

Retrospective Study

Value of ultrasound and magnetic resonance imaging combined with tumor markers in the diagnosis of ovarian tumors

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Grade E (Poor): 0**P-Reviewer:** Hirota Y, Japan; Mier W, Germany**Received:** August 23, 2023**Peer-review started:** August 23, 2023**First decision:** September 13, 2023**Revised:** September 21, 2023**Accepted:** October 25, 2023**Article in press:** October 25, 2023**Published online:** November 6, 2023**Qian Yang, Hui Zhang, Hai-Bao Wang**, The First Affiliated Hospital of Anhui Medical University, Hefei 230022, Anhui Province, China**Qian Yang**, Taihe Hospital of Traditional Chinese Medicine, Fuyang 236000, Anhui Province, China**Pei-Qi Ma, Bin Peng**, Fuyang People's Hospital, Fuyang 236000, Anhui Province, China**Gui-Tao Yin**, No. 2 People's Hospital of Fuyang City, Fuyang 236000, Anhui Province, China**Nan-Nan Zhang**, Linquan People's Hospital, Fuyang 236000, Anhui Province, China**Corresponding author:** Hai-Bao Wang, MD, PhD, Chief Physician, Professor, The First Affiliated Hospital of Anhui Medical University, No. 218 Jixi Road, Hefei 230022, Anhui Province, China. wang23415@126.com**Abstract****BACKGROUND**

Compare the diagnostic performance of ultrasound (US), magnetic resonance imaging (MRI), and serum tumor markers alone or in combination for detecting ovarian tumors.

AIM

To investigate the diagnostic value of US, MRI combined with tumor markers in ovarian tumors.

METHODS

The data of 110 patients with ovarian tumors, confirmed by surgery and pathology, were collected in our hospital from February 2018 to May 2023. The dataset included 60 cases of benign tumors and 50 cases of malignant tumors. Prior to surgery, all patients underwent preoperative US and MRI examinations, as well as serum tumor marker tests [carbohydrate antigen 125 (CA125), human epididymis protein 4 (HE4)]. The aim of the study was to compare the diagnostic performance of these three methods individually and in combination for ovarian tumors.

RESULTS

This study found statistically significant differences in the ultrasonic imaging characteristics between benign and malignant tumors. These differences include echo characteristics, presence or absence of a capsule, blood flow resistance index,

clear tumor shape, and blood flow signal display rate ($P < 0.05$). The apparent diffusion coefficient values of the solid and cystic parts in benign tumors were found to be higher compared to malignant tumors ($P < 0.05$). Additionally, the time-intensity curve image features of benign and malignant tumors showed significant statistical differences ($P < 0.05$). The levels of serum CA125 and HE4 in benign tumors were lower than those in malignant tumors ($P < 0.05$). The combined use of US, MRI, and tumor markers in the diagnosis of ovarian tumors demonstrates higher accuracy, sensitivity, and specificity compared to using each method individually ($P < 0.05$).

CONCLUSION

US, MRI, and tumor markers each have their own advantages and disadvantages when it comes to diagnosing ovarian tumors. However, by combining these three methods, we can significantly enhance the accuracy of ovarian tumor diagnosis, enabling early detection and identification of the tumor's nature, and providing valuable guidance for clinical treatment.

Key Words: Ovarian tumors; Ultrasound; Magnetic resonance imaging; Tumor markers; Differential diagnosis

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Core Tip: Ultrasound, magnetic resonance imaging, and tumor markers each have their own advantages and disadvantages when it comes to diagnosing ovarian tumors. However, by combining these three methods, we can significantly enhance the accuracy of ovarian tumor diagnosis, enabling early detection and identification of the tumor's nature, and providing valuable guidance for clinical treatment.

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INTRODUCTION

In recent years, there has been a steady increase in the occurrence of ovarian tumors, with a trend towards affecting younger age groups[1]. Benign tumors generally have a positive prognosis and are often treated through surgical resection. On the other hand, malignant tumors have a high fatality rate and pose a significant risk to the lives of patients, resulting in a poor prognosis. Early identification of whether an ovarian tumor is benign or malignant is crucial for patients to make informed decisions about treatment options and to improve their overall prognosis[2].

