Evaluation of the efficacy of gastric lymphoma treated with non-surgical therapy using oral and double contrast-enhanced gastric ultrasonography

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Abstract

Aim: To investigate the feasibility of assessing the efficacy of non-surgical treatment for gastric lymphoma using oral contrast-enhanced ultrasound (OCEUS) and double contrast-enhanced ultrasound (DCEUS). Material and methods: A total of 27 patients with gastric lymphoma treated nonoperatively were included in this retrospective study. The efficacy was evaluated using OCEUS and CT, respectively, and the results were tested for kappa concordance. Sixteen of the 27 patients underwent multiple DCEUS examinations before and after treatment. Micro-perfusion of the lesion in DCEUS is represented by the Echo Intensity Ratio (EIR), (echo intensity of the lymphoma lesion/echo intensity of the normal gastric wall), and one-way ANOVA was used to compare the differences between groups in EIR values before and after treatment. Results: OCEUS and CT were highly consistent in assessing the efficacy of gastric lymphoma, with a Kappa value of 0.758. During a median follow-up of 8.8 months, there was no statistical difference between the complete remission rate obtained by OCEUS and that obtained by endoscopic and CT (25.93% vs. 44.44%, p=0.154; 25.93% vs. 33.33%, p=0.766). There was also no statistical difference in the time to achieve complete remission using OCEUS assessment and endoscopy and CT (4.71±1.03 months vs. 6.01±2.14 months, p=0.088; 4.47±1.84 months vs. 6.01±2.14 months p=0.143). The difference in EIR between the groups before treatment and after different numbers of treatments was statistically significant (p<0.05), and post hoc analysis revealed this difference as early as after the second treatment (p<0.05). Conclusions: Transabdominal OCEUS and CT are comparable in the assessment of gastric lymphoma treatment efficacy. DCEUS is a noninvasive, cost-effective, and widely available method for gastric lymphoma therapeutic effect evaluation. Therefore, transabdominal OCEUS and DCEUS have the potential to be used for the early assessment of the efficacy of the non-surgical treatment of gastric lymphoma.

Keywords: Gastric lymphoma; Efficacy evaluation; Oral contrast-enhanced ultrasound; Double contrast-enhanced ultrasound

Introduction

Primary gastric lymphoma is relatively rare, accounting for about 5% of primary gastric tumors [1]. However, the stomach is the most common extranodal site for lymphoma, accounting for 30-40% of all extra-nodal lymphomas and 60-70% of gastrointestinal lymphoma [2]. Most of the primary gastric lymphomas are non-Hodgkin’s lymphomas, usually of B-lymphocyte origin, and gastric Hodgkin’s lymphoma is relatively rare. The main histological types of primary gastric lymphoma are high-grade diffuse large B-cell lymphoma (DLBCL) and low-grade mucosa-associated lymphoid tissue (MATL) lymphoma, while other types of lymphoma are rare [3]. The role of surgery in the treatment of gastric lymphoma has been diminishing over the past few decades, and the treatment of gastric lymphoma has now shifted from a predominantly surgical approach to a multimodal treat-
ment that includes *Helicobacter pylori* eradication treatment, radiation therapy, chemotherapy, and immunotherapy [4,5]. Surgical treatment is only recommended for patients with severe complications such as uncontrollable gastrointestinal bleeding and perforation [6].

Primary gastric lymphoma is a group of heterogeneous diseases. Different histological types and disease stages have different treatment methods, and the prognosis is usually good with appropriate treatment [7,8]. Therefore, the efficacy evaluation during the treatment is of great significance for optimizing the treatment plan, improving the survival rate of patients, and early detection of recurrence. Histological evaluation of repeat biopsies is the gold standard for efficacy assessment [9]. However, when the lesion is extremely mild or growing in the submucosa or intrinsic muscular layer, it is difficult to identify the lesion by endoscopy alone, which would result in a false negative biopsy result [10]. Endoscopic ultrasound (US) helps to improve the accuracy of biopsy and has advantages in providing local staging information [11]. However, the discomfort caused by invasive examination and the related infection risk result in low patient acceptance of multiple endoscopies during long-term follow-up [10]. Computed tomography (CT) and positron emission tomography-computed tomography (PET-CT) are the methods recommended by the European Society for Medical Oncology and the Chinese Society of Clinical Oncology to assess the efficacy of lymphoma and are the most commonly used methods in clinical practice [12,13]. However, high doses of ionic radiation and high cost limit their clinical application.

