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DVP parametric imaging for characterizing ovarian masses in contrast-enhanced ultrasound

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Summary

Aim: To evaluate whether parametric imaging with contrast-enhanced ultrasound is an approach capable of for the differential diagnosis of ovarian masses. *Materials and Methods:* The authors analysed 50 cases of ovarian masses by routine ultrasound and contrast-enhanced ultrasound with a new dedicated parametric image processing software—Sonoliver. The angiogenesis and blood perfusion mode on a digital video recorder were recorded and the morphological characteristics of time-intensity curve (TIC) and dynamic vascular pattern (DVP) curve were subsequently described. The quantity factor, including time to peak (TTP), maximum intensity (IMAX), rise time, (RT), mean transit time (mTT), generated by Sonoliver software were compared in both histological gradings. *Results:* There were 24 cases (86%) displaying mainly hypo-enhanced with blue imaging in those with benign masses and 15 cases (68%) displaying mainly hyper-enhanced imaging with red in those with malignant masses. The difference was statistically significant ($p < 0.05$). DVP curves were unipolar below the baseline in 23 cases (82%) of benign masses and unipolar above the baseline in 15 cases (68%) of malignant masses. IMAX, TTP, and mTT were all significantly higher in those with malignant masses than those with benign ones (all $p < 0.05$), but, no statistical difference in the RT between the two groups was found ($p > 0.05$). *Conclusions:* According to the results, DVP parametric imaging is a new approach capable of differential diagnoses of overian masses with contrast-enhanced ultrasound.

Key words: Angiogenesis; Contrast-enhanced ultrasound; Time-intensity curve; Dynamic vascular pattern curve; Ovarian mass.

Introduction

Tumor-associated neoangiogenesis is believed to be a critical requirement for tumor growth and metastasis in ovarian cancer as well as other organs [1]. Therefore, ovarian tumor-associated neoangiogenesis represents the potential to be an early detection target for ovarian cancer, the third leading cause of mortality from female genital cancer. Ultrasound (US) is recognized as the initial diagnostic modality of choice and the least invasive method for the evaluation of most pelvic masses. However, US shows limitation in the detection of small blood vessels and slow flow owing to the lack of reflection from red blood cells and low signal to noise ratio [2-4]. Many studies have shown that US did not improve the sensitivity and specificity of predicting malignancy. Therefore, an in vivo non-invasive method of detecting ovarian tumor-associated neoangiogenesis and blood perfusion is currently an important clinical concern so as to improve the early detection of early-stage disease and have a positive impact on the prognosis of this dreaded disease.

Parametric image processing in contrast enhanced ultrasound (CEUS) is increasingly being recognized as a promising and powerful molecular imaging tool [5]. In CEUS parametric imaging, the assessment of perfusion with ultrasound contrast agents (UCA) is beneficial in many diagnostic indications in molecular imaging, scattering strength of which is orders of magnitude higher than that of red blood cells. Moreover, UCA can be targeted to specific molecular markers through the attachment of appropriate ligands to the surface of the microbubbles and is administered intravenously. Microbubbles could enhance bubble non-linearities of ultrasound waves while not enhancing tissue nonlinearity [6-7]. This feature, by means of contrast-specific imaging software, makes dynamic monitoring of tumor angiogenesis and perfusion become possible. Because of that their size is comparable to red blood cell, currently available contrast microbubbles are able to stay predominantly in the blood circulation and pass through the lung microcirculation [8].

In this study, the authors examined women with suspicious but not clinically obvious malignant ovarian masses who were about to undergo surgery after conventional US. The authors' aim was to evaluate whether parametric imaging with CEUS is an approach capable of for the differential diagnosis of ovarian masses.

Materials and Methods

Patient selection

Fifty consecutive patients ages 24 to 70 years (average ± SD, 44 ± 12) who were diagnosed to have an ovarian mass that was difficult to confirm by conventional US in the present hospital from January 2009 to December 2011.

