Am J Cancer Res 2016;6(10):2375-2385
www.ajcr.us /ISSN:2156-6976/ajcr0039686

Original Article

Predictive values of serum VEGF and CRP levels combined with contrast enhanced MRI in hepatocellular carcinoma patients after TACE

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Received September 8, 2016; Accepted September 15, 2016; Epub October 1, 2016; Published October 15, 2016

Abstract: This study explored the predictive value of serum vascular endothelial growth factor (VEGF) and C-reactive protein (CRP) levels combined with enhanced magnetic resonance imaging (MRI) in hepatocellular carcinoma (HCC) patients after transcatheter arterial chemoembolization (TACE). One hundred and seventeen patients who received TACE from June 2010 to December 2012 in our hospital were included in this study. Serum VEGF and CRP levels before and after TACE were determined by ELISA and single immunodiffusion method for analyzing the association of serum levels with pathological features. Enhanced MRI was utilized before and after TACE to measure tumor size and ADC value in enhanced region and non-enhanced region. MRI data were combined with serum VEGF and CRP levels to analyze the predictive value in efficacy and prognosis for HCC patients after TACE. The serum VEGF and CRP levels after TACE were increased, but can return to normal levels in a certain time. VEGF and CRP levels were not statistically associated with tumor location, tumor staining or presence of membrane (all $P > 0.05$), but closely correlated with combined portal vein tumor thrombus, combined arteriovenous fistula and distant metastasis (all $P < 0.05$). Low levels of serum VEGF and CRP, small tumor size and low ADC value before treatment indicated a better prognosis. The sensitivity and specificity of serum VEGF and CRP levels, tumor size and ADC value were respectively 92.31% and 88.46%, 93.85% and 90.38%, 81.54% and 78.85% as well as 47.69% and 84.62%. Serum VEGF and CRP levels, tumor size and ADC value could predict the efficacy of TACE for HCC patients. Serum VEGF and CRP levels combined with enhanced MRI may serve as markers for efficacy and prognosis evaluation in HCC patients after TACE.

Keywords: Hepatocellular carcinoma, vascular endothelial growth factor, c-reactive protein, enhanced magnetic resonance imaging, transcatheter arterial chemoembolization, clinical feature, efficacy, prognosis

Introduction

Primary liver cancer is the 5th most common malignant tumor with high incidence and mortality, among which hepatocellular carcinoma (HCC) is the most prevalent histologic type, accounting for 85%~90% of primary liver cancer [1]. HCC as the second leading cause of tumor associated death worldwide, results in approximately 0.6 million annual mortalities internationally [2]. The main risk factors for HCC are hepatitis B virus/hepatitis C virus infection, alcoholism, nonalcoholic steatohepatitis and diabetes [3]. Besides, evidence supported that majority of the HCC patients had unfavorable liver function and only those patients in early stage or resectable HCC were suitable for hepatic resection [4]. As a result, transcatheterarterial chemoembolization (TACE) becomes one of the available choices for those patients in intermediate- or advanced-stage HCC [5]. This treatment works through specifically restricting blood supply to the tumor and "starve" the tumor to death; meanwhile, the chemical drugs from the embolization agents retain for a prolonged period of time, resulting in more deposit in the tumor tissues and much longer in vivo life of the drugs [6].

However, the efficiency of TACE treatment is limited by recurrence and metastasis of residual nodule [7]. As the molecular biology of HCC was further explored, researchers found that certain genes and proteins involved in regulation of tumor cell differentiation might contribute to HCC relapse and metastasis [8]. Therefore, finding a reliable molecular tag and method to predict relapse post TACE treatment becomes imperative as it is critical for planning appropriate further medical measurements for HCC in advanced stage.

As the most important means for prognostic evaluation, Magnetic Resonance Imaging (MRI) can discover and scan the lesion at various positions, on multiple layers, and though different angles; multiple sequences scanning can indirectly reveal different lesion pathology change after treatment, with extremely high resolution, sensitivity and specificity [9, 10]. Diffusion-weighted MRI (DWI) can determine the lesion nature by the quantification of apparent diffusion coefficient (ADC) value; dynamic contrast-enhanced scanning MRI (DCE-MRI) can specifically reveal the tumor neovascularization after intervention procedure [11]. Despite the aforementioned advantages, MRI does have several disadvantages with a limited sensitivity. Evidence showed that MR imaging features have unsatisfied sensitivity regarding the identification of cervical lymph node metastases in patients with thyroid cancer [12]. The emergence of neovessels is critical for tumor invasion and metastasis and vascular endothelial growth factor (VEGF) is regarded as the most potent pro-angiogenic factors secreted by tumor cells and is closely associated with growth, invasion and metastasis of tumor cells [13]. It has been demonstrated that abnormal expression of VEGF was associated with occurrence of HCC, and the more expression of VEGF was suggested for a higher chance of recurrence [14]. C-reactive protein (CRP) is an acute-phase protein of hepatic origin. Excessive expression of CRP usually indicates occurrence of malignant tumor or metastasis [15]. It has been shown that serum CRP level is the prognosis biomarker of a variety of cancers [16, 17].

