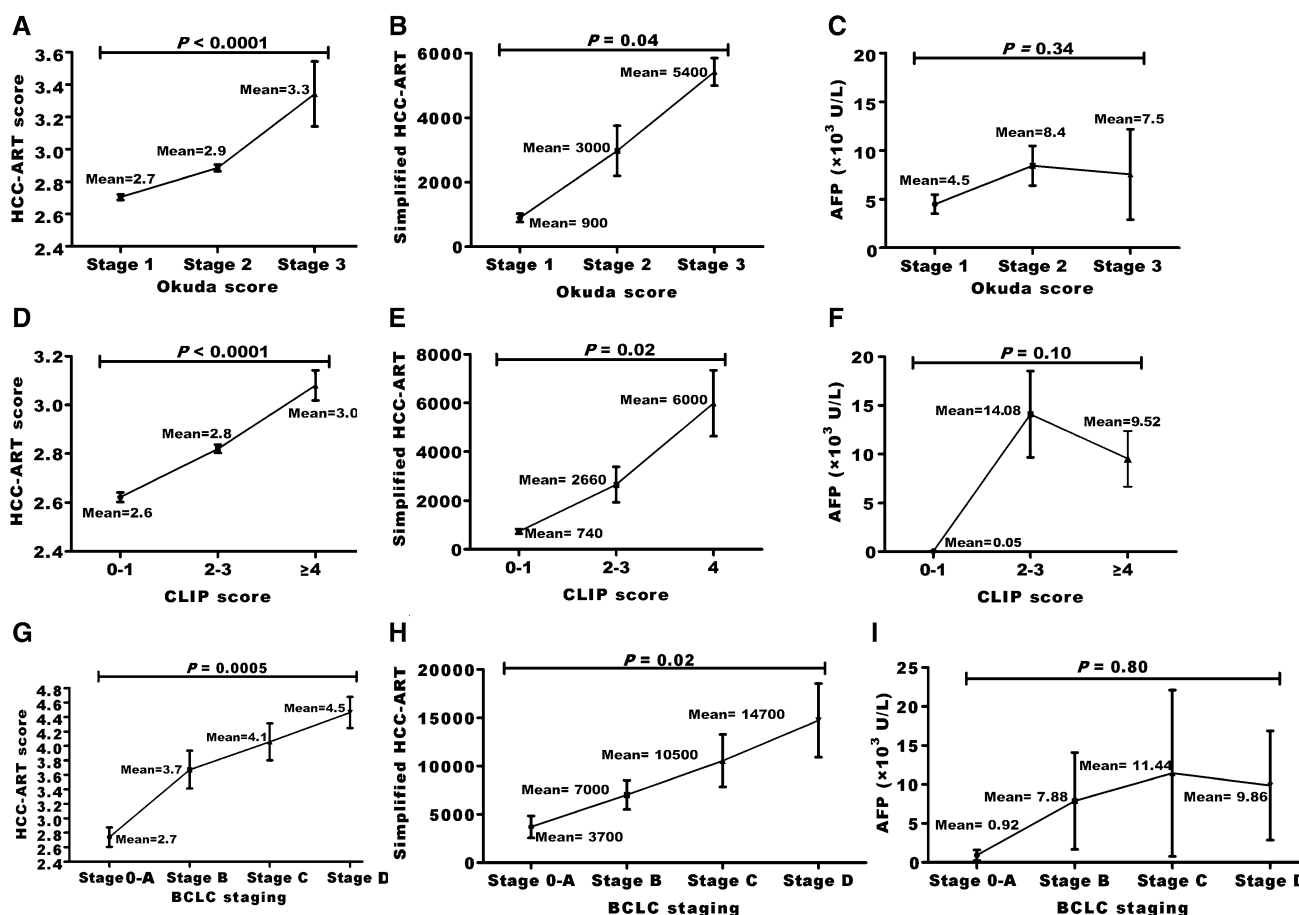
### BCLC (Barcelona Clinic Liver Cancer)

Furthermore, in contrast to AFP, HCC-ART and its simplified form were able to distinguish with high statistical significance between BCLC stages ranging from early to end-stage (Fig. 1g–i). Sensitivities to early-stage BCLC were 85, 100, 8, and 8 for HCC-ART, simplified HCC-ART, AFP ( $\geq 400$  U/L), and AFP ( $\geq 200$  U/L), respectively (Table 3).