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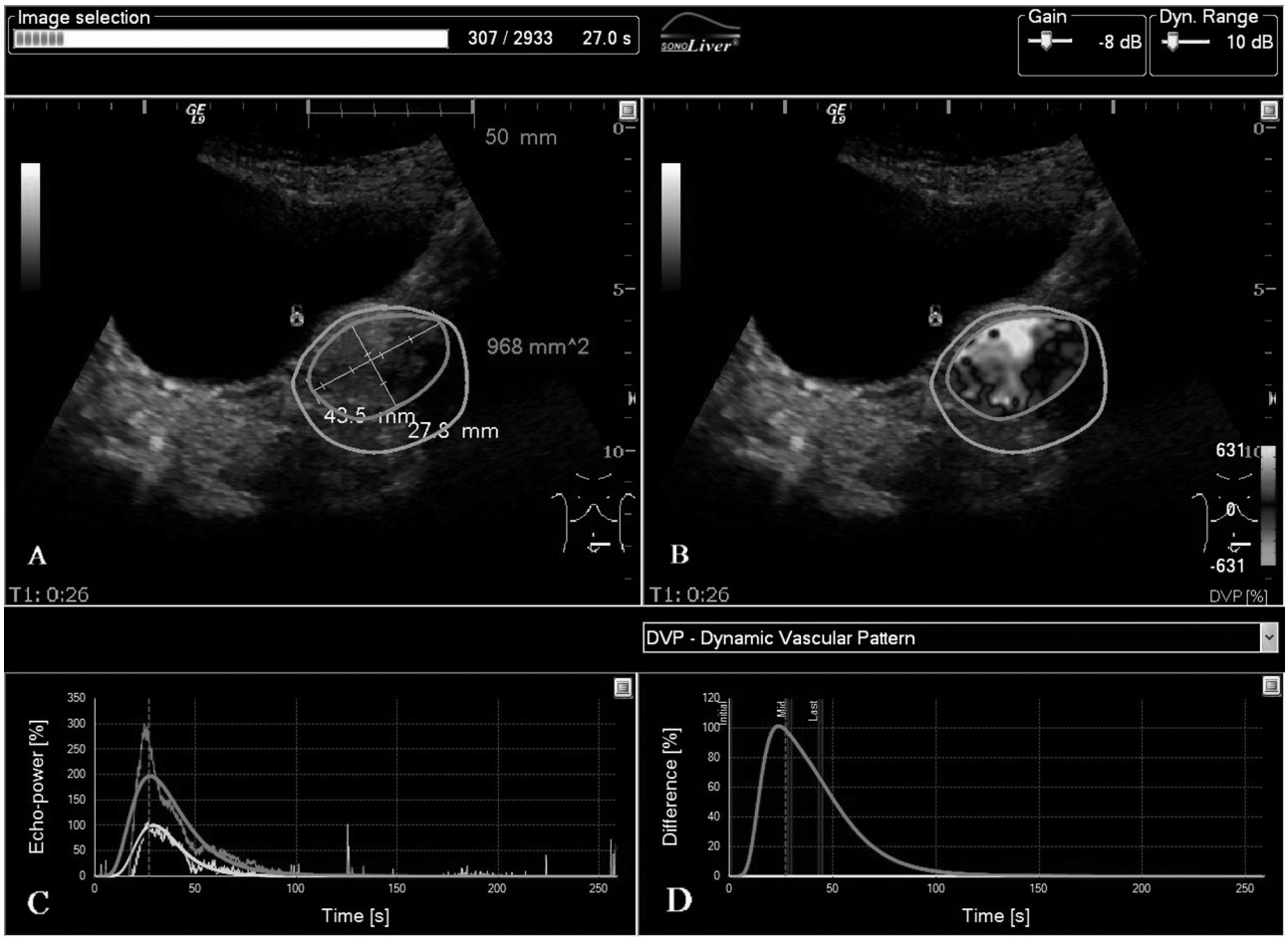

Figure 1. — Screenshot of Sonoliver at time to peak (TTP) of an epithelial ovarian cancer. (A)Three regions of interest (ROIs) are drawn: delimitation ROI (blue border), analysis ROI (green border), and reference ROI (the blue border minus the green border). (B) The DVP parametric images are interpreted according to warm-and-cold color. (C) Time-intensity curve (TIC). (D) DVP curve, with the healthy ovarian tissue taken as reference.