US is a non-invasive, radiation-free, economical, and widely accessible imaging modality. Transabdominal oral contrast-enhanced ultrasound (OCEUS) using an oral contrast agent clearly visualizes the gastric wall and gastric lesions [14]. Double contrast-enhanced ultrasound (DCEUS), which combines an oral contrast agent and intravenous microbubble contrast agent, can not only reveal the anatomical features of the gastric wall lesions but also the perfusion features [15]. At present, OCEUS and DCEUS have been widely used for the diagnosis, staging, or efficacy evaluation of gastrointestinal diseases [16,17]. However, its application in the efficacy evaluation of gastric lymphoma is still unclear. Hence, we hypothesized that OCEUS and DCEUS could be used to assess the efficacy of non-surgical treatment of gastric lymphoma. The purpose of this study was to investigate the feasibility of OCEUS and DCEUS for assessing the efficacy of non-surgical treatment of gastric lymphoma.

**Materials and methods**

This study was approved by the Ethics Committee of our Hospital. Informed consent was waived as this study was retrospective.

**Patients**

From January 2015 to October 2022, 59 consecutive patients diagnosed with gastric lymphoma at our hospital were included in this study. Inclusion criteria were as follows: 1) The patient’s diagnosis was confirmed by pathology and immunohistochemistry; 2) All patients received non-surgical treatment modalities; 3) undergo at least two or more times CT and trans-abdominal OCEUS examinations during the same period. Exclusion criteria were as follows: 1) patients with a histological diagnosis of other diseases; 2) patients who underwent surgical treatment; 3) incomplete image data. Finally, 27 patients were included. The inclusion process is shown in figure 1.

**Equipment and contrast agent**

US imaging was performed using a HI VISION Preirus US system (Hitachi Medical, Tokyo, Japan) and an Aplio i800 US system (Canon Medical, Tokyo, Japan) with a choice of convex array probes (2.5-5.0 MHz) and linear array probes (7-10 MHz). An oral contrast agent (Brand Tianxia, Huzhou East Asia Medical Co., Ltd., Zhejiang, China), mainly composed of cereal, was used for abdominal US examination to improve imaging quality, and suspicious gastrointestinal perforation and intestinal obstruction were contraindications [18]. Intravenous contrast agents were SonoVue (Bracco, Italy), composed of sulfur hexafluoride microbubbles.

**US examinations**

Before the examination, the patient was required to fast at least for 8 hours. A transabdominal gastric scan
was performed after the administration of 500 ml of pasty oral contrast. During the procedure of scan, the patient was mainly placed in the supine and right lateral positions, and the cross-section and longitudinal section of the entire stomach were scanned sequentially, including the cardia, fundus, body, and antrum of the stomach [18,19]. After the location of the lesion was determined, the probe orientation was adjusted to clearly show the maximum cross-section and the maximum longitudinal section, the images were saved, and the size, shape, echo characteristics, and blood flow information of the lesion were recorded. After completion of the OCEUS examination, the contrast mode was activated, the mechanical index was set to 0.08, and 2.4 ml of microbubble suspension was injected through a median elbow vein, flushed with 5 ml of saline, and the entire procedure was videotaped. Scanning and diagnosis were performed by a senior physician with at least 5 years of experience in gastrointestinal US diagnosis.

**Data collection and analysis**

Text and image data from transabdominal OCEUS, abdominal CT examinations, and endoscopy were collected for the 27 included patients. US image data were saved in picture or video format, CT image data were saved in a serial image or picture format, and endoscopy image data were saved in picture format. US and CT image data were analyzed by two sonographers and radiologists with at least 5 years of experience in gastrointestinal tract imaging, respectively.