Overall, HCC-ART at a cutoff of  $\geq 2.5$  and simplified HCC-ART at a cutoff of  $\geq 280$  appeared to be superior, more sensitive markers than serum AFP for detecting different HCC stages (Table 3). Likewise, the diagnostic value of these formulae was high; there was a large area under the ROC curve for distinguishing early-stage tumors according to the Okuda, CLIP, and BCLC schemes (Table 3). Increased HCC-ART ( $r = 0.5$ ,  $P < 0.0001$ ) and simplified HCC-ART ( $r = 0.8$ ,  $P < 0.0001$ ) values were found to be correlated with HCC tumor size significantly better than the Okuda ( $r = 0.2$ ,  $P = 0.03$ ), CLIP ( $r = 0.2$ ,  $P = 0.02$ ), BCLC ( $r = 0.2$ ,  $P = 0.03$ ), and AFP ( $r = 0.1$ ,  $P = 0.03$ ) (Table 4).

### Discussion

No treatment appears to be capable of prolonging survival in patients with large HCC tumors [12]. The conventional



**Fig. 1** Distribution of HCC-ART scores, simplified HCC-ART scores, and AFP values in different common HCC staging systems. In contrast to AFP, HCC-ART and simplified HCC-ART scores (mean  $\pm$  SEM) increased significantly with the Okuda stage (a HCC-ART, b simplified HCC-ART, c AFP), CLIP stage (d HCC-ART, e simplified HCC-ART, f AFP), and BCLC stage (g HCC-ART, h sim-

plified HCC-ART, i AFP) of the HCC tumor. AFP values fluctuated with HCC stage progression in a nonsignificant manner. One-way ANOVA was used to check differences between groups for statistical significance.  $P > 0.05$  was considered to indicate a nonsignificant and  $P < 0.05$  to indicate a significant difference

biomarker AFP, its glycosylated isoforms, and other modalities are widely used to detect HCC [5, 6]. However, their accuracy for detecting early-stage HCCs is poor [7]. So far, no reliable biomarker for detecting small HCCs has been reported; thus, novel biomarkers are urgently needed for these tumors.

Recently, an exciting study suggested that the use of a novel predictive model, the HCC-ART score, could increase the accuracy of HCC screening and surveillance [8]. In the present work, we have made significant improvements to the reliability of that model by verifying its applicability to the detection of small-sized and early-stage HCC. We found that the HCC-ART score increased significantly as tumor size increased from small to very large. For small tumors ( $\leq 2$  cm), the calculated AUC values for the HCC-ART, the simplified HCC-ART, and AFP were 0.871, 0.934, and 0.560, respectively, indicating that

the HCC-ART is a superior small-HCC biomarker to AFP. The sensitivities of these biomarkers to small ( $\leq 2$  cm) and medium-sized (3–5 cm) HCCs were 70 and 97% for the HCC-ART, 82 and 100% for the simplified HCC-ART, and 12 and 100% for AFP, respectively. Excellent results were obtained when the HCC-ART was used to distinguish between different tumor sizes.

Due to these favorable characteristics, the differential diagnostic accuracy of the HCC-ART for detecting early-stage HCC according to the Okuda (stage 1), CLIP (0-1), and BCLC (stage A) staging systems was assessed. Another attractive aspect of the HCC-ART is that it has a higher AUC and sensitivity than and comparable specificity to AFP for the detection of early-stage HCC (Table 3). Increased HCC-ART score was significantly correlated with HCC tumor size as well as Okuda, CLIP, and BCLC stage.



**Table 3** Predictive power of HCC-ART, simplified HCC-ART, and AFP in the diagnosis of HCC, using Okuda, CLIP, and BCLC as gold-standard staging systems

