Quantitative Analysis of Contrast-Enhanced Ultrasound in Evaluation of Preoperative Chemotherapy for Wilms Tumor in Children—Pilot Study

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Abstract

Background : Contrast enhanced ultrasound (CEUS) has been actively used in the detection of lesions in many organs especially in kidney and liver, but there is no report in evaluation of preoperative chemotherapy for Wilms tumor in children.

Methods: 8 children with Wilms tumor were enrolled in this study. All patients accepted preoperative chemotherapy for 4-8 weeks. Each child accepted CEUS examination for 3 times. The image files and the data were analyzed through software.

Results: The group included 8 cases with tumor volume from 155.71 to 1624.16 cm³. 6 patients accepted preoperative chemotherapy for 8 weeks before surgery, 2 cases underwent preoperative chemotherapy for 4 weeks. Each child accepted CEUS examination for 3 times and no one showed discomfort to the SonoVue contrast agent. By the comparison from the 3 times CEUS, tumor volume in most of the children was significantly reduced, and the blood perfusion intensity in the tumor was prominently weakened. The results of linear regression showed that the changes of PI between the first and second CEUS examinations and between the second and third CEUS examinations had correlation with the reduction volume of Wilms tumor.

Conclusions: It is the first time to apply quantitative analysis of CEUS in evaluation of preoperative chemotherapy for Wilms tumor in children. CEUS is a safe examination in children. CEUS videos could show clearly the changes of tumor perfusion during preoperative chemotherapy. PI can precisely reflect the degree of sensitivity of tumor to chemotherapy in early stage of preoperative chemotherapy.

Background

Wilms tumor, also known as nephroblastoma, is the most common urinary system malignant tumor in children^[1]. Chemotherapy plays a crucial role in the treatment of Wilms tumor. Whether Children's Oncology Group (COG) Renal Tumor Committee or International Society of Pediatric Oncology (SIOP) Renal Tumor Committee, has its preoperative chemotherapy protocols^[2,3]. Preoperative chemotherapy reduces the size of tumor and the length of tumor extension, probability of tumor rupture during the operation and the surgical complications are also reduced^[4]. Preoperative chemotherapy is usually recommended for 4-8 weeks in children, no more than 12 weeks^[3]. Evaluation of the effect of preoperative chemotherapy has been routinely repeated with conventional ultrasound (CUS), contrast-enhanced computed tomography (CECT) and magnetic resonance imaging (MRI)^[5,6]. Conventional ultrasound can measure the tumor size and distinguish cystic from solid in renal tissue, however, lower accuracy and incapable detection in microcirculation are limitations. The ionizing radiation from repeated CT when monitoring chemotherapy response is harmful to children. Expensiveness, time-consumption and potential nephrotoxicity of contrast agent are the limitations of MRI.

Recently, the development of microbubble contrast media and imaging techniques, contrast enhanced ultrasound (CEUS) has been actively used in the detection and differentiation of lesions in many organs especially in kidney and liver [7]. Under CUS, CEUS not only has the advantages of CUS but also can clearly show the microcirculation perfusion level of the tissue, and the precise analysis of tissue perfusion could be acquired by software. Ultrasound contrast agents (UCAs) are known to be well-tolerated and safe in adults. Because of no excreting through the kidneys, UCAs can be safely administered to patients with renal insufficiency with no risk of contrast-related nephropathy [8]. Dynamic observation of CEUS could provide an accurate characterization and microcirculation of tumor. However, there are no reports on the application of CEUS in pediatric Wilms tumors. Based on these advantages, CEUS may be with more values to assess preoperative chemotherapy for Wilms tumor in children. This study aimed to explore whether CEUS might be helpful to evaluate preoperative chemotherapy effect for Wilms tumor.

Methods

Patients' information and study design

This study and the using of SonoVue (a kind of UCAs) were approved by the Ethics Committee of Beijing Children's Hospital (National Center for Children's Health of China). Informed consent was signed with each child's parents.

From April to October in 2018, 8 children, diagnosed with Wilms tumor, were enrolled at the first visit to the department of Urology Surgery in Beijing Children's Hospital. All patients were unilateral tumors and diagnosed for the first time without any treatment before. All patients accepted preoperative chemotherapy with vincristine and actinomycin D for 4-8 weeks and no one underwent preoperative radiotherapy (**Table 1**). Each child accepted CEUS examination for 3 times, which respectively were before preoperative chemotherapy treatment, 3 weeks after chemotherapy treatment and before the nephrectomy. After the preoperative chemotherapy, radical nephrectomy was performed. All the pathological types were proved to be Wilms tumor finally.