Due to the subjective nature of gynecological examinations and the challenges in accurately assessing surrounding infiltration, various auxiliary examination methods have been developed for diagnosing ovarian tumors[3]. Non-invasive imaging techniques such as conventional ultrasound (US), three-dimensional US, color and power Doppler, computed tomography, magnetic resonance imaging (MRI), and positron emission tomography are commonly used. While these techniques have significantly improved the diagnostic rate of ovarian tumors, each has its limitations when used alone[4, 5]. US is widely used in clinical diagnosis and treatment due to its simplicity, speed, affordability, and portability. Real-time US, in particular, is often employed for localized puncture of peritoneal effusion, providing relief to patients and aiding in disease diagnosis[6]. However, US can be affected by factors such as intestinal gas, lung gas, far-field and near-field attenuation, and artifacts, which may lead to misdiagnosis. On the other hand, MRI is extensively used for diagnosing other pelvic diseases due to its stable image quality and ability to clearly reveal signs of metastasis. MRI images provide a clear depiction of tissue structures, accurate location, and intuitive signs of lesion metastasis by examining the relationship with surrounding tissues. This information serves as valuable evidence for selecting appropriate clinical staging and surgical methods[7,8]. Currently, there are several main methods used in clinical practice to screen for ovarian cancer. One of these methods is gynecological pelvic examination, which involves physically examining the ovaries. However, this method has some limitations. It is subjective and dependent on the expertise of the examiner. Additionally, it is challenging to determine the nature of tumors solely through this examination[9]. On the other hand, imaging techniques have advanced and can detect most ovarian tumors. However, interpreting the imaging features of the ovaries is complex, and there is still a lack of specific and sensitive indicators to distinguish between benign and malignant ovarian tumors[10,11]. This is particularly true when distinguishing between benign ovarian tumors and early-stage ovarian cancer. The interpretation of imaging results is also influenced by subjective factors. In comparison, tumor marker detection is a more objective method and has a wide range of applications in screening, diagnosing, evaluating, and monitoring ovarian cancer[12].

The most commonly used ovarian tumor markers in clinical practice are carbohydrate antigen 125 (CA125) and human epididymis protein 4 (HE4)[13]. While CA125 has high sensitivity, its diagnostic specificity is poor. An increase in serum CA125 can be found in many tumors, including breast cancer, endometrial cancer, prostate cancer, and some gastrointestinal tumors. Additionally, benign female reproductive system diseases like endometriosis and reproductive system

inflammation, as well as pregnancy and menstrual periods, may also cause a certain degree of CA125 increase[14]. Therefore, CA125 alone is not suitable as an independent indicator for ovarian cancer screening. However, it is highly suitable as a monitoring indicator for ovarian cancer treatment. Further exploration is needed to determine how CA125 can be combined with other diagnostic indicators, such as laboratory and imaging indicators, to improve diagnostic specificity[15]. HE4 is a well-researched ovarian tumor marker that has gained significant attention in recent years[16]. Multiple studies have revealed that HE4 is highly expressed in ovarian cancer tissues, while its expression in normal ovarian tissues is almost negligible. The expression of HE4 in ovarian cancer cells is not only associated with histopathology but also with the degree of tumor differentiation. Current research indicates that combining CA125 and HE4 as markers significantly enhances the sensitivity and specificity in predicting ovarian malignancies. This combination is particularly effective in cases where the serum expression trends of CA125 and HE4 are opposite, reducing the chances of missed diagnoses and misdiagnosing certain benign diseases with high CA125 expression as ovarian cancer[17].

This study aims to evaluate the diagnostic efficiency and value of preoperative US and MRI examinations, as well as serum tumor markers (CA125, HE4), for different types of ovarian cancer patients. The study will compare the diagnostic performance of US, MRI, and serum tumor markers alone or in combination for detecting ovarian tumors.

MATERIALS AND METHODS

General information

From February 2018 to May 2023, a total of 110 patients with ovarian tumors were selected as the subjects of our study. These patients were confirmed to have tumors through surgery and pathology, and their complete clinical and imaging data were available. Among them, 60 cases were diagnosed with benign tumors and 50 cases were diagnosed with malignant tumors. The age range of patients with benign tumors was between 24 and 70 years, with an average age of (50.22 ± 6.13) years. Patients with malignant tumors had an age range of 25 to 73 years, with an average age of (52.16 ± 5.89) years. The comparison of general data between patients with benign and malignant tumors did not show any statistical significance ($P > 0.05$). The use of patient's tissues was reviewed and approved by the Ethics Committee of the First Affiliated Hospital of Anhui Medical University. The consent was obtained from all patients before specimen collection.