Classification system	Number	HCC-ART at a cutoff of $\geq 0.5$			Simplified HCC-ART at a cutoff of $\geq 280$			AFP at a cutoff of $\geq 400$ U/L		
		AUC	Sen. (%)	Spe. (%)	AUC	Sen. (%)	Spe. (%)	AUC	Sen. (%)	Spe. (%)
Okuda stage										
Stage 1 (early)	115	0.988	90	100	0.950	84	99	0.732	42	100
Stage 2 (intermediate)	169	1.000	100	100	0.973	95	99	0.828	43	100
Stage 3 (advanced)	34	1.000	100	100	0.993	100	99	0.875	56	100
CLIP stage										
CLIP 0–1 (early)	100	0.992	93	100	0.945	84	99	0.632	10	100
CLIP 2–3 (intermediate)	175	0.997	98	100	0.972	94	99	0.850	55	100
CLIP $\geq 4$ (advanced)	43	1.000	100	100	1.000	100	99	1.000	99	100
BCLC stage										
Stage 0-A (early)	76	0.968	85	100	1.000	100	99	0.487	8	100
Stage B (intermediate)	86	0.975	86	100	0.998	100	99	0.684	31	100
Stage C (advanced)	76	1.000	100	100	1.000	100	99	0.871	40	100
Stage D (end-stage)	80	1.000	100	100	1.000	100	99	0.852	50	100

AUC was generated by comparing HCC to liver cirrhotic patients

Sen. sensitivity, Spe. specificity, AUC area under the curve

**Table 4** Correlation of HCC-ART and AFP with tumor size and different HCC classification systems

Variables	AFP	ART	Simplified HCC-ART	Tumor size	Okuda	CLIP	BCLC
AFP	1.0	0.2 ( $P = 0.001$ )	0.2 ( $P = 0.001$ )	0.1 ( $P = 0.03$ )	0.1 ( $P = 0.3$ )	0.2 ( $P = 0.001$ )	0.3 ( $P = 0.003$ )
HCC-ART	0.2 ( $P = 0.001$ )	1.0	0.9 ( $P < 0.0001$ )	0.5 ( $P < 0.0001$ )	0.5 ( $P < 0.0001$ )	0.5 ( $P < 0.0001$ )	0.7 ( $P < 0.0001$ )
Simplified HCC-ART	0.2 ( $P = 0.001$ )	0.9 ( $P < 0.0001$ )	1.0	0.8 ( $P < 0.0001$ )	0.5 ( $P < 0.0001$ )	0.5 ( $P < 0.0001$ )	0.5 ( $P = 0.001$ )
Tumor size	0.1 ( $P = 0.03$ )	0.5 ( $P < 0.0001$ )	0.8 ( $P < 0.0001$ )	1.0	0.2 ( $P = 0.03$ )	0.2 ( $P = 0.02$ )	0.2 ( $P = 0.03$ )
Okuda	0.1 ( $P = 0.3$ )	0.5 ( $P < 0.0001$ )	0.5 ( $P < 0.0001$ )	0.2 ( $P = 0.03$ )	1.0	0.4 ( $P < 0.0001$ )	0.3 ( $P = 0.005$ )
CLIP	0.2 ( $P = 0.001$ )	0.5 ( $P < 0.0001$ )	0.5 ( $P < 0.0001$ )	0.2 ( $P = 0.02$ )	0.4 ( $P < 0.0001$ )	1.0	0.5 ( $P < 0.0001$ )
BCLC	0.3 ( $P = 0.003$ )	0.7 ( $P < 0.0001$ )	0.5 ( $P = 0.001$ )	0.2 ( $P = 0.03$ )	0.3 ( $P = 0.005$ )	0.5 ( $P < 0.0001$ )	1.0

\* Pearson's correlation was used.  $P > 0.05$  is considered nonsignificant,  $P < 0.05$  is considered significant

The outcomes derived from this study compared favorably with other biomarkers used for the diagnosis of early-stage HCC. For example, high serum concentrations of DCP are present in 50–60% of all HCC patients, but in only 15–30% of early-stage HCC cases [13]. AFP-L3 exhibits a sensitivity of only 35% in patients with small HCC (<2 cm) [14]. Recently, similar promising results were obtained when the biomarker Dickkopf-1 (DKK1), a secretory antagonist of the canonical Wnt signaling pathway, was employed to detect early-stage HCC (70.9% sensitivity and 90.5% specificity) [15]. However, it is unlikely that DKK1 is specific for HCC. Overexpression of DKK-1 has been reported in many malignant tissues, including breast, lung, esophageal, ovarian, and gastric cancers [16]. In contrast, our model is based on inflammatory markers, markers

associated with liver fibrosis, and AFP, all of which are associated with liver cell proliferation; they reflect the disease status of the liver.