This study was approved by the Ethics committee, and written informed consent was obtained from each patient. All patients were scheduled for surgical treatment after evaluation by gynecologic oncologists and radiologists and were screened for contraindication, particularly pregnant or lactating women, interventional therapy or chemotherapy preoperative, cardiac shunts, recent history of thrombosis, and hypersensitivity to this ultrasound contrast agent.

Contrast-enhanced ultrasound examination

Patients were examined using Logiq 9 color Doppler ultrasonic diagnostic apparatus with a curved 4C transducer (between 1.0~4.0 MHz). An intravenous bolus injection within 3-5 s of 2.4 ml of SonoVue was used, which consists of microbubbles containing sulfur hexafluoride (SF6) gas encapsulated within a phospholipid monolayer shell, followed by a flush of five ml of saline.

First of all, a baseline US examination is recommended to scan the whole pelvic cavity and record the ovarian mass size, echogenicity, border, and color Doppler flow distribution. Then, determine the optimum scanning section and switch to Real-time CEUS mode with low mechanical index (MI) ranging from 0.1 to 0.15. As soon as the intravenous bolus injection of SonoVue, the real-time enhancement pattern of contrast agent inside the tumor was observed for three to five minutes and the imaging video was recorded in Dicom format for later analysis. Patients were required to maintain supine position and minimize inspiration extent the overall process.

Analysing of parametric imaging in contrast-enhanced ultrasound

The information contained in a dynamic sequence of enhancement can be exploited as a diagnostic aid by means of a new dedicated parametric image processing software SonoLiver quantitative software. This software generates a processed sequence by subtracting the mean pixel signal processing obtained in a reference region from the original pixel signal processing, a processing called dynamic vascular patterns (DVP) [9-11]. Such a processing can be used for enhancing the differences in perfusion kinetics between malignant and benign ovarian masses, which generates a sequence of images in warm and cold colors by subtracting the mean pixel-value obtained in a reference region from the original pixel values. DVP processing was designed to help clinicians to confirm their diagnoses of benign and malignant masses. The DVP parametric images were interpreted ac-