CEUS examination and data analysis

CEUS was performed by two experienced chief physicians (N.W and X.M.W) in ultrasound specialty, who also was in the charge of film reading and data analysis. The IU 22 (Philips, Bothell, WA) type ultrasound system (frequency 2.5-7.0 MHz) was used not only in ordinary detection such as tumors size, boundary and echo features but also in contrast-enhanced ultrasound (CEUS) to observe blood microcirculation in and around renal tumors. The US contrast agent used in this study was SonoVue (Bracco, Milan, Italy) contrast agent (Bracco) was used, which consists of sulfur hexafluoride (SF₆)-filled microbubbles stabilized with phospholipids. First step, an ampoule of 5 mg of SonoVue should be shaken with 5 ml of normal saline to serve as a microbubble suspension. According to 0.03ml/Kg of the patient's weight, the volume of injection agent was extracted each time. The injection agent was quickly injected through the

superficial forearm vein, followed by 5ml saline immediately. And then, the tumor tissue and peripheral renal parenchymal perfusion were observed continuously and dynamically. When SonoVue was injected, the timer and video recorder were simultaneously activated. The conditions such as gain, depth and so on were kept unchanged during the procedure. Probe position was fixed, and the children were asked to keep quiet and breathe peacefully. If the children were too young to cooperate, chloral hydrate sedation was used to ensure the scanning plane to be stable. The tumor was observed continuously for more than 3 min. CEUS imaging data video clips for each detected tumor were saved in a hard drive for subsequent observation and analysis. After the examination, each child was kept in our hospital for 2 hours to observe whether there were any symptoms of discomfort.

After examination, the image files were export by DICOM format and the data was analyzed through Philips Q-Lab software. 5 regions of interest (region of interest, ROI) were selected in the tumor tissue to analyze. We chose a solid area in tumor tissue with a good blood perfusion to select the ROIs when the first-time examination on children before chemotherapy treatment. The following two times examination, we tried to keep the observation area of interest basically to be the same as the first time **Figure 1**. The time-signal intensity curve was plotted by Q-Lab software, and the CEUS parameters of tumor were selected for analysis. Parameters include: time to peak (TTP), wash in slope (WIS), peak intensity (PI).

Statistical analysis

Linear-regression analysis was used to compare the changing data of CEUS parameters with the reduced volume of the tumor. Continuous data were expressed as a percent or mean $\pm$ standard deviation (SD). $P < 0.05$ was considered significant. Statistical analysis was performed using SPSS software version 24.0.

Results

The 8 cases, with half of boys and girls, included 5 tumors in the right kidney, and 3 in the left. The group aged from 1.9 to 6.1 years (mean 3.6 ± 1.2 years); tumor size from $8.3 \times 6.5 \times 6.8$ to $12 \times 17 \times 16$ cm (length $\times$ width $\times$ height); tumor volume from 155.71 to 1624.16 cm³ (mean 628.45 ± 584.16 cm³). All patients had been found relatively large Wilms tumors, in which the smallest tumor volume was 155.71 cm³, and some tumors had ruptured in 6 cases (**Figure 4**). In order to completely remove tumors, preoperative chemotherapy needed to be performed in these patients. Because of a continuous decrease in the tumor volume, 6 patients accepted preoperative chemotherapy for 8 weeks before surgery. One child (**NO.2 case**) accepted nephrectomy only after 4 weeks chemotherapy because the tumor had shrunk significantly to a viable size for operation. With the tumor little shrinkage and relatively rich blood supply after 4 weeks chemotherapy, one patient **NO.8 case** underwent nephrectomy because the surgery is not very difficult (**Figure 2 and 3**). All children accepted 3 times CEUS during the process of preoperative chemotherapy, and no one showed discomfort to the SonoVue contrast agent. After the preoperative chemotherapy, all patients underwent radical nephrectomy. And the pathological types included 5 (62.5%) cases of mixed type, 1 (12.5%) cases of mesenchymal type, 1 (12.5%) case of germ type, 1 (12.5%) case of epithelial type. (**Table 1**)