The application of a staging system is important not only for obtaining prognostic information but also in management planning. For HCC, there are many proposed staging systems [17]. The classifications most commonly used for HCC are the Okuda, CLIP, and BCLC staging systems. Although the Okuda staging system was the first classification to include both tumor parameters and liver function factors [10], its main problem is that the assessment of tumor burden is applicable only to advanced-stage tumors [18]. The most robust predictors of HCC prognosis are portal vein thrombosis, tumor size, AFP, and Child–Pugh class. Interestingly, the CLIP system, one of the most commonly used HCC-staging

systems, includes all of these predictors [19]. A consensus conference, the Americas Hepato-Pancreato-Biliary Association/American Joint Committee on Cancer Conference, endorsed the use of CLIP for prognostic HCC staging because it was the only externally validated staging system [20]. The CLIP score has been well validated for the detection of HCC in Western patients. However, a CLIP score of zero is a tumor extent of less than 50% of the liver, and 50% of the liver is a fairly large tumor considering that many early-stage HCCs with nodules less than 2 cm in diameter can routinely be detected in many countries [21]. The BCLC staging for HCC is currently the only system that includes an integrated assessment of liver disease, tumor extension, and presence of constitutional symptoms, providing an indication of the first-line treatment [22]. It is based on the possibility of curative intervention, so, the BCLC staging system is actually an extension of the treatment-decision algorithm rather than a prognostic system [18]. Studies that have evaluated more than these three systems have shown that the BCLC system is the most useful [23]. We found that, among the three staging systems, the HCC-ART score showed the strongest correlation ( $r = 0.7$ ,  $P < 0.0001$ ) with the BCLC system. There was a strong positive correlation between the simplified HCC-ART ( $r = 0.8$ ;  $P < 0.0001$ ) and the tumor size in contrast to Okuda, CLIP and BCLC scores.

The HCC-ART is based on simple, noninvasive, and routinely measured biomarkers (age, AFP, aspartate aminotransferase/alanine aminotransferase ratio (AAR), alkaline phosphatase, and albumin). Oxidative DNA damage is known to contribute to the progression of chronic liver damage to HCC, and is correlated with the induction of telomerase and repair enzyme activity [24, 25]. On the other hand, these repair mechanisms, such as CuZn superoxide dismutase [26], are known to degrade with age, and the *hOGGI* gene encodes a DNA glycosylase [27] that contributes to hepatocarcinogenesis in the presence of persistent oxidative damage. AFP alone cannot be considered the ideal biomarker for HCC. However, the combined detection of AFP along with other complementary serum biomarkers could improve the early diagnostic rate of HCC [28]. A higher AAR has been reported in patients with HCC compared with those with cirrhosis, likely due to an increase in cytosolic aspartate aminotransferase [29]. AAR in combination with age, AFP, albumin, and other liver-related seromarkers were recently included in predictive models for the risk of HCC [30]. Recent studies suggest that ALP could serve as an easy-to-assess biomarker that permits the early prediction of HCC recurrence after hepatectomy of large tumors [31, 32]. Liver carcinogenesis is associated with a decrease in serum albumin due to liver impairment. The level of serum human albumin is usually normal in chronic liver diseases until the onset of cirrhosis and the development of primary liver cancer.

The most robust predictors of death in HCC patients are tumor-related (e.g., AFP) and liver-related (e.g., AAR, ALP, and albumin), and these are included in the HCC-ART [19]. Thus, applying the HCC-ART may improve the prognosis of HCC. Nevertheless, in order to determine the actual prognostic value of the HCC-ART in HCC cases, further studies comparing its accuracy in a large population to other staging systems are required. Although validation studies that apply the HCC-ART in different institutions, different patient groups, and different countries are needed, we propose that the HCC-ART is a highly sensitive and specific score for differentiating small-sized and early-stage HCC from liver cirrhosis. Because it is quite easy to determine the HCC-ART score via routine laboratory tests, we strongly recommend its routine use in clinical practice. Also, because it gives similar promising results to the original HCC-ART and its advantage of minimal variability and a wide continuous scale for assessing HCC severity, the simplified HCC-ART has the potential to be even more widely used than the original HCC-ART.

#### Compliance with ethical standards

**Conflict of interest** None.

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