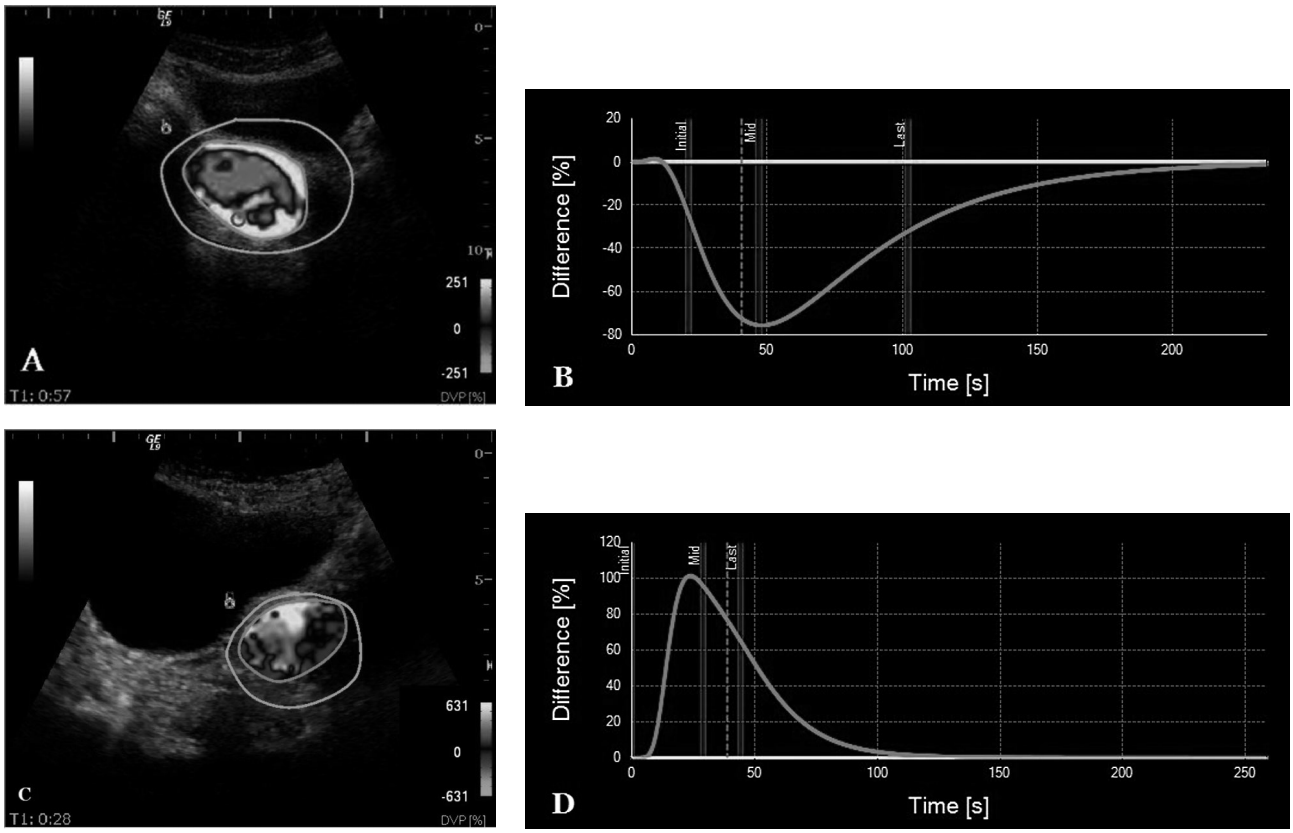

Figure 2. — (A) The DVP parametric images displays mainly hypo-enhanced with blue imaging in those with benign masses. (B) DVP curves are unipolar below the baseline in benign masses. (C) The DVP parametric images display mainly hyper-enhanced imaging with red in those with malignant masses. (D) DVP curves are unipolar above the baseline in malignant masses.

cording to warm-and-cold color (dark blue - light blue – yellow – orange - red), indicating that the perfusion intensity increased in turn (Figure 1B).

The SonoLiver software was designed for real-time evaluation of tissue perfusion obtained by CEUS examination, which also provides an objective quantification of perfusion parameters. Three regions of interest (ROIs) are outlined in the angiogram: (1) delimitation ROI, a blue border delimiting the region of the entire region to be analysed; (2) analysis ROI, a green border sketching out the contours of suspicious lesions; (3) reference ROI, the blue border minus the green border region-wide (Figure 1A). Then the automatic motion compensation of the software was used to correct the respiratory motions. At the same time, the system automatically drew out the time-intensity curve (TIC), from which a series of semi-quantitative perfusion parameters is extracted and analysed, and the DVP curve, with the healthy ovarian tissue taken as reference (Figures 1C, D).

The parameters obtained in this study included: time to peak (TTP); maximum intensity (IMAX), which was the percentage ratio of intensity of ROIs in lesions and ROI reference at the highest point of the perfusion process; rise time (RT), from 10% to 90% of IMAX; mean transit time (mTT) corresponding to the center of gravity of best-fit function of echo-power.

DVP parametric images were read by a clinician experienced in ultrasound who was blinded to the nature of the lesions. Finally, diagnoses made in this way were compared against the biopsy to calculate efficacy scores.