**Criteria and methods for efficacy evaluation using CT and OCEUS**

In the absence of criteria dedicated to the assessment of efficacy in gastric lymphoma, the Response Evaluation Criteria in Solid Tumors (RECIST) was used to assess treatment response in CT [20]. There are currently no relevant criteria for OCEUS to evaluate the therapeutic efficacy of gastric lymphoma. According to our experience and RECIST standard, we set the following as the reference for OCEUS efficacy evaluation: 1) complete response means complete disappearance of the lesion and normalization of the layers of the gastric wall; 2) partial response refers to a reduction in the number of lesions or a reduction in lesion size by more than 30%; 3) stable disease refers to essentially no change in the number of lesions or lesion size; and 4) progression disease refers to an increase in the number of lesions or an increase in lesion size by at least 20%, or the appearance of new lesions. Considering the great morphological changes in the stomach and the diverse morphology of lymphoma lesions, we used the lesion area to indicate the lesion size, which was outlined and measured with SONOMATH software (AMBITION T. C., Chongqing, China).

**Efficacy evaluation using DCEUS**

Echo intensity of the lesion in CEUS can indicate the micro-perfusion of the lesion [21]. Sixteen subjects underwent multiple DCEUS examinations before and after treatment. We reviewed the video data of these DCEUS examinations and selected the frame in which enhancement of the lymphoma lesion was highest. We measured the echo intensity of the normal gastric wall mucosal layer and lesions using the SONOMATH software and then calculated the echo intensity ratio (EIR= echo intensity of lymphoma lesions/echo intensity of mucosal layer of the normal gastric wall). These EIR values were grouped according to the number of therapy sessions. Finally, we had six groups: before treatment, after one treatment, two treatments, three treatments, four treatments, and after eight treatments. Then the differences in EIR values among the groups were analyzed.

**Statistical analysis**

All statistical analyses were performed with SPSS software (Windows version 24.0). A chi-square test or Fisher’s exact test was used to analyze the differences in each parameter between DCBCL and MALT lymphoma, as well as the differences in complete remission rates between endoscopy, CT, and transabdominal OCEUS. Differences in time to achieve complete remission evaluated using endoscopy, CT, and OCEUS were examined by independent samples t-tests. Differences in EIR values between subgroups were analyzed using one-way ANOVA with a post hoc test using the Bonferroni method. The kappa test was used to analyze the concordance between the results of the transabdominal OCEUS and CT assessments, with a kappa value greater than 0.75 indicating high consistency. \( p < 0.05 \) were considered statistically significant.

**Results**

**Patient characteristics**

The demographic and disease data of the 27 patients included in the study are detailed in Table I. The most common first symptom was abdominal pain, followed by abdominal distention and epigastric discomfort.

**Ultrasound features of gastric lymphoma**

Gastric lymphoma lesions appear on transabdominal OCEUS as hypoechoic lesions of different morphology, which can be classified into four types: superficial, mass forming, ulcerative, and diffuse infiltrative. The OCEUS image types of gastric lymphoma and their corresponding endoscopic images are shown in figure 2. In our data, abundant blood flow signals were detected within the lymphoma lesions in only three cases using color Doppler flow imaging. The most common ultrasound image types
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Table I. Patients information

<table>
<thead>
<tr>
<th>N=27</th>
<th>Gender</th>
<th>Age</th>
<th>Histology</th>
<th>Serum LDH</th>
<th>Ki67</th>
<th>Stage</th>
<th>Helicobacter pylori</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Male</td>
<td>≤60 years</td>
<td>DLBCL</td>
<td>Normal</td>
<td>≤65%</td>
<td>I-II</td>
<td>Positive</td>
<td>Anti-HP treatment</td>
</tr>
<tr>
<td>19</td>
<td>13</td>
<td>17</td>
<td>13</td>
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<td>14</td>
<td>11</td>
<td>20</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td>&gt;60 years</td>
<td>&gt;60 years</td>
<td>MALT</td>
<td>Elevated</td>
<td>&gt;65%</td>
<td>III-IV</td>
<td>Negative</td>
<td>Chemotherapy</td>
</tr>
<tr>
<td>14</td>
<td>14</td>
<td>9</td>
<td>1</td>
<td>12</td>
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<td>16</td>
<td>7</td>
<td>9</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>MCL</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Both</td>
</tr>
<tr>
<td>1</td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>14</td>
</tr>
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</table>