To the best knowledge of the authors, there has not been any literature reporting the combined use of serum VEGE and CRP levels with enhanced MRI results as prognostic biomarker of HCC after TACE. Therefore, in this study we examined the prognostic value of serum VEGF and CRP levels combined with MRI results from HCC patients who underwent TACE treatment with the aim to identify new prognostic approach for TACE treatment and improve the overall survival of the HCC patients.

Materials and methods

Ethical statement

The First Affliated Hospital of Soochow University approved the study design. All patients had to sign informed consent in written form to undergo diagnostic and therapeutic procedure at the time of hospitalization. This study complied with the guidelines and principles of the declaration of Helsinki [18].

Subjects

A total of 117 intermediate or advanced HCC patients who were diagnosed according to TNM classification of liver cancer [19] and Barcelona Clinic Liver Cancer (BCLC) staging classification [20], were included in this study. Among them, there were 86 males and 31 females, age ranged from 31 to 74 years with median age of 51.74 ± 7.92 years. All patients were diagnosed with HCC by clinical examination, imageology and α-fetoprotein (AFP) examination, and or further confirmed by puncture biopsy pathology. None of them had the indications for surgery resection or the subjects refused the surgical resection. The patients were included according to following criteria: older than 18 years and expected survival time longer than 3 months; no history of surgery, chemotherapy, radiotherapy or other loco-regional therapy; capable of action competence; signed the consent voluntarily; willing to follow doctors instructions; no liver transplantation history or any indicator for surgical resection; liver function categorized to class A according to Child classification or categorized as class B but improved to A after regular treatment. The exclusion criteria were: liver function were severely damaged with hepatic cirrhosis decompensation and were categorized to class C according to Child classification; overall health status is poor and unable to tolerate surgical operation; other medical conditions such as diabetes, lung dysfunction, impaired renal function and severe heart disease; blood circulation, coagulation

disorders; serious infection or local active infection; large scale metastasis to multiple organs other than liver.

TACE treatment strategy

All patients underwent hematology tests (complete blood count, biochemical test, blood type, blood coagulation, liver function, renal function, AFP and CA19-9), electrocardiogram, postero-anterior and lateral X- ray chest films and ultra-sound examination or CT examination. Patients were fasted for 12 hours and underwent iodine allergy test or iopromide injection, and routine skin cleaning before surgery operation. Fifteen minutes before operation, 8 mg of Ondansetron (Qilu Pharma, Jinan, Shangdon, China; H10970065) were injected intravenously on right hand to prevent vomiting. After local anesthesia through 5-10 ml 2% lidocaine (Bayer, Leverkusen, Germany) injection 1 cm under inguinal ligament, TACE was performed using Seldinger technology. After left femoral arteriopunction, 5F shealth and catheter was placed. Then high pressure injector was used for visualization on ceoliac trunk artery or superior mesenteric artery, to locate the position of tumor lesion, diameter and quantity, and also to confirm supportive artery for the tumor. Superselective arterial cannula chemotherapy and hepatic arterial infusion chemotherapy were performed using chemotherapy agents including 1.0 gram of Tegafur (Fresenius Medical Care, Bad Homburg, Germany), 30~50 mg of hydroxycamptothecin (HCPT, Zhejiang Kancheer Pharmaceutical, Dongyang, Zhejiang, China) and 30 mg of Epirubicin (EPI, Chongqing Laimei Pharmaceutical, Chongqing, China; batch number, 1304010), mixed with 5~20 ml of lipiodol emulsion. After super-selective embolization of tumor blood supply artery, the chemotherapy mixture was supplied to the tumor in adjusted volume according to the tumor size and ADC value. For patients with thin artery or tortuous vessels, 3F micro-catheter was used for segmental embolization. For patients with excessive blood supply and/or AVF + AP-shunt, PVA or gelatin sponge particle embolization was used until the entire tumor blood supply artery was blocked and tumor staining vanished completely.

After operation, the puncture point was pressed for 15 minutes, bandaged and sent back to ward, followed by careful monitoring of blood pressure, heart rate, pulse and breathe. Patients were fasted for further 6 hours and left lower limb was immobilized for 24 hours. The puncture point was further checked for potential hemorrhage and dorsal pedis artery was checked for pulse.