Inclusion and exclusion criteria

Inclusion criteria: (1) The image collection is comprehensive and clear, fulfilling the clinical diagnostic criteria for the disease; (2) The ovarian tumor is unilateral, consisting of both cystic and solid components, with the solid tissue showing enhancement; and (3) The clinical data is complete, and the patient has provided informed consent by signing an agreement. Exclusion criteria: (1) The artifact is large and the image quality is poor; (2) Patients with contraindications such as MRI and US; and (3) Patients with radiotherapy and chemotherapy before examination.

Ultrasonic examination

The Canon 790 color Doppler ultrasonic diagnostic instrument with a probe frequency of 3-5 MHz is used for this examination. The patient should lie supine on the examination table after filling the bladder. The probe is placed at the pubic symphysis in the patient's lower abdomen for scanning. During the scan, the position, shape, size, capsule, and echo of the ovary and tumor are observed. Additionally, the color Doppler flow imaging (CDFI) is used to visualize the inside of the tumor, surrounding blood flow shape, blood flow distribution, and to measure the blood flow resistance index.

MRI plain scan

The Canon 3T MRT-3010 magnetic resonance scanner was utilized for the examination, along with a 6-channel phased array coil. To minimize respiratory artifacts, sandbags were placed on the abdomen. Fast spin echo sequences were employed. For diffusion-weighted imaging (DWI), cross-sectional scanning and single-shot echo-planar imaging were adopted. The DWI image obtained after scanning was transmitted to the workstation to generate an apparent diffusion coefficient (ADC) map. The region of interest (ROI) was positioned in both the solid and cystic parts of the lesion, and the ADC value was measured. In addition, dynamic contrast-enhanced MRI (DCE-MRI) was performed. Initially, a T1-vibe-fs scan was conducted, followed by the injection of the contrast agent gadopentetate dimeglumine. The injection was administered through the middle cubital vein at a rate of 4 mL/s using a high-pressure syringe, with a rinse of 0.9% normal saline in 20 mL. Subsequently, dynamic enhanced scanning was performed. The collected images were imported into the post-processing workstation, where the ROI was manually outlined in both the solid and cystic parts of the tumor. The time-signal intensity curve (TIC) was automatically generated, and the TIC type was recorded. The types were classified as type I (inflow type), type II (platform type), and type III (outflow type).

Detection of serum tumor markers

Fasting peripheral venous blood 5 mL was collected within 24 h after admission, and 10 min was centrifuged at 3500 r/min speed. The supernatant was taken for examination. The following indexes were detected by chemiluminescence method: Serum CA125 level and HE4 level.

Observation index

(1) The study recorded the ultrasonographic characteristics of both benign and malignant tumors. These characteristics included the shape of the lesion, presence of a capsule, echo intensity, blood flow resistance index, and blood flow signal display rate; (2) MRI features include recording the ADC values of both the solid and cystic components of the tumor, as well as determining the type of time-intensity curve (TIC) after contrast enhancement; (3) The levels of serum CA125 and HE4 were compared between benign and malignant tumors; and (4) The diagnostic accuracy, sensitivity and specificity of US, MRI, serology and the combination of the three methods were recorded, in which CA125 > 35 U/mL was positive and HE4 > 140.0 pmol/L was positive. When one of the serological results was positive, it was judged to be positive. When one of the three methods is positive, it is judged to be positive.

Statistical method

Statistical analysis was conducted using SPSS 18.0 software. Count data were presented as the number of cases and percentage ($n\%$) and compared using the χ^2 test. Measurement data were presented as mean \pm SD, and the independent sample t -test was used for comparison. A statistically significant difference was considered when $P < 0.05$.

RESULTS

Comparison of ultrasonographic features between benign and malignant tumors

Table 1 presents the statistical analysis of the US features of ovarian benign and malignant tumors. The results indicate significant differences in the echo, capsule, blood flow resistance index, shape, and blood flow signal display rate between benign and malignant tumors ($P < 0.05$).

MRI feature comparison

Table 2 presents the ADC values of ovarian benign and malignant tumors along with the statistics of dynamically enhanced TIC image features. The results indicate that the ADC values of the solid and cystic parts of malignant tumors were lower compared to benign tumors, and this difference was statistically significant ($P < 0.05$). Additionally, there were significant differences observed in the TIC image characteristics between benign and malignant tumors ($P < 0.05$).