All CEUS data was analyzed by Philips Q-Lab software. By the comparison from the 3 times CEUS, we found that the tumor volume of most of the children was significantly reduced, and the blood perfusion intensity in the tumor was prominently weakened. Only one case showed no significant tumor volume and blood perfusion reduction. The mean values of parameters (WIS, TTP, PI) from the 5 ROIs was calculated for each patient. The difference of parameters between the first and second CEUS examinations, the second and third CEUS examinations and the first and third CEUS examinations were calculated for each patient (**Table 2**). We used SPSS software version 24.0 to conduct linear regression analysis between each difference of parameters and tumors volume reduction in the group, to explore the correlations between changes of each parameter and tumor volume reduction after preoperative chemotherapy. The results showed in **Table 3**. The changes of PI between the first and second CEUS examinations and between the second and third CEUS examinations had correlation with the reduction volume of Wilms tumor (1st CEUS-2nd CEUS: $R=0.94; P=0.001<0.05$; 2nd CEUS-3rd CEUS: $R=0.75; P=0.031<0.05$). Although, during the second and third CEUS evaluations, the value of correlation (R value) between the changes in blood perfusion intensity and tumor volume shrinkage had decreased compared with the first and second CEUS. However, they still have a clear correlation (P value <0.05). In the whole process of preoperative chemotherapy, the P value of the correlation between PI and tumor shrinkage is greater than 0.05.

Discussion

Wilms tumor, also known as nephroblastoma, is the most common urinary system solid tumor in children, with the morbidity about 7.8 per million ^[9,10]. Because of the multidisciplinary approach of combined surgery, chemotherapy, and use of radiotherapy (RTH) when necessary, the cure rate in children has been dramatically improved during the last three decades. The latest report showed the five-year event-free survival (EFS) and overall survival were 77.2% and 87.5% ^[11]. Chemotherapy plays a crucial role in the treatment of nephroblastoma. SIOP protocol is used mainly in Europe and an essential part of it has been preoperative chemotherapy. On the North-American NWTs/COG protocols, there are some situations wherein preoperative chemotherapy is recommended, including children for whom renal-sparing surgery is planned, tumors inoperable at surgical exploration, and tumor extension into the IVC above the hepatic veins ^[12,13].

Traditional evaluation strategy of the effect of preoperative chemotherapy has been routinely measured the tumor size with CUS, enhanced CT and MRI. Conventional ultrasound can measure the tumor size and distinguish cystic from solid in tumor tissue, however, its lower accuracy and incapable of detection microcirculation are limitations. Contrast-enhanced CT was an important noninvasive modality in evaluating preoperative chemotherapy of Wilms tumor. The disadvantage of CT modality was an exposure to ionizing radiation. Because children are at the stage of rapid body development, their bodies and organs are more sensitive to the destructive effects of ionizing radiation comparing to adults. The carcinogenic effects of ionizing radiation from radiological diagnostic procedures are random. The probability of secondary malignancies increases with increasing radiation exposure and has a

cumulative effect.^[14] Contrast-enhanced MRI was also used in the diagnosis and characterization of renal tumors^[15,16]. However, its expensiveness, time-consumption and potential nephrotoxicity of contrast agent limited its use.

CEUS is an imaging examination that combines ultrasound and UCAs. SonoVue is a kind of UCAs, containing microcapsules filled with sulfur hexafluoride (SF₆) and composed of lipid-encapsulated microvesicles. Because microbubbles range in diameter from 1 to 10 µm, SF₆ is able to pass through the pulmonary bed, but cannot penetrate the vascular endothelium, so it is excreted by breathing rather than metabolized by the kidney^[17]. This feature of SonoVue enables real-time visualization of renal micro- and macro-circulation. In addition, CEUS tests have no risk of nephrotoxicity or nephrogenic systemic fibrosis (NSF), and there is no ionizing radiation^[18]. These characteristics make the examination appealing for patients with those requiring long-term, repeated, low radiation burden imageological examination. Therefore, CEUS is very suitable for children with nephroblastoma who need multiple texts to evaluate the effect of preoperative chemotherapy.