Blood sample collection

Blood samples of 6 ml were taken from patients with empty stomach on 1 day before TACE treatment, and 1 day, 3 days, 7 days, 14 days and 28 days after treatment. The samples were collected into sterile glass tube with 0.667 ml of 0.129 mol/L trisodium citrate, and then centrifuged at 4°C at 3000 rpm for 5 minutes within 2 hours of collection. The separated serum was stored in -70°C.

ELISA detection for VEGF and immune scatter turbidimetry for CRP

Blank control, standard and test wells were set up in the ELISA assay. The wells were added with diluted samples, then sealed and incubated at 37°C for 2 hours. After the liquid were abandoned, the wells were added with detection solution A100 (formulated immediately before use) and sealed for 1 hour incubation at 37°C. Then the samples were undergone aspiration of the solution, washing three times and soaking for 1 to 2 minutes and drying. After added detection solution B, the plate was sealed, 37°C incubated for 1 hour, washed for four times. In the final step, 100 μL of coloration solution was added to the wells and then the plate was incubated at room temperature in darkness for 15 minutes, before adding 100 μL termination solution. The absorbance (OD) was measured by spectrometer (Thermo lab system, Finland) with a wavelength of 450 nm. The concentration of each sample was calculated from the standard curve. The VEGF assay kit was purchased from Jinmei Biotechnology (Shenzhen, Guangdong, China). The standard concentration is 227.34 ng/L [21], so concentration higher than 227.34 ng/L would be categorized to high expression and lower one would be considered as low expression.

Blood (3 ml) collected as mentioned above was centrifuged to separate the serum within 1 hour of collection. The detections were performed on the same day of collection. Immune scatter turbidimetry was employed to deter-

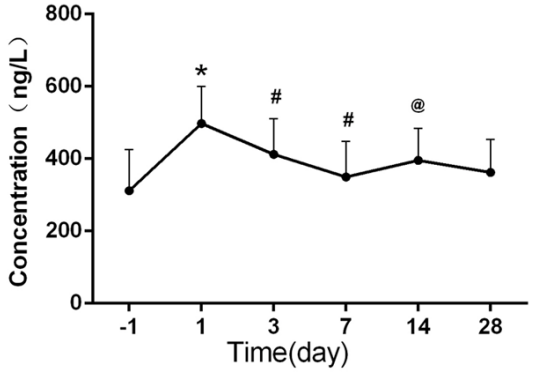

Figure 1. Curve of VEGF level variation after TACE treatment. Note: *P < 0.05, compared to before treatment; #P < 0.05, compared to one day before treatment; @P < 0.05, compared to 7 days after treatment; TACE, transcatheter arterial chemoembolization; VEGF, vascular endothelial growth factor.

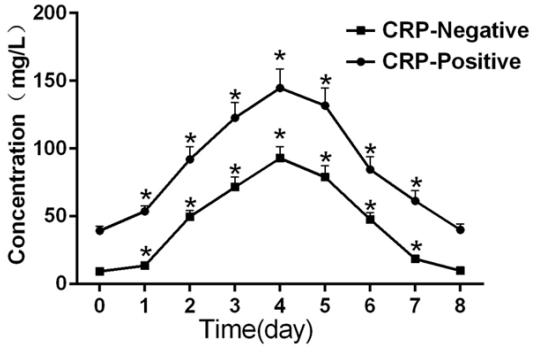

Figure 2. CRP concentration variations from patients with either positive CRP or negative CRP level. Note: *P < 0.05, compared to before treatment; CRP, C-reactive protein.

mine the serum CRP level using BNPProSpec automatic protein analyzer and accompany reagents (Dade Behring BN Prospec, USA). The reference value is < 10 mg/L with detection limit of 3.08 mg/L [22]. Therefore in this study, CRP ≥ 10 mg/L will be regarded as positive and < 10 mg/L will be negative.

Detection on tumor lesion activity and evaluation on treatment efficacy

Patients underwent MR scan on the day before and 28 days after embolization treatment. MR routine scan, DWI and dynamic contrast enhanced scan and liver acceleration volume acquisition (LAVA) sequences were employed using cross-sectional LAVA method. Gd-DTPA of 0.2 mmolkg was injected into vein using high pressure injector (at inject rate of 2 ml/s), arterial phase was collected 20~22 seconds after injection, portal phase was collected 60~70 seconds after injection, and equilibrium phase was started 150 seconds after injection. Scanning parameters were as follows: TR/TE3.2 ms/1.6 ms; twist angle at 15°; inversion time at 70 ms; receiver band width of 83.3 kHz, FOV of 38~40 cm; matrix of 256 × 224; ephase FOV of 0.9; slice thickness of 2 mm. Array space sensitive coding technique (ASSET) was also employed with phase accelerator of 2 Ph, reconstructed slice thickness of 2 mm and scan time of 16~18 seconds, using zero fill interpolation processing (ZIP × 2).