Statistical Analysis

Statistical analyses were carried out using SPSS version 13.0 . The differences of measurement data were compared with the t test while the counting data were using Chi-square test. All data were described as mean ± standard deviation. A value of $p < 0.05$ indicated statistical significance.

Results

Most benign ovarian masses are cystic, with clear borders. The performance for malignant masses are mainly mixed or solid, with irregular borders. In the group of benign ovarian masses, the mean diameters before CEUS (7.2 ± 3.4 cm) and after CEUS (7.4 ± 3.6 cm) were not statistically significantly different ($p = 0.398$). In the group of malignant ovarian masses, the mean diameters was significantly higher ($p = 0.006$) after CEUS (9.7 ± 3.0 cm) than before CEUS (8.5 ± 2.9 cm).

The majority of benign tumor blood vessels located peripherally in focus, with regular figure, and the intra-tumor with no enhancement or homogeneous enhancement. On the contrary, blood vessels in malignant tumors located penetrating or central and became coarser, with increasing tortuosity, branching patterns, vascular loops, and shunts.

Table 1. — *Quantitative parameters in the ROIs of 50 cases of ovarian masses ($\bar{x} \pm s$).*

Pathology	Number	IMAX (%)	RT (s)	TTP (s)	mTT (s)
Benign	28	67±32	44±11	65±19	154±52
Malignant	22	365±125	23±10	28±11	74±29
t Value		-12.170	7.265	8.364	5.269
p Value		0.000	0.813	0.042	0.006

ROI: region of interest, IMAX: maximum intensity, RT: rise time, TTP: time to peak, mTT: mean transit time.

In CEUS imaging of 50 cases, there were 24 cases (86%) displaying mainly hypo-enhanced with blue imaging in those with benign masses, including: four cases of internal non-enhanced imaging (two cases of inflammatory encapsulated fluid showed no internal enhancement and two cases of endometrial cysts showed ring-like enhancement in cystic wall) (Figure 2A). What is interesting is the mainly hyper-enhancement with red imaging in four cases of benign masses. They were, respectively, two cases of teratoma and two cases of ovarian fibroma. Meanwhile, there were 15 cases (68%) displaying mainly hyper-enhanced imaging with red and seven cases (32%) displaying mainly hypo-enhanced imaging with blue in those with malignant masses (Figure 2C). The difference was statistically significant ($p < 0.05$).

Twenty-three (82%) of the 28 benign masses were correctly classified as benign on the basis of that the DVP curves were unipolar below the baseline while malignant ones were opposite (Figure 2B). That is, 15 cases (68%) of malignant masses, DVP curves were unipolar above the baseline (Figure 2D). Respectively, the other five cases (18%) of benign masses were two cases of teratoma, one case of fibroma, two cases of corpus luteum cyst hemorrhage, of which DVP curves were unipolar above the baseline. Moreover, the other six cases (27%) of malignant masses were above the baseline at the perfusion period and early clearance, and reduced to below the baseline during the middle-late clearance. One case (5%) of malignant mass was unipolar below the baseline during the whole process of the CEUS.

IMAX, TTP, and mTT were all significantly higher in those with malignant masses than those with benign ones (all $p < 0.05$), but, no statistical difference in the RT between the two groups was found ($p > 0.05$) (Table 1).

Discussion

The significant differences of vascular anatomy and hemodynamics between the benign and malignant ovarian masses supplied pathophysiological basis of imaging studies. Use of CEUS has fully revealed the malignant characteristics of tumor angiogenesis and reflected the enhancing process of real-time dynamic observation of tumor microvessel perfusion on the pathological basis of characteristic of blood vessels inside the tumour, which has proved the discrimination between benign and malignant tumors [11-14]. In the present study, the authors have demonstrated that tumor angiogenesis can be visualized by targeted contrast-enhanced ultrasound imaging using ultrasound contrast agent microbubbles and related quantitative parameters were extracted from TIC. Meanwhile, TIC from ROIs generated a complex shape that was applied to tumours in an attempt to compare the echoes in analysis ROI and reference ROI. At the same time, the DVP parametric images read by Sonoliver software, which can be used as a tool for enhancing the differences in perfusion kinetics between focal lesions and normal ovarian organs.

In the present study, vascularity and enhancement patterns were intuitively reflect by warm-and-cold color (warm color stands for hyper-enhancement, cold color indicates the opposite). The majority of DVP parametric images showed hyper-enhancement in malignant masses (68%, 15 of 22) and hypo-enhancement in benign masses (86%, 24 of 28). This observation is consistent with early reports, indicating that malignant overian masses which had abundant blood stream would lead to large flow of ultrasound contrast agent microbubbles, whereas benign overian masses had light vascularity. However, in the above cases, the circle enhancement was shown in the cystic wall, which was different from that in malignant tumors. As a result, the DVP parametric images of benign and malignant ovarian masses seldom partially overlapped .

With regards to hyper-enhancement with red imaging in four cases of benign masses, two cases were teratomas, attributing to the increase of blood supply from increased thyroid and glial elements, angiogenesis. However, the other two cases with hyper-enhancement of benign masses were ovarian fibroma, of which enhancement was flocculent, sparse and slow. This was different from that of malignant masses. Concerning the hypo-enhanced imaging with blue, perhaps these were associated with liquefactive necrosis of malignancy. The aforementioned show the limitations in the present study. That is, some benign ovarian tumors with abundant blood supply might show the same ultrasound characteristics as the malignant tumors. In addition, although there were morphological and distributional difference of microvessels between benign and malignant lesions, their number and enhanced performance might overlap.

Difference between the original flow signal and the reference flow signal were obtained by DVP curve and reflected different vascular signatures of overian masses compared with contrast agent uptake in adjacent tissues. In the present study, analysis of DVP curve of contrast enhancement dynamics confirmed that the malignant masses, which were unipolar above the baseline, were hyper-enhanced with rich blood vessels. Also, the hypo-enhanced were corresponding to unipolar below the baseline. Six cases of malignant masses with bipolar DVP curve illustrated that hyper-enhancement in the during the perfusion period were followed by hypo-enhancement during the mid-

dle-late clearance. Indirectly, the DVP curve reflected the intensity change of the contrast agent microbubbles and hemodynamic differences. These findings suggested that DVP parametric imaging may enhance the individual diagnostic confidence in the subjective analysis of overian masses.

A published study showed that RT and TTP were associated with the number of microbubbles considering RT and TTP reflected the contrast agent arrival velocity, IMAX reflected the blood flow, and mTT indirectly responded to the emptying process of the contrast agent [15]. In the present study, there was significant difference in TTP of ovarian masses in two histological gradings, which suggested that the number of microbubbles accessing the vascular bed of malignant masses is much more than that of benign ones and reflect the richness of the malignant vascular. Current data showed that there were significant differences between TTP and mTT of ovarian masses in both histological gradings. The major feature of malignant vascular tumor is arteriovenous fistula, vascular deformation, arteriovenous shunt, multi-branch blood supply leading to large flow, and fast speed of microbubbles. Furthermore, current data showed that there were no significant differences in RT between both histological gradings, perhaps associated with less samples.

Time-intensity curves may provide an objective measure to demonstrate the more rapid enhancement of tumor relative to normal parenchyma. Three-dimensional presentation and other postprocessing image enhancements may increase the conspicuity of cancers. In the final analysis, a simplified protocol will be needed with clear enhancement of malignant foci if enhanced transrectal sonography is to be generally applied in screening for prostate cancer.

The ability to identify malignant tumor with a noninvasive method is important to decide the need for surgery and on the type of surgery required. The present pilot study demonstrated that the DVP curve contained a wealth of quantitative information and objectively reflected distribution of contrast agent in the normal and diseased tissue and the dynamic changes of perfusion. In conclusion, DVP parametric imaging is a new approach capable of differential diagnoses of ovarian masses with contrast-enhanced US.

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