UCAs have a very low rate of anaphylactoid reactions (1:7000 patients, 0.014 %), significantly lower than the rate with iodinated state-of-the-art CT agents (35-95:100 000 patients, 0.035-0.095 %). Data from 75 completed studies showed that the most frequent adverse events were headache (2.1 %), nausea (0.9 %), chest pain (0.8 %) and chest discomfort (0.5 %)^[8]. All the children in our study signed the informed consent and stayed in the hospital for more than 2 hours after the injection of UCAs. None of them showed symptoms of discomfort.

There have been reports of CEUS applied to adult renal tumors. Chen^[19] and et al. evaluated the diagnostic value of CEUS in small renal cell carcinoma (RCC) and angiomyolipoma (AML). The study showed CEUS had value in diagnosing small RCC and AML and outperformed conventional ultrasound (CUS) in differentiation of small RCC and AML. Gulati^[20] and et al. evaluated the diagnostic value of CEUS in cystic and solid renal lesions, which showed CEUS with an emphasis on imaging features could different various cystic and solid renal lesions. CEUS has been applied in the evaluation and percutaneous treatment of hepatic and renal tumors^[7]. CEUS has advantages over CECT and CUS in assessing cystic renal mass. CEUS is superior to CUS and CECT in visualizing the number and thickness of cystic renal lesions and the composition of cystic renal mass. In the diagnosis of renal cystic lesions, CEUS is similar to CECT and better than CUS.^[21] Only a small number of studies were conducted by quantitative analysis of CEUS parameters. For example, Gerst^[22] and et al. used CEUS to evaluate renal masses by parameter comparison. This study showed ultrasound features of gray-scale heterogeneity, lesion washout, grade of contrast enhancement, and quantitative measure of PI may be useful for differentiating clear cell carcinoma and non-clear cell renal tumors. As far as we know, there is no research about applying CEUS in chemotherapy effects of renal tumors. Because of the importance of evaluating the effect of preoperative chemotherapy for children with Wilms tumor, it is significant to study quantitative analysis of CEUS in evaluation of preoperative chemotherapy in Children with Wilms tumor which may improve accuracy of the evaluation.

Two experienced chief physicians (N.W and X.M.W) in ultrasound specialty performed CEUS. The CEUS videos and data in this study were analyzed by the two physicians respectively, then they came to a conclusion that both of them supported it. The CEUS videos showed that the tumors' volume was significantly reduced and the blood perfusion in the tumor was significantly weakened in most of the children with the progress of preoperative chemotherapy. Particularly, some tumors showed liquefactive necrosis and the blood perfusion even disappeared (**Figure 5**). Simultaneous enhanced CT also confirmed this changes (**Figure 4**). However, one case (**N0.8**) was not very sensitive to chemotherapy, the tumor's volume did not change very much, and CEUS showed little significant reduction in tumor blood perfusion, which was also indicated by the quantitative analysis, the limited decrease in PI (**Figure 2 and 3**). Postoperative pathology indicated the diagnosis of mesenchymal type in Wilms tumor. Study showed that stromal type, including mesenchymal type, and epithelial type predominant tumors may demonstrate a poor clinical response to therapy but have an excellent prognosis if the tumor is completely excised ^[23].

In the stage of analysis of CEUS data, we chose a solid area in tumor tissue with an abundant blood perfusion to select the RIOs when the first-time examination on children before chemotherapy treatment. The following two times examination, we tried to keep the observation area of interest basically to be the same as the first time (**Figure1**). Only in this way can the stability of the selection of interest area be guaranteed, and the data can be compared between each CEUS examination.

It was very different in the tumor blood perfusion in these 8 cases. In some cases, SonoVue in tumor tissue could be cleared up in 3 minutes, while in others showed more than 5 minutes, so area under the curve (AUC) was not included as a parameter in this study. The difference of parameters (WIS, TTP, PI) between the first and second CEUS examinations, the second and third CEUS examinations and the first and third CEUS examinations were calculated for each patient (**Table 2**). Linear regression analysis showed the changes of PI between the first and second CEUS examinations and between the second and third CEUS examinations had correlation with the reduction volume of Wilms tumor. Experience in SIOP has shown that the majority of reduction (48%) in tumor volume occurs in the first 4 weeks of therapy but that reduction extends out through 8 weeks (62%) ^[24]. Therefore, when the second CEUS examination just 3 weeks after chemotherapy, most tumors were in the stage of significant shrinkage, partial tumor cells were necrotic under the action of chemotherapy drugs, and the blood microcirculation was decreasing observed by CEUS. Changes of blood perfusion may turn up earlier than tumor shrinkage during chemotherapy ^[25]. Therefore, detection of changes in blood microcirculation maybe more sensitive to evaluate the effect of tumor chemotherapy. Thus, CEUS could evaluate the effect of preoperative chemotherapy for Wilms tumor more sensitivity by showing the level of blood perfusion within tumor than traditional method. With the progress of preoperative chemotherapy, a large amount of necrosis has generated in the tumor tissue, so the degree of blood perfusion changing is also reduced than previous stage. At this point, with the absorption of the necrotic tissue, the tumor begins to shrink more significantly, however, the changes of PI became less obvious compared to the previous stage. This phenomenon may be related to early happening in change of tumor perfusion during the process of chemotherapy, and the less obvious changes of blood flow perfusion after the tumor has partially