The size and location of tumors were recorded from images with decent quality; with most of the entire tumor lesion included in the image. The Region of Interesting (ROI) and ADC value of tumor lesion before and after TACE was measured by Functool software in a GEAW4.3 workstation. ADC value was taken from each slice of the tumor lesion and the lesion ADC value was the average value of ADC values from all the slices. Combined with the enhanced results from dynamic contrast enhanced scan, ROI was established in the lesion enhanced region and non-enhanced region, and the average ADC values of tumor enhanced region and non-enhanced region after TACE treatment was measured.

The evaluation of HCC treatment efficacy was based on response evaluation criteria in solid tumors (RECIST) [23]: complete remission (CR), partial remission (PR), stable disease (SD) and progressive disease (PD), and (CR + PR) % is the response rate (RR). The tumor size drops to normal or more than 50% was regarded as CR, between 25% to 50% as PR, less than 25% as SD and unchanged size or enlargement and signs of metastasis as PD.

Follow up

To calculate the survival time after treatment, a telephone follow up or home visit follow up was made every 1~3 months to collect cumulative survival rate. The treatment efficiency was evaluated 3 years after the treatment.

Statistical analysis

Data were analyzed using statistic software SPSS21.0. Numeration data was presented in form of percentage or rate and compared using

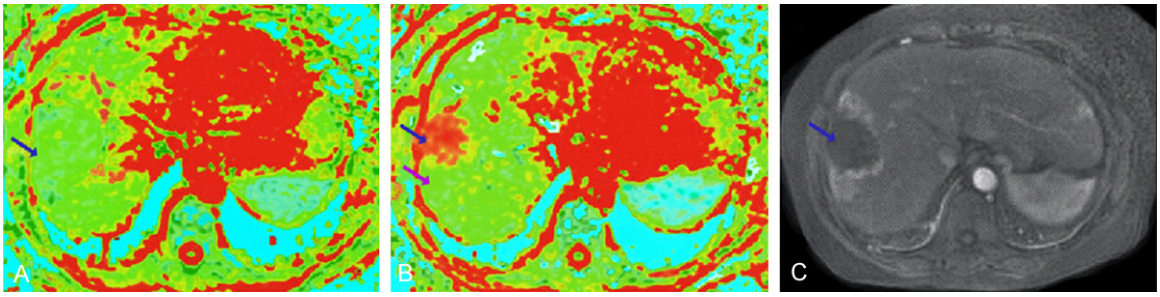

Figure 3. ADC map of HCC before and after TACE treatment. Note: A. Low signal in liver tissues surrounding the tumor (arrow); B. ADC map from the same patient 28 days after TACE treatment: strong signal in non-enhanced region (blue arrow), low signal in enhanced region (purple arrow); C. Arterial phase image from the same patient 28 days after TACE treatment: apparent non-homogenous enhanced in the tumor (arrow), non-enhanced necrosis can be seen inside the tumor; ADC, apparent diffusion coefficient; TACE, transcatheter arterial chemoembolization; HCC, hepatocellular carcinoma.

Chi-square test between groups. The measurement data were represented as mean ± standard deviation (mean ± SD). *T*-test is carried out for comparison between two groups; one-way ANOVA was used for multiple group comparisons (a test for homogeneity of variance was performed before the comparison); LSD-t test was utilized for the pair-wise comparison of average from multiple groups. The survival rate is calculated based on Kaplan-Meier method and comparison of survival rate between two groups was performed using Log-rank method. Two tailed $P < 0.05$ is regarded as significant difference.

Results

Serum VEGF and CRP levels before and after TACE

The data from this study revealed that VEGF concentration from the 117 patients after TACE treatment changed dynamically compared with pre-treatment concentration. As presented in **Figure 1**, the post-treatment VEGF was considerably elevated on Day 1 after treatment, and dropped on Day 3 and Day 7, and slightly increased on Day 14. Usually the oxygen tension in tumor after treatment leads to the up regulation of VEGF mRNA which further leads to more VEGF translation, resulting in enhancement of neovascularization to ameliorate the hypoxia. A clear pattern for CRP expression variation before and after TACE treatment was noticed. There were 68 CRP positive cases (58.12%) and 49 CRP negative cases (41.88%). After TACE treatment, the serum CRP concentration increased dramatically, then peaked on Day 4 and gradually decreased afterwards to pre-treatment level on Day 8, with a bigger rangeability in the CRP positive groups than negative group (**Figure 2**).