Table II. Ultrasound features of diffuse large B cell lymphoma (DLBCL) and mucosa associated lymphoid tissue (MALT)

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>DLBCL (N=17)</th>
<th>MATL (N=9)</th>
<th>p</th>
</tr>
</thead>
<tbody>
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<td>Ultrasound image pattern</td>
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<tr>
<td>Superficial</td>
<td>0</td>
<td>6</td>
<td></td>
</tr>
<tr>
<td>Mass forming</td>
<td>3</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Diffuse infiltrating</td>
<td>12</td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>Ulcer</td>
<td>2</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Infiltration depth</td>
<td>0</td>
<td>0.001</td>
<td></td>
</tr>
<tr>
<td>Mucosa and submucosa</td>
<td>0</td>
<td>6</td>
<td></td>
</tr>
<tr>
<td>Muscularis propria</td>
<td>6</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>Serosa layer</td>
<td>7</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Beyond the serosa layer</td>
<td>4</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

N, number of patients

of DLBCL and MATL lymphoma were diffuse infiltrative type (70.59%) and superficial type (66.67%), respectively. The ultrasound image types of DLBCL and MALT lymphoma were statistically different (p=0.001). In addition, MALT lymphomas tend to be limited to the mucous layer, submucosal layer, or muscularis propria while DLBCL is more aggressive, often reaching the serous layer of the stomach wall or even breaking through the serous layer. There is a statistically significant difference in the depth of invasion between DLBCL and MALT lymphoma (p=0.001). Infiltration of the entire gastric wall occurred in four patients with DLBCL resulting in gastric wall stiffness and luminal obstruction. The ultrasound features of DLBCL and MALT are shown in Table II.

Fig 2. The OCEUS image types of gastric lymphoma and their corresponding endoscopic images. The first row shows the ultrasound aspect and the second row shows the corresponding endoscopic images: a) superficial type: the hierarchical structure of the gastric wall at the lesion is intact, showing only the thickening of the mucosa and submucosa; b) mass forming type: the lesion appears as a well-defined hypoechoic mass, which may be accompanied by local ulceration; c) diffuse infiltrating type: the stratified structure of the gastric wall disappears and the lesion infiltrates the entire gastric wall; d) ulcer type: the mucosal layer of the lesion loses continuity and the ulcer reached the bottom, with the entry of a hyperechoic oral contrast visible inside.
Consistency analysis of the evaluation of the efficacy of gastric lymphoma

A total of 37 efficacy evaluations were performed in 27 subjects, and 37 sets of paired evaluation data were obtained, of which two subjects underwent three assessments, six subjects underwent two assessments, and the remaining 19 subjects underwent only one time assessment, the results of which are shown in Table III. Kappa concordance test showed that OCEUS and CT were highly concordant in evaluating the effect of gastric lymphoma with a Kappa value of 0.758.

During a median follow-up period of 8.8 months (2.1 to 47.3 months), 12 (44.44%) patients achieved complete remission by endoscopy, 9 (33.33%) by CT, and 7 (25.93%) by OCEUS. There was no significant difference in the complete response rate obtained by OCEUS compared with those obtained by endoscopy and CT (25.93% vs. 44.44%, p=0.154; 25.93% vs. 33.33%, p=0.766). Time to complete remission obtained with endoscopy and abdominal CT was 4.71±1.03 months and 4.47±1.84 months, respectively, compared to 6.01±2.14 months with transabdominal OCEUS. Although complete remission was obtained later with OCEUS than with endoscopy and abdominal CT, the difference was not statistically significant (p=0.088 and p=0.143, respectively).