Comparison of serum tumor markers between benign and malignant tumors

The statistics of serum tumor markers for ovarian benign and malignant tumors are presented in **Table 3**. The results indicate that the levels of serum CA125 and HE4 were significantly higher in patients with malignant tumors compared to those with benign tumors. This difference was found to be statistically significant ($P < 0.05$).

Comparison of diagnostic efficacy of different methods in ovarian tumors

There was no significant difference in the diagnostic accuracy, sensitivity, and specificity of US, MRI, and tumor markers for ovarian tumors ($P > 0.05$). However, when US, MRI, and tumor markers were combined, the diagnostic accuracy, specificity, and sensitivity for ovarian tumors were higher compared to using a single method of detection ($P < 0.05$, see **Tables 4** and **5**).

DISCUSSION

Ovarian tumors can be classified into benign and malignant based on their characteristics. Benign tumors are usually asymptomatic and may be discovered during routine gynecological examinations. Malignant tumors are often asymptomatic in the initial stages but eventually present symptoms such as abdominal distension, abdominal mass, and ascites[18]. While some ovarian malignant tumors may be detected during physical examinations, most patients seek medical attention after experiencing typical symptoms. Unfortunately, by this time, the majority of cases have already progressed to advanced stages, resulting in a poor prognosis. Therefore, early detection and targeted treatment are crucial for improving the prognosis of patients with ovarian tumors[19].

Currently, there are several diagnostic methods available for ovarian tumors, including US, CT, MRI, and serum tumor marker detection[20]. Among these methods, US is widely used due to its affordability, simplicity, and reliable results. However, the deep location of the female ovary within the pelvic cavity poses challenges in clearly visualizing small ovarian tumors using transabdominal ultrasonography. Factors such as exploration depth, intestinal cavity inflation, and bladder reflections can hinder the clear display of small tumors and the fine internal structures, thereby affecting the accuracy of ovarian tumor diagnosis[21]. In the differential diagnosis of benign and malignant ovarian tumors, ultrasonography can be used to observe tumor hemodynamic parameters and other information through CDFI examination, which aids in the diagnosis of these tumors[22]. This study identified distinct differences in US images between benign and malignant ovarian tumors. For instance, malignant tumors are highly invasive and grow rapidly, often exhibiting abundant new blood vessels and fast blood flow. In contrast, benign tumors grow slowly, have fewer blood vessels, and typically lack blood flow or show slow blood flow. Evaluating the vascular features of ovarian tumors through ultrasonographic examination can provide valuable assistance in distinguishing between benign and malignant tumors[23].

Table 1 Ultrasonographic characteristics of benign and malignant tumors

	Echo		Capsule		Blood flow resistance index	Form		Blood flow signal display rate
	Rules	Irregularity	Yes	No		Clear	Not clear	
Benign tumor	55	5	46	14	0.79 ± 0.05	54	6	15 (25%)
Malignant tumor	8	42	12	38	0.44 ± 0.49	5	45	50 (100%)
χ^2/t	63.809		30.350		24.010	70.189		63.462
<i>P</i> value	0.000		0.000		0.000	0.000		0.000

Table 2 Comparison of apparent diffusion coefficient value (mm²/s) and time-signal intensity curve image features [*n* (%)] between benign and malignant tumors

	Solid part ADC value	ADC value of cystic part	TIC image features		
			I type	II type	III type
Benign tumor	1789.74 ± 122.53	2799.33 ± 89.88	46 (76.67)	14 (23.33)	0 (0.00)
Malignant tumor	867.67 ± 15.87	2260.03 ± 91.75	0 (0.00)	4 (0.08)	46 (92.00)
χ^2/t	33.353	20.615	65.885	4.685	94.875
<i>P</i> value	0.000	0.000	0.000	0.030	0.000

ADC: Apparent diffusion coefficient; TIC: Time-signal intensity curve.

Table 3 Serum tumor markers in patients with benign and malignant tumors

	<i>n</i>	CA125 (U/mL)	HE4 (pmol/L)
Benign tumor	60	22.67 ± 4.57	75.33 ± 9.84
Malignant tumor	50	397.60 ± 180.02	298.70 ± 40.66
<i>t</i>		-11.462	-28.982
<i>P</i> value		0.000	0.000

CA125: Carbohydrate antigen 125; HE4: Human epididymis protein 4.