MR scan results

The diameter of tumor after TACE treatment was 8.78 diameter of tumor after TACE treatment oup ± 2.08 cm before TACE ($t = 3.969$, $P < 0.001$, **Table 1**). Meanwhile, the average ADC value of the whole HCC lesion after TACE was $(1.33 \pm 0.19) \times 10^{-3}$ mm^2/s, higher than the $(1.12 \pm 0.23) \times 10^{-3}$ mm^2/s value before TACE ($t = 7.614$, $P < 0.001$). After TACE all the lesions was unevenly enhanced during the dynamic contrast enhancement, and no enhancement region was observed within the lesion (**Figure 3**). After TACE, the non-enhanced region in each lesion all showed strong ADC signal in the ADC map, with average ADC value of $(1.06 \pm 0.17) \times 10^{-3}$ mm^2/s. The enhanced region of the tumors showed ADC value clearly lower than that before TACE ($t = 2.269$, $P = 0.024$) while the ADC value of the tumor non-enhanced region was significantly higher than that before TACE ($t = 18.548$, $P < 0.001$, **Table 1**).

The association between treatment efficacy and combination of VEGF and CRP levels with MRI

RECEST criteria combined with clinical pathology data were used to evaluate the HCC treatment efficiency one month after TACE treatment. The patients were categorized into two

Table 1. The comparisons on MRI pre and post TACE

Subject	Pre-TACE	Post-TACE	t	*P*
Diameter variation (cm)	9.69 ± 2.08	8.78 ± 1.35	3.969	< 0.001
ADC value (mm^2/s)	(1.12 ± 0.23) × 10^{-3}	A: (1.33 ± 0.19) × 10^{-3}	7.614	< 0.001
		B: (1.06 ± 0.17) × 10^{-3}	2.269	0.024
		C: (1.69 ± 0.24) × 10^{-3}	18.548	< 0.001

Note: A represents average ADC value after TACE treatment; B represents ADC value in the enhancement region after TACE treatment; C represents ADC value in non-enhancement region after TACE treatment; TACE, transcatheter arterial chemoembolization; ADC, apparent diffusion coefficient; MRI, magnetic Resonance Imaging.

Table 2. Relationship between serum VEGF & CRP level, MRI and clinical pathology & treatment efficacy

Parameter	CR + PR	SD + PD	X^2/t	*P*
Age (year)	51.23 ± 9.26	52.15 ± 6.72	0.622	0.535
Gender			0.009	0.925
Male	38	48		
Female	14	17		
Presence of tumor membrane			0.147	0.702
Yes	12	17		
No	40	48		
Presence of portal vein tumor thrombus			15.56	< 0.001
Yes	13	25		
No	39	40		
Presence of arteriovenous fistula			5.003	0.025
Yes	15	8		
No	37	57		
Occurrence of distant metastasis			8.603	0.003
Yes	14	35		
No	38	30		
Pre-treatment VEGF (ng/L)	230.49 ± 79.48	376.85 ± 92.64	9.037	< 0.001
Pre-treatment CRP (ng/L)	12.80 ± 9.51	38.05 ± 7.11	16.429	< 0.001
Pre-treatment tumor diameter (cm)	8.55 ± 1.96	10.60 ± 1.69	6.072	< 0.001
Pre-treatment ADC value (ss^2/m)	1.04 ± 0.22	1.19 ± 0.22	3.665	< 0.001

Note: VEGF, vascular endothelial growth factor; CRP, C-reactive protein; MRI, magnetic Resonance Imaging; CR, complete remission; PR ,partial remission; SD, stable disease; PD, progressive disease; ADC, apparent diffusion coefficient.

groups, with 52 patients with CR + PR in responsive group, and 65 patients with SD + PD patients in non-responsive group. A careful comparison revealed that the serum VEGF and CRP levels one day before treatment from SD + PD patients were significantly higher than those from CR + PR patients (both $P < 0.05$). Similarly, the tumor size (one day before treatment) of the SD + PD patients was significantly larger than that from the CR + PR patients (both $P < 0.05$). Our results also revealed that the number of cases with portal vein tumor thrombus, combined arteriovenous fistula and distant metastasis in SP + PD group were significantly different from that in the CR + PR group (both $P < 0.05$), meanwhile gender, age and cases of tumor capsule were not significantly different between the two groups (all $P > 0.05$, **Table 2**).