Evaluation of therapeutic response to gastric lymphoma using DCEUS

Altogether 16 subjects underwent 54 DCEUS examinations during the whole procedure of therapy and a total of 54 EIR mean values were obtained. Before treatment, the mean EIR values of MALT lymphoma and DLBCL lymphoma were 1.48±0.01, and 1.52±0.06, respectively. One-way ANOVA showed that the EIR values of lesions in DCEUS were statistically different among groups (p<0.05). Before treatment, the lesion was much more enhanced than the normal gastric wall, with a mean EIR value of 1.48±0.07. As the number of treatments increased, the EIR values gradually decreased. After the third treatment, the enhancement of the lesion was similar to that of the normal gastric wall, with a mean EIR value of 1.08±0.04. The subsequent increase in the number of treatments did not result in a significant change in EIR values. The EIR curve is shown in figure 3.

Post-hoc tests showed there was no significant difference in EIR values of lesions before and after the first treatment (p=0.511). However, after 2, 3, 4, and 8 courses of treatment, EIR values of the lesions were significantly different from those before treatment (p<0.05). In addition, there was no significant difference in EIR value among lesions after 3, 4, and 8 treatment courses (p=1). The change of the EIR value revealed differences in the micro-perfusion of the lesion before and after treatment. As the number of treatments increased, the micro-perfusion of the lesion decreased and the enhancement pattern changed from hyperenhancement to isoenhancement, which is similar to the enhancement intensity of the normal gastric wall (fig 4). This marked change in micro-perfusion was first seen after the second treatment, indicating that DCEUS can be used for early evaluation of therapeutic effects through micro-perfusion changes of early lesions.

Discussion

In this retrospective analysis, the evaluation results of transabdominal OCEUS and abdominal CT on the treatment response of gastric lymphoma are in good agreement (kappa value = 0.758). However, the evaluation results of transabdominal OCEUS were inconsistent with CT in 5 cases, all of which were assessed as complete response by abdominal CT, while partial remission by OCEUS. Of these, two MALT lymphoma patients had negative pre-treatment baseline CT findings and were therefore assessed as having a complete response, which
is consistent with previous studies that gastric MALT lymphomas may present as false negatives on abdominal CT scans [22]. As for the remaining three patients, we reviewed the transabdominal OCEUS and abdominal CT images and found that inadequate preparation before the examination may have contributed to the inconsistent assessment results. In these three cases, transabdominal OCEUS images showed hypoechoic lesions growing along the gastric wall with insignificant thickening (thickness 5-8 mm) and loss of gastric wall layers. Therefore, these three cases were evaluated as partial remission by transabdominal OCEUS. However, the mildly thickened gastric wall was incorrectly perceived as a fold of the gastric wall due to inadequate filling of the gastric lumen prior to CT examination. Therefore, these three patients were evaluated by CT as being in complete remission.

Gastric lymphoma, especially MALT lymphoma, tends to grow diffusely in the horizontal direction of the gastric wall. When the thickening of the gastric wall is not significant, it can be easily confused with the folds of the gastric wall, so it is important to fully fill the gastric cavity before the examination. Oral contrast agent not only dilates the gastric cavity to expel air but also creates a uniform hyperechoic background in the gastric cavity, making the gastric wall and hypoechoic gastric lymphoma lesions easier to visualize [23,24]. In addition, since oral contrast agents are mainly composed of cereals, their residence time in the stomach cavity is longer than water, thus providing sufficient examination time.

The time to achieve complete remission obtained by OCEUS assessment was longer comparing to endoscopic assessment. This is consistent with the results of previous studies on the efficacy of US endoscopy in assessing gastric lymphoma. This may be related to the inability of OCEUS to distinguish between residual lesions and post-treatment tissue fibrosis, thereby overestimating residual lesions [25,26]. Complete remission was obtained also later with OCEUS when comparing with abdominal CT. This may be related to the fact that OCEUS provides information about the hierarchy of the gastric wall. The criteria for assessing a complete remission by OCEUS include, in addition to the complete disappearance of the lesion, the normal restoration of the graded structures of the gastric wall, whereas CT cannot distinguish the hierarchy of the gastric wall [27]. The time to achieve complete remission assessed by transabdominal OCEUS was longer than that assessed by CT, suggesting that normalization of the gastric wall layers is usually later than normalization of the gastric wall thickness.

