Contrast-enhanced ultrasound for evaluating the pathologic response of breast cancer to neoadjuvant chemotherapy: A meta-analysis

Kun Jia, MD, Li Li, MD, Xiao Jing Wu, MD, Mei Jin Hao, MBBS, Hong Yuan Xue, MD

Abstract
Objective: Recent reports have suggested that contrast-enhanced ultrasound (CEUS) can be used to monitor the pathologic responses of breast cancer (BC) to neoadjuvant chemotherapy (NAC); however, the diagnostic performance of CEUS in BC has yet to be confirmed. Thus, we conducted a meta-analysis of related studies to explore the relationship between CEUS and pathologic responses of BC to NAC.

Materials and methods: We searched PubMed, Embase, Web of Science, ScienceDirect, and China National Knowledge Infrastructure databases for studies published until September 31, 2018. Study-specific odds ratios (ORs) and 95% confidence intervals (CIs) were calculated, and then ORs with 95% CIs were pooled to estimate the prognostic role of CEUS for the pathologic responses of BC to NAC.

Results: Pooled meta-analysis of the 9 eligible studies that included 424 patients indicated the high performance of CEUS for monitoring pathologic responses to NAC (OR = 31.83, 95% CI: 16.69–60.67, \( P < .001 \)), with no significant heterogeneity (\( I^2 = 0.0\% \), \( P = .529 \)). The pooled sensitivity, specificity, positive likelihood ratio, negative likelihood ratio, and diagnostic odds ratio were 87% (95% CI: 0.81–0.92), 84% (95% CI: 0.74–0.91), 5.5 (95% CI: 3.3–9.2), 0.15 (95% CI: 0.10–0.23), and 36 (95% CI: 18–70), respectively. An area under the curve of 0.92 (95% CI: 0.89–0.94) suggests a high ability for prognostic detection. Although Beggs funnel plot (\( P = .057 \)) indicated the presence of publication bias among the included studies, the trim-and-fill method verified the stability of the pooled outcomes. Sensitivity analysis suggested that the pooled OR was robust.

Conclusion: Our results suggest that CEUS has a high diagnostic performance for the pathologic responses of BC to NAC. Further and better-designed studies should be performed to verify the clinical applications of CEUS for monitoring BC responses to NAC.

Abbreviations: CEUS = contrast-enhanced ultrasound, CI = confidence interval, NAC = neoadjuvant chemotherapy, OR = odds ratio, pCR = pathological complete response.

Keywords: breast cancer, contrast-enhanced ultrasound, neoadjuvant chemotherapy, pathologic response, prognosis

1. Introduction
Breast cancer (BC) is a major health problem and is the most common cancer in women worldwide, affecting 12% of all women and leading to 450,000 deaths each year.[1] Neoadjuvant chemotherapy (NAC) is the standard treatment for locally advanced BC and inflammatory BC. NAC not only reduces tumor size to make surgery feasible but may also allow breast conserving surgery in women requiring a mastectomy. Achieving a pathological complete response (pCR) is a predictor of an improved disease free and overall survival, and it is used as a surrogate clinical endpoint for long-term outcome.[2–4] Notably, BC is highly heterogeneous with distinct molecular subtypes, and the same NAC chemotherapy regimen yields diverse responses. Therefore, clinically applicable biomarkers should be developed to predict the response of BC to NAC.

Contrast-enhanced ultrasound (CEUS) has gained vast interest in the last decade because of its capability to gather macro- and microvascular information in various organs. Thus, this technique can be used to understand the complexity of angiogenesis in different types of tumors.[5,6] CEUS is a quantitative kinetic imaging modality that assesses intravascular blood flow in breast tumors even at the capillary level. Some previous studies[7–11] suggested that CEUS can be used to monitor the pathologic responses of BC to NAC. However, the small sample size of each study might lack statistical power to draw definitive conclusions. Thus, we conducted a meta-analysis of related studies to explore the relationship between CEUS and pathologic responses of BC to NAC.

2. Materials and methods

This meta-analysis was conducted in compliance with the Preferred Reporting Items for Systematic Reviews and Meta-analyses checklist.[16] The present meta-analysis was based on

---

Editor: Sayed S. Daoud.
The authors have no conflicts of interest to disclose.