The association of survival rate with serum VEGF and CRP level, tumor size and ADC value

Follow up of 117 patients (with no drop out) was continued for 3 years (according to the last follow up date). The patients were categorized into two groups based on serum VEGF value

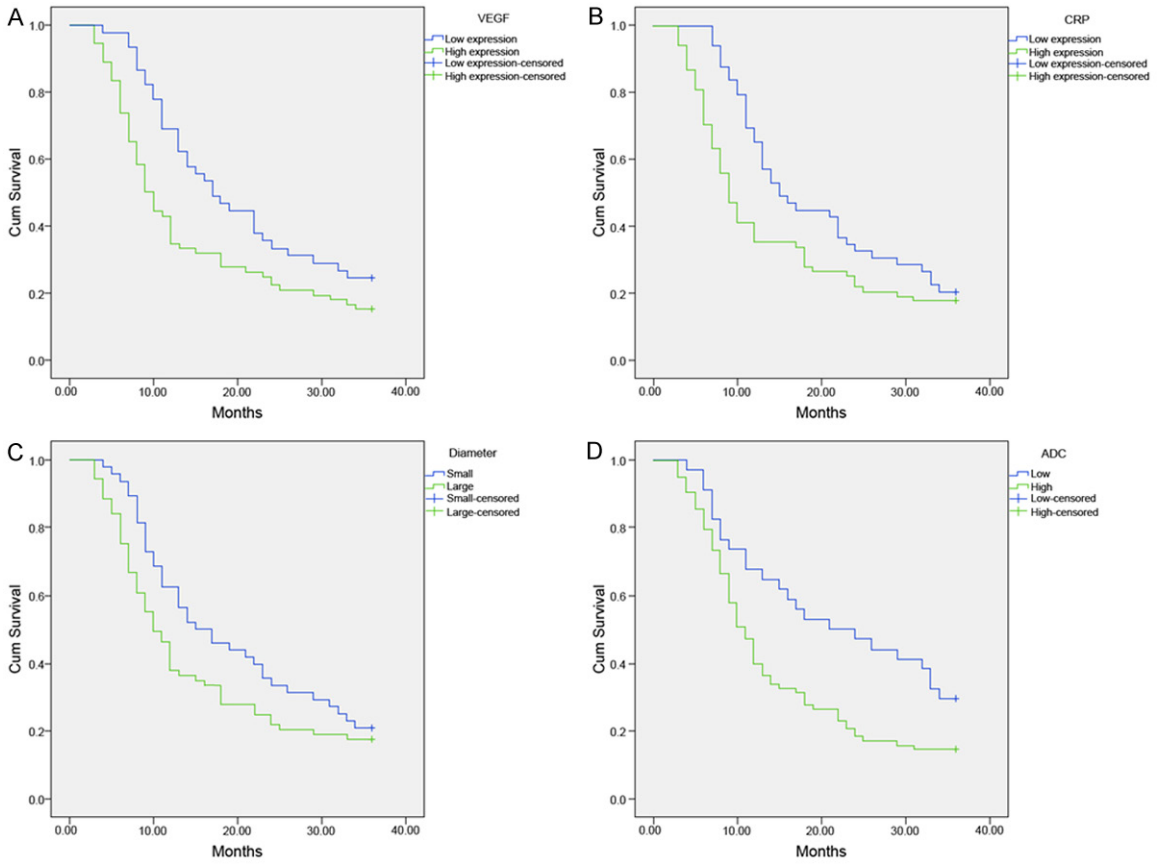

Figure 4. Survival curve analysis. Note: A. Survival curve of patients with different VEGF expression level: low (1) and high (2) serum VEGF level group one day before treatment; B. Survival curve of patients with different CRP expression level: low (1) and high (2) serum CRP level group one day before treatment; C. Survival curve of patients with different tumor size: small (1) and large (2) tumor size group; D. Survival curve of patients with different MR determined ADC value: low (1) and high (2) ADC value group; ADC, apparent diffusion coefficient; VEGF, vascular endothelial growth factor; CRP, C-reactive protein; MRI, magnetic Resonance Imaging.

before treatment. For the 72 cases in the high level group, the survival rates at 12, 24 and 36 months were 40.28%, 23.61% and 15.28% respectively; for the 45 cases in the low lever group, these rates were 71.11%, 33.33% and 24.44% respectively (**Figure 4A**). Similarly, the patients can categorize into two groups based on serum CRP value before treatment. For the 68 cases in the high level group, the survival rates at 12, 24 and 36 months were 44.12%, 23.19% and 17.39% respectively; for the 49 cases in the low lever group, these rates were 63.27%, 33.33% and 20.83% respectively (**Figure 4B**). Using 10 cm tumor size before treatment as standard, the patients can categorize into two groups. For the 69 cases in the large tumor size group, the survival rates at 12, 24 and 36 months were 44.93%, 23.19% and 17.39% respectively; for the 48 cases in the small tumor size group, these rates were 62.50%, 33.33% and 20.83% respectively (**Figure 4C**). Finally, with pre-treatment lesion ADC value of 1.0 ss^2/m as standard, the patients can categorized into two groups. For the high ADC value group, the survival rates at 12, 24 and 36 months were 46.99%, 19.28% and 14.46% respectively; for the low ADC value group, these rates were 64.71%, 47.06% and 29.41% respectively (**Figure 4D**). Log-rank non-parameter test were used to compare these results and it indicated that groups with low level of pre-treatment serum VEGF and CRP and small contrast-enhanced ADC value have higher survival rate.