necrosis during the later stage of chemotherapy. On the other hand, the number of cases in this study was small, and large sample size studies should be conducted in the future in order to explain the relationships more accurately.

Conclusion

It is the first time to apply quantitative analysis of CEUS in evaluation of preoperative chemotherapy for Wilms tumor in children. CEUS is a safe and valuable examination in evaluation of preoperative chemotherapy for Wilms tumor in children. High accuracy, no ionizing radiation and lower financial burden are the advantages of CEUS. CEUS videos could show clearly the changes of tumor perfusion during preoperative chemotherapy. Under the analysis of the software, one of the parameters, PI can precisely reflect the degree of sensitivity of tumor to chemotherapy in early stage of preoperative chemotherapy. Due to the small number of cases included in this study, there is no clear result on other parameters such as TTP and WIS. Further large sample size studies should be conducted in the future.

Declarations

Ethics approval and consent to participate

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. This study and the using of SonoVue (a kind of UCAs) were approved by the Ethics Committee of Beijing Children's Hospital (National Center for Children's Health of China). Informed consent was signed with each child's parents.

Consent for publication

Not applicable.

Availability of data and materials

The datasets generated and analyzed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

SX and NS conceptualized and designed the study, acquired, analyzed and interpreted data, drafted the initial manuscript. WPZ and HCS acquired, analyzed, coordinated and supervised data collection. NW and XMW, two experienced chief physicians in ultrasound specialty, had performed CEUS, who were also in charge of film reading and data analysis. DFL, PL, GNW, YY and SFC critically reviewed the manuscript. All authors approved the final manuscript as submitted and agree to be accountable for all aspects of the work.

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Not applicable.

Abbreviations

CEUS: contrast enhanced ultrasound; CUS: conventional ultrasound; CECT: contrast-enhanced computed tomography; MRI: magnetic resonance imaging; UCAs: ultrasound contrast agents; TTP: time to peak; WIS: wash in slope, PI: peak intensity; COG: Children's Oncology Group; SIOP: International Society of Pediatric Oncology.

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Tables

Table 1. The Baseline Characteristics of Patients

Sr	Age (year)	Tumor Size (Length*Width*Height) (cm)			Preoperative Chemotherapy Time (week)	Histological type
		before chemotherapy	3 weeks chemotherapy	before surgery		
a	3	17*12*16	15*10*12	12*10*12	8	mixed
	4.1	9.9*4.8*6.6	5*4*4	4*3*4	4	germ
	3.7	7.2*8.4*9	5.5*7*7.6	5*5.5*6.2	8	mixed
a	3.1	8.3*6.5*6.8	7.2*5.5*6.2	4.3*4.3*3.8	8	epithelial
	6.1	18*12*13	16*10*11	8.6*6.1*7.1	8	mixed
	1.9	7.2*6.5*8	5.8*5.3*6.5	3*3*5.5	8	mixed
a	2.7	12*10*13	11*9.3*12	10*9.7*12	8	mixed
	3.9	8*10*11	7.5*10*10	7.5*10*9.5	4	mesenchymal

Table 2. Difference of CEUS Parameters and Tumor volume During Preoperative Chemotherapy