Department of Ultrasound, Hebei General Hospital, Heping West Road, Xinhua Qu, Shijiazhuang, China.

Correspondence: Kun Jia, Department of Ultrasound, Hebei General Hospital, No.348 Heping West Road, Xinhua Qu, Shijiazhuang 050051, China (e-mail: jkun0311@hotmail.com).

Copyright © 2019 the Author(s). Published by Wolters Kluwer Health, Inc. This is an open access article distributed under the terms of the Creative Commons Attribution-Non Commercial-No Derivatives License 4.0 (CCBY-NC-ND), where it is permissible to download and share the work provided it is properly cited. The work cannot be changed in any way or used commercially without permission from the journal.

Medicine (2019) 98(4):e14258
Received: 10 November 2018 / Received in final form: 27 December 2018 / Accepted: 2 January 2019
http://dx.doi.org/10.1097/MD.0000000000014258
previously published studies, and no ethical approval or patient consent was required. This study has been registered in PROSPERO (CRD42018111899).

2.1. Literature search
The PubMed, Embase, Web of Science, ScienceDirect, and China National Knowledge Infrastructure databases were searched for studies published until September 31, 2018. The following terms were used as keywords in the literature search by 2 individual authors (KJ and LL): “breast cancer,” “neoadjuvant chemotherapy,” and “contrast-enhanced ultrasound.” We also performed a full manual search of the bibliographies of selected studies to identify additional studies.

2.2. Inclusion and exclusion criteria
The inclusion criteria were as follows: the study subjects were pathologically diagnosed with BC; studies that evaluated the association between CEUS and pathologic responses of BC to NAC; sufficient data available for calculating standardized odds ratios (ORs) with 95% confidence intervals (CIs). When several studies were available for the same cohort, we retained the one with the largest number of cases for analysis.

The exclusion criteria were as follows: lack of sufficient survival data, inability to obtain the full text, reviews, letters, case reports, conference abstracts, and duplicate articles.

2.3. Data extraction
Two researchers (KJ and XJW) independently extracted detailed information using a predesigned data extraction form and assessed the quality of the individual studies. Disagreements were resolved by discussion or consensus with a third reviewer (HYX). After strict selection and evaluation, basic information, including first author, publication year, country, age, number of patients, pathologic response characteristics, tumor stage, and study period, was extracted from the included studies. The ORs and 95% CIs obtained directly from the published articles were integrated in the meta-analysis according to the study conducted.

Figure 1. Flow diagram of the inclusion and exclusion of studies.
2.4. Quality assessment

The quality of the included studies was assessed by 2 authors (KJ and MJH) using the Newcastle-Ottawa quality assessment scale (NOS)[17] and studies awarded with 6 or higher were classified as high-quality studies.[18,19] Any disagreement was resolved by discussion and consensus.

2.5. Statistical analysis

STATA 14.0 software (StataCorp, College Station, TX) was used to analyze the extracted data. The predictive value of CEUS in this meta-analysis was performed using the pooled OR and its 95% CI. Heterogeneity between studies was evaluated using the Cochran Q test and the I² test. Studies were considered to have high, moderate, or low heterogeneity when $I^2$ was >75%, 50% to 75%, or 25% to 50%, respectively. [20] Fixed-effect models were adopted only for a high-quality studies.[18,19] Any disagreement was resolved by adjustment for the funnel plot.

2.6. Subgroup analyses

We performed 4 subgroup analyses (Table 2): with published language; with pCR or response; sample size; and country. No significant deviations from the main results were found for any of the subgroups.

3. Results

3.1. Literature search

Figure 1 shows the inclusion process, in which 346 potentially relevant studies were screened. After scanning titles and abstracts, 177 studies were excluded for duplication, leaving 14 to be read in full. Eventually, the 9 remaining articles involving 424 patients were included in this meta-analysis.

3.2. Characteristics and quality assessment of the included studies

Table 1 shows the characteristics and quality assessment of the 9 included studies. The sample sizes of the 9 papers ranged from 18 to 63 with a total of 424 participants. Of these studies, 7 originated from China, one from Japan, and one from the United States. The scores of the eligible studies from the NOS ranged from 6 to 7, with a mean of 6.8, indicating that the included studies were of high quality.