Previous studies have confirmed that assessment criteria based on lesion size changes underestimate the effect of treatment in the early stages. The morphology of the lesion does not change immediately in the early stages of treatment and changes in lesion size take longer to be observed [28,29]. Therefore, 18F-FDG PET/CT, as an imaging method that can reflect the early treatment response through tumor metabolic changes, has been widely applied in clinical practice [30,31]. However, the use of PET/CT in some types of lymphoma is controversial. Some studies have shown that 18F-FDG PET/CT is not a useful imaging modality for MALT lymphoma because 18F-FDG uptake by MALT lymphoma is low, so 18F-FDG PET/CT is less sensitive to gastric MALT lymphoma and often produces false-negative results [32]. This also occurred in our study, where four subjects with negative 18F-FDG PET/CT findings but subsequent biopsy confirmed mucosa-associated lymphoma were finally selected abdominal CT and OCEUS to evaluate the efficacy. In addition, the physiological uptake of 18F-FDG in the gastrointestinal tract and inflammatory radioactive accumulation in the stomach may also affect the diagnosis results [33].

Tumor micro-perfusion responds to treatment effects earlier than morphologic changes [34]. Similar to PET-
CT, DCEUS, as an imaging method for obtaining information on tumor micro-perfusion, can provide perfusion information on lesions for efficacy evaluation. Furthermore, DCEUS is non-radioactive and non-nephrotoxic compared to PET-CT, as the contrast agent of DCEUS is metabolized by the lungs rather than the kidneys [35]. More importantly, DCEUS is less than one-tenth the price of PET-CT, which is more acceptable to most patients. The value of DCEUS in the evaluation of efficacy has been confirmed in gastric tumors [36]. In this study, the difference of EIR before treatment and after different times of treatment was significant, and this difference was shown as early as after the second treatment. That reveals the value of DCEUS in the efficacy evaluation of gastric lymphoma, especially in the early efficacy evaluation. Furthermore, when evaluating efficacy using transabdominal OCEUS alone, it is difficult to distinguish between residual lesions and post-treatment gastric wall fibrosis. DCEUS can help determine the nature of persistent hypoechoic lesions after normalization of gastric wall thickness, whether they are residual lesions or post-treatment tissue fibrosis.

Although PET-CT is currently the most recommended and commonly used imaging modality in clinical practice, its high cost imposes a significant financial burden on patients. And the complete response standard of PET-CT depends only on the maximum standard uptake value (SUVmax) of the tumor without considering the residual lesion, which would impose a great psychological burden on the patient. As an imaging method that can reflect the effect of early treatment, due to its low price, DCEUS can be used multiple times. This would obviously reduce the psychological and financial burden of the patient. Moreover, the application of DCEUS is not limited by the type of histology. Therefore, DCEUS has great potential for clinical application in evaluating the efficacy of gastric lymphoma.

However, there are some limitations in this study. First, the incidence of gastric lymphoma is low, and the overall sample size remains small after inclusion and exclusion of noncompliant cases despite retrospective collection of cases over a longer period. Second, for a heterogeneous disease such as gastric lymphoma, it is meaningful to perform statistical analysis in subgroups according to different pathological types, disease stages and treatments. The inability to perform grouping studies due to the small sample size is a drawback. In addition, only localized lesions in the stomach were evaluated in this study, and infiltration elsewhere was not addressed here. Finally, it is hoped that a prospective pilot study with a larger sample size would be conducted in the future.

In conclusion, transabdominal OCEUS and CT are comparable in the assessment of gastric lymphoma efficacy. DCEUS is a noninvasive, cost-effective, and widely available method for gastric lymphoma therapeutic effect evaluation. Transabdominal gastric US with the aid of oral contrast and microbubble contrast has the potential to be used for earlier assessment of the efficacy of nonsurgical treatment of gastric lymphoma.

Conflict of interest: none

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References