Table 4 Comparison of diagnostic results and pathological results of three examination methods for ovarian tumors

	<i>n</i>	US		MRI		Tumor marker		Joint detection method	
		Benign	Malignant	Benign	Malignant	Benign	Malignant	Benign	Malignant
Benign tumor	60	48	12	51	9	49	11	59	1
Malignant tumor	50	12	38	8	42	4	46	0	50
Total	110	60	50	59	51	53	57	59	51

US: Ultrasound; MRI: Magnetic resonance imaging.

MRI, which does not involve ionizing radiation, offers the advantage of imaging in multiple planes and directions, as well as providing high soft tissue resolution[24]. As a result, it has become a reliable method for diagnosing ovarian tumors. However, it is important to note that MRI examinations can be time-consuming and noisy. Additionally, patients with birth control rings or metal foreign bodies may not be suitable for MRI examinations[25]. In recent years, DWI, a high-field MRI sequence, has gained popularity in clinical practice. It allows for the observation of microscopic movement of water molecules in living tissues by detecting the diffusion of water molecules[26]. Moreover, DWI can also quantitatively analyze vascular permeability and blood volume using ADC. Another useful technique, DCE-MRI, relies on the pharmacokinetic characteristics of contrast agents to measure blood perfusion and outflow in lesions. It also enables dynamic observation of the entire enhancement process and provides valuable information on tumor blood supply. Fur-

Table 5 Comparison of diagnostic efficiency of different methods

Method	Diagnostic accuracy (%)	Diagnostic sensitivity (%)	Diagnostic specificity (%)
US	78.18 (86/110)	80.00 (48/60)	76.00 (38/50)
MRI	84.55 (93/110)	85.00 (51/60)	84.00 (42/50)
Tumor marker	86.36 (95/110)	81.67 (49/60)	92.00 (46/50)
Joint detection method	99.09 (109/110)	98.33 (59/60)	100.00 (50/50)
χ^2	22.705	16.590	30.303
<i>P</i> value	0.000	0.001	0.000

US: Ultrasound; MRI: Magnetic resonance imaging.

Furthermore, DCE-MRI has the ability to comprehensively evaluate tumor morphology and dynamics. Whole abdominal MRI examinations can detect the presence of distant metastasis in malignant tumors. It is worth noting that the ADC values of benign tumors tend to be higher than those of malignant tumors, particularly in the solid and cystic parts. This paragraph discusses the relationship between the rapid proliferation of malignant tumor cells, high cell density, and the limited diffusion movement of water molecules, which results in a reduction in the ADC value[27,28]. The occurrence and development of tumors rely on the formation of new blood vessels, and analyzing the angiogenesis mechanism and process of tumor tissue can help evaluate the tumor status. The TIC image features of benign and malignant tumors differ. Benign tumors typically exhibit type I (inflow type) and type II (platform type) curves, while malignant tumors mostly display type II (platform type) and type III (outflow type) curves. These findings align with the pathological characteristics of benign and malignant lesions[29]. The absence of a prominent enhancement peak in the type I curve suggests a benign lesion. The maximum enhancement peak in the type II curve is lower than that of the myometrium, indicating a borderline lesion. On the other hand, the type III curve exhibits a high enhancement peak in the myometrium, indicating a malignant lesion. This highlights the significant role of DCE-MRI in distinguishing between benign and malignant ovarian tumors[30].

Serum tumor markers, such as CA125 and HE4, are widely used in the clinical diagnosis of ovarian cancer[31]. CA125 is a broad-spectrum marker commonly used in gynecological tumors. Its level is associated with the size of the tumor and the amount of antigen produced by it. When cancer cells invade tissues like the uterus, fallopian tubes, and intrahepatic bile ducts, they disrupt intercellular connections and basement membranes. This leads to the activation and release of a significant amount of CA125 into the bloodstream, resulting in a notable increase in serum CA125 levels. In ovarian tumors, even when there are no obvious symptoms or difficulties in pathological identification, a significant rise in CA125 levels indicates a malignant lesion and serves as a highly sensitive indicator for diagnosing ovarian cancer[32]. HE4 was initially discovered in human epididymal epithelial cells. Subsequent studies have revealed its abundant expression in ovarian cancer, particularly in serous ovarian cancer and endometrioid cancer[33]. HE4 plays a crucial role in the diagnosis of ovarian cancer, disease detection, and postoperative recurrence detection. It holds significant clinical value in distinguishing between benign and malignant tumors, and when combined with CA125, it can serve as an early screening indicator for ovarian cancer. Notably, the levels of serum CA125 and HE4 were found to be significantly lower in patients with benign tumors compared to those with malignant tumors[34].