1st CEUS-2nd CEUS				2nd CEUS-3rd CEUS				1st CEUS-3rd CEUS			
(dB/sec)	ΔTTP(s)	ΔPI(dB)	ΔV(cm ³)	ΔWIS(dB/sec)	ΔTTP(s)	ΔPI(dB)	ΔV(cm ³)	ΔWIS(dB/sec)	ΔTTP(s)	ΔPI(dB)	ΔV(cm ³)
0.36	-7.57	4.74	728.45	0.14	3.44	1.43	176.55	-0.22	-4.13	6.17	905.12
0.42	-3.79	1.38	116.19	0.01	0.07	0.87	15.60	-0.41	-3.72	2.25	131.90
0.38	5.73	1.75	125.71	0.10	-1.05	1.46	59.88	1.39	4.68	3.21	185.59
0.33	-6.88	1.56	67.85	1.15	-5.57	2.23	85.83	0.82	-12.45	3.79	153.69
0.45	1.50	4.01	513.92	0.35	4.16	2.47	680.47	0.80	5.66	6.48	1194.52
0.69	-2.49	2.33	85.24	-0.23	3.25	1.12	73.69	1.46	0.76	3.45	159.04
0.95	-4.93	1.42	162.97	-0.13	-2.58	1.03	31.20	1.08	-7.51	2.45	194.16
0.64	1.83	1.05	63.69	0.57	-2.96	0.86	18.33	1.21	-1.13	1.91	82.14

h in slope; TTP: time to peek; PI: peak intensity; V: tumor volume; Δ: the change of parameters;
r volume measurements: $V = L \times W^2 / 2$ [26]

Table.3 Linear Regression Analysis between Difference of Parameters and Tumors Volume

parameter	1st CEUS-2nd CEUS			2nd CEUS-3rd CEUS			1st CEUS-3rd CEUS		
	R	R ²	P	R	R ²	P	R	R ²	P
WIS	0.24	0.057	0.57	0.11	0.12	0.79	0.16	0.03	0.71
TPP	0.24	0.058	0.57	0.33	0.11	0.42	0.21	0.04	0.62
PI	0.94	0.89	0.001*	0.75	0.57	0.031*	0.53	0.28	0.18

WIS: wash in slope; TTP: time to peek; PI: peak intensity; *The changes of PI between the 1st and 2nd CEUS examinations and between the 2nd and 3rd CEUS examinations had correlation with the reduction volume of Wilms tumor.

Figures

Figure 1

This boy (NO.5 in the table 1) was diagnosed with mixed type of wilms tumor after nephrectomy. A: A solid area in tumor tissue with a good blood perfusion was chose to select the 5 RIOs when the first-time examination on children before chemotherapy treatment. B: The following CEUS examination, we tried to keep the observation area of interest basically to be the same as the first time.

Figure 2

The patient (NO.8 in the table 1), a 3.9 years old boy, was not very sensitive to chemotherapy, the tumor's volume did not change very much, and CEUS showed little significant reduction in tumor blood perfusion. A: before chemotherapy, CEUS showed that the blood perfusion inside the tumor is abundant; B: after 4 weeks preoperative chemotherapy, preoperative CEUS showed no significant decrease in blood perfusion and prominent tumor shrinkage.

Figure 3

The patient (NO.8 in the table 1), was not very sensitive to chemotherapy, the tumor's volume did not change very much, and contrast-enhanced CT showed little significant reduction in tumor's volume. A: before chemotherapy, contrast-enhanced CT showed that a large wilms tumor occupied the abdominal cavity of the child, tumor size was 8*10*11cm. B: after 4 weeks preoperative chemotherapy, the tumor volume did not change significantly, the size was 7.5*10*10 cm. The perfusion of large blood vessels inside the tumor was relatively weakened, and the tumor boundary was relatively clear.

Figure 4

The patient (NO.5 in the table 1), a 6 years old boy, was diagnosed with mixed type of wilms tumor after nephrectomy. During preoperative chemotherapy, contrast-enhanced CT showed that the tumors' volume was significantly reduced in 8 weeks. A: before preoperative chemotherapy; B: after 8 weeks preoperative chemotherapy.

Figure 5

This 6 years old boy (NO.5 in the table 1) was diagnosed with mixed type of wilms tumor after nephrectomy. During preoperative chemotherapy, The CEUS videos showed that the tumors' volume was significantly reduced and the blood perfusion in the tumor was significantly weakened in most of the children with the progress of preoperative chemotherapy. Particularly, some tumors showed liquefactive necrosis and the blood perfusion even disappeared. A: before preoperative chemotherapy; B: after 3 weeks preoperative chemotherapy. C: after 8 weeks preoperative chemotherapy.