The diagnostic value of serum VEGF and CRP level, tumor size and ADC value

The Areas under the ROC Curve (AUC) of serum VEGF, serum CRP, pre-treatment tumor size and

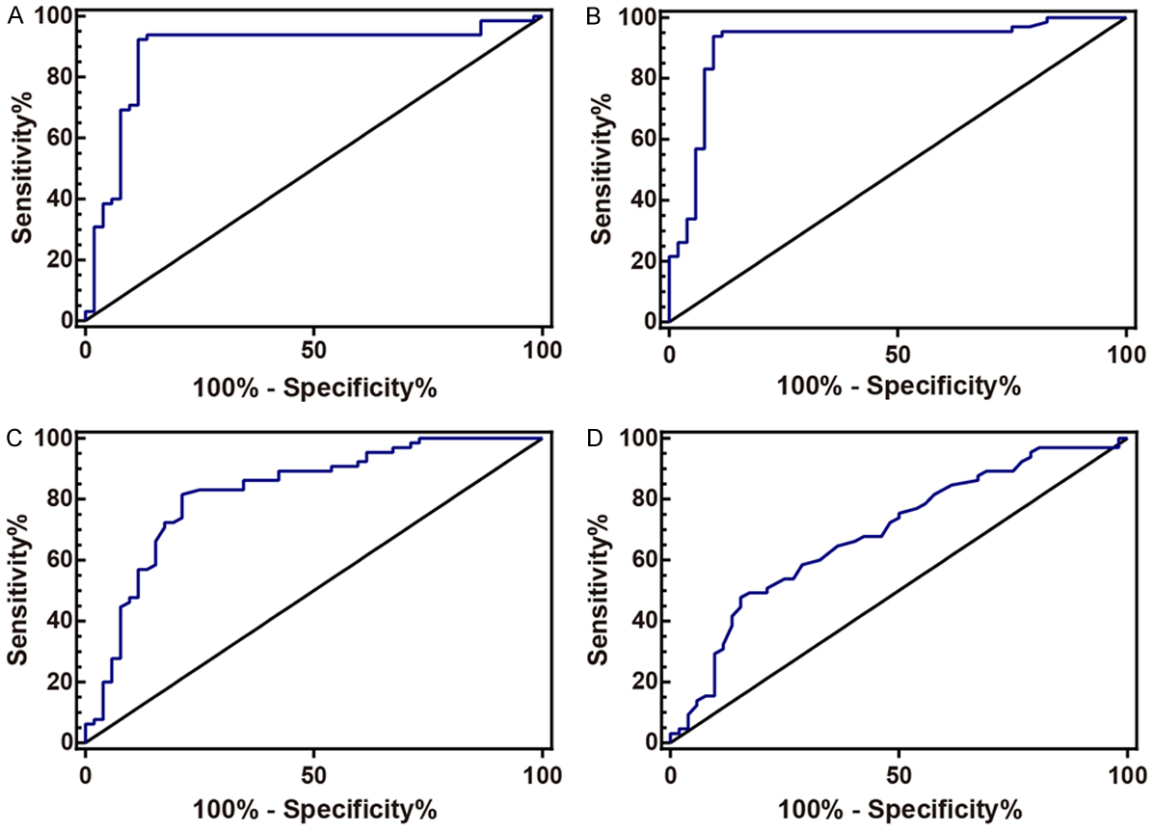

Figure 5. The ROC curves of pre-treatment VEGF, CRP, tumor diameter and ADC value. Note: A. ROC analysis of VEGF level before treatment; B. ROC analysis of CRP level before treatment; C. ROC analysis tumor size before treatment; D. ROC analysis of ADC value before treatment; ADC, apparent diffusion coefficient; VEGF, vascular endothelial growth factor; CRP, C-reactive protein; MRI, magnetic Resonance Imaging; ROC, receiver operating characteristic.

pre-treatment ADC value were 0.885 (95% CI: 0.814~0.956), 0.914 (95% CI: 0.853~0.975), 0.824 (95% CI: 0.746~0.903) and 0.688 (95% CI: 0.591~0.785) respectively with the best cut-off score of 247.1, 34.08, 10.10 and 1.24 respectively. The sensitivity and specificity for VEGF, CRP, tumor size and ADC value at this cut-off score were 92.31% and 88.46%, 93.85% and 90.38%, 81.54% and 78.85%, 47.69% and 84.62% respectively (**Figure 5A-D**).

Discussion

In this study, the serum VEGF and CRP concentration in combination with contrast-enhanced MRI were evaluated for its application in predicting TACE treatment efficacy and prognosis in HCC, in a purpose to provide new prognostic predictor for TACE treatment in the future. Our results indicated that serum VEGF level increased considerably one day after TACE treatment in patients with HCC. VEGF plays an important role in carcinogenesis as it promotes vascular endothelial cells proliferation, accelerates degradation of extra-cellular matrix and suppress apoptosis, resulting in tumor angiogenesis [24]. The TACE treatment intensified hypoxia in tumor micro-environment, and the hypoxia induces cells to produce hypoxia induced factor 1 α (HIF-1 α) which would bind to DNA, resulting in up regulation of VEGF mRNA and subsequently VEGF translation [25]. The unregulated synthesis of VEGF promotes cancer angiogenesis and improves the oxygen supply of cancer cells. It has been shown that the HIF-1 α level was correlated with VEGF concentration in TACE treated PHC patients' serum, implying that HIF-1 α can increase cancer metastasis through upregulation of VEGF level to increase tumor angiogenesis [26]. It has also been proven that intervention of VEGF expression with drugs can inhibit the emergence of HCC [27].

In this study we also found that serum CRP concentration increased drastically and peaked on Day 4 after TACE treatment. CRP is an acute-phase protein synthesized by hepatocytes and its concentration is usually upregulated in cases of infection or severe injury [28]. During TACE treatment, iodine oil embolization leads to tumor hypoxia and further increases inflammation reaction in tumor and liver, releasing a variety of pro-inflammation cytokines such as IL-1, IL-6 and IL-17. IL-6 is correlated with HCC carcinogenesis and metastasis and it can strongly induce synthesis of CRP in hepatocytes [15, 29, 30]. This study revealed that CRP rangeability before and after TACE treatment in CRP positive group is much larger than that in the CRP negative group while the survival rate CRP positive group is lower than that of the negative group, which proved that CRP may be a reliable indicator for TACE efficacy or HCC prognosis, as an increased CRP before TACE treatment is associated with a poor prognosis. Consistent with our results, a study conducted by Jun CH et al demonstrated that a high serum CRP level was proved to have certain relation with a large tumor size and a poorly defined tumor type for patients with HCC undergoing TACE [31].

The DCE-MRI results of this study indicated that tumor size became smaller and the average ADC value in HCC lesion increased after TACE treatment and the ADC value in the tumor necrotic region was much higher than that in the survived region, suggesting that ADC value helps evaluation of tumor viability. ADC value, as a quantitative parameter, is the apparent diffusion coefficient value of water molecule quantified by DWI [32, 33]. After embolization, the tumor cells undergo necrosis and apoptosis, with cell membrane permeability increased, cell gap enlarged, and these leads to more water molecule movement and increased ADC value [34]. Therapeutic regimens, physical or radiation results necrosis and cell fragmentation, further leads to increased water diffusion and ADC value, which proves the effectiveness of the therapies [35]. Therefore, ADC variance before and after treatment indirectly indicates tumor necrosis, and this further implies that MR data can be used as biomarker for HCC patient TACE treatment prognosis evaluation [36]. Bonekamp S studied the ADC value variation HCC lesion in patients before TACE, as well as 1 and 6 months after TACE, and found that increase in ADC value provides an early assessment of long term treatment of TACE in HCC [37].

Although currently imaging is an indispensable method for prognostic evaluation of TACE treatment, factors such as iodized oil deposition after embolization, inflammation after treatment and intestinal gas, can all interfere with the imaging examination, therefore other examination methods need to be combined with imaging method for a precise prognostic evaluation. Our studies indicated that serum VEGF and CRP level, tumor diameter and ADC value of the ineffective SD + PD group patients on the day before treatment, is higher than that of the effective CR + PR group. Besides, the survival results in the following three years revealed a higher survival rate in the group with lower serum VEGF and CRP level and decreased ADC values before treatment. Therefore, parameters such as VEGF and CRP level and tumor size can be combined with MRI in the evaluation of embolization treatment effect.

In summary, current study clearly stated that combination of the four parameters (serum VEGF, serum CRP, tumor size before treatment, and ADC value before treatment) may provide evaluation for the efficacy of TACE treatment and HCC prognosis, and may be recommended as a new prediction method for evaluation of TACE efficacy and prognosis. Due to the limited sample quantity, more clinical observations are needed to verify the results in this study. Moreover, the molecular mechanism of VEGF and CRP level variation was not investigated in this study and will need to be elucidated in the future.

Acknowledgements

This study was supported by a grant from National Natural Science Foundation of China (No. 81501563) and Science and Technology Research Project of Suzhou (KJXW2013003) and (SYS201611). We would like to acknowledge the helpful comments on this paper received from our reviewers.

Disclosure of conflict of interest

None.

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