US, MRI, and serum tumor markers each have their own advantages and disadvantages in diagnosing ovarian tumors. The combined use of these three methods in diagnosing ovarian tumors has shown significantly higher diagnostic accuracy, sensitivity, and specificity compared to using a single method alone. This indicates that the combined application of these three methods can complement each other, aiding in localizing and qualitatively diagnosing ovarian tumors, detecting them early, and distinguishing between benign and malignant tumors[35]. These findings provide valuable guidance for clinical treatment.

CONCLUSION

Ultrasonography is a valuable tool in diagnosing ovarian tumors as it provides information about their location, internal structure, and blood flow characteristics. It is capable of making definite and differential diagnoses for most ovarian tumors. MRI serves as a supplementary imaging method to US, enhancing the diagnostic value of ovarian tumors when used in combination. Although serum tumor markers alone cannot be used for localizing tumors, their combined application with US and MRI improves the sensitivity and specificity of ovarian tumor diagnosis. This combined approach is particularly useful for preliminary screening, early diagnosis, and differential diagnosis of benign and malignant tumors. Overall, the integration of these three methods contributes to the early detection and accurate differentiation of ovarian tumors, making them valuable in clinical practice.

ARTICLE HIGHLIGHTS

Research background

Ultrasound (US), magnetic resonance imaging (MRI), and serum tumor marker detection are currently effective clinical tools for diagnosing ovarian cancer. However, there are currently limited studies that investigate their individual or combined use for detection.

Research motivation

This study aimed to investigate the diagnostic value of US, MRI, and tumor marker detection alone or in combination for ovarian tumors.

Research objectives

Comprehensive comparison of US, MRI combined with tumor markers in the diagnosis of ovarian tumors.

Research methods

A total of 110 ovarian cancer patients were selected as research subjects from our hospital, spanning from February 2018 to May 2023. These patients were confirmed to have ovarian cancer through surgery and pathology, with 60 cases being benign tumors and 50 cases being malignant tumors. Prior to surgery, all patients underwent preoperative US and MRI examinations, along with serum tumor marker testing for carbohydrate antigen 125 (CA125) and human epididymis protein 4 (HE4).

Research results

This study investigated the differences in ultrasound imaging characteristics between benign and malignant tumors. The study found that there were statistically significant differences in echogenic characteristics, presence or absence of capsule, blood flow resistance index, clear tumor shape, and blood flow signal display rate ($P < 0.05$). The apparent diffusion coefficient values of the solid and cystic parts of benign tumors were observed to be higher than those of malignant tumors ($P < 0.05$). Moreover, significant statistical differences were found in the time-intensity curve image features of benign and malignant tumors ($P < 0.05$). The levels of serum CA125 and HE4 were found to be lower in benign tumors compared to malignant tumors ($P < 0.05$). The combined use of ultrasound, MRI, and tumor markers resulted in higher accuracy, sensitivity, and specificity in diagnosing ovarian tumors than using each method alone ($P < 0.05$).

Research conclusions

The use of US, MRI, and tumor markers in diagnosing ovarian tumors has both advantages and disadvantages. However, combining these three methods can greatly enhance the accuracy of diagnosis, facilitate early detection, identify the nature of the tumor, and offer valuable guidance for clinical treatment.

Research perspectives

The early detection and targeted treatment are crucial for improving the prognosis of patients with ovarian tumors.

FOOTNOTES

Author contributions: Yang Q and Zhang H contributed to the conceptualization of this study; Ma PQ, Peng B, Yin GT involved in the methodology of the manuscript; Zhang NN took part in the formal analysis; Wang HB contributed to the investigation; Yang Q prepared the original draft; Wang HB involved in the writing-review and editing, and supervision; and all authors have read and agreed to the published version of the manuscript.

Institutional review board statement: This study was performed in line with the principles of the Declaration of Helsinki. Approval was granted by the Ethics Committee of the First Affiliated Hospital of Anhui Medical University.

Informed consent statement: The informed consent was obtained from all patients before specimen collection.

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Data sharing statement: The datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

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