3.3. Relationship between CEUS and the NAC response

The pooled results indicated the high performance of CEUS for monitoring pathologic responses to NAC (OR = 31.83, 95% CI: 16.69–60.67, $P < .001$; Fig. 2), with no significant heterogeneity ($I^2 = 0.0\%$, $P = .529$). The pooled sensitivity, specificity, positive likelihood ratio, negative likelihood ratio, and diagnostic OR were 87% (95% CI: 0.81–0.92), 84% (95% CI: 0.74–0.91), 5.5 (95% CI: 3.3–9.2), 0.15 (95% CI: 0.10–0.23), and 36 (95% CI: 18–70), respectively (Fig. 3). The sROC AUC of CEUS for the pathologic responses of BC to NAC was 0.92 (95% CI: 0.89–0.94; Fig. 4).

3.4. Subgroup analyses

We performed 4 subgroup analyses (Table 2): with published language; with pCR or response; sample size; and country. No significant deviations from the main results were found for any of the subgroups.

3.5. Sensitivity analysis and publication bias assessment

Sensitivity analyses implied that the pooled results of our meta-analysis are robust (Fig. 5). Publication bias was observed among studies using Begg’s ($P = .175$; Fig. 6A) and Egger’s ($P = .057$; Fig. 6B) tests. Results of the trim-and-fill method showed that 3 necessary studies were missed. After filling these 3 in the comprehensive analysis, the adjusted fixed-effects pooled OR of 21.38 (95% CI: 11.760–38.883, $P < .001$; Fig. 6C) calculated using the trim-and-fill method was consistent with that in the original analysis (OR = 31.83, 95% CI: 16.69–60.67, $P < .001$).

4. Discussion

BC is a vascular-dependent lesion, and its growth, infiltration, and metastasis are closely related to neo-vascularization.[122] Folkman[22] proposed that the inhibition of angiogenesis arrests solid tumors. Angiogenesis occurs at the capillary level; thus, CEUS may be one of the most direct imaging tools for visualizing perfusion changes in the tumor. CEUS can objectively depict tumor vascularity and intratumoral perfusion by reconstructing stereoscopic images.[123]

Table 1

| First author | Publication year | Country | Age, years | Number of patients | Pathological response | Clinical stage | Study period | NOS score |
|--------------|------------------|---------|------------|-------------------|----------------------|---------------|--------------|-----------|
| Amioka et al | 2016             | Japan   | 53.0±10.2  | 63                | pCR                  | I–III B       | 2012–2015   | 7         |
| Cui et al    | 2014             | China   | 45–62      | 48                | pCR                  | NA           | 2011–2013   | 7         |
| Guo et al    | 2015             | China   | 58.2±3.4   | 54                | Response             | II B–II      | 2013–2015   | 7         |
| Han et al    | 2018             | China   | 44.63±11.25| 55                | Response             | II A–III C   | 2015–2017   | 7         |
| Jia et al    | 2016             | China   | 28–63      | 48                | pCR                  | II–III       | 2010–2012   | 7         |
| Lee et al    | 2017             | USA     | 24–64      | 18                | pCR                  | NA           | 2014–2015   | 6         |
| Li et al     | 2015             | China   | 45.36±3.40 | 60                | pCR                  | II B–III     | 2011–2014   | 7         |
| Wan et al    | 2018             | China   | 50.9±9.6   | 51                | pCR                  | NA           | 2015–2016   | 6         |
| Zhang et al  | 2014             | China   | 44.04±7.61 | 27                | response             | II B–III     | 2011–2013   | 7         |

NOS = Newcastle-Ottawa scale; pCR = pathological complete response.

Continuous variable is presented as means ± SD or range.
In the present meta-analysis, we searched several major databases for studies exploring the relationship between CEUS and pathological response of BC to NAC. By combining the data from the 9 studies, CEUS presented a diagnostic sensitivity of 87%, a specificity of 84%, and an AUC of 0.92. These 3 representative parameters confirmed the accuracy of CEUS as a valuable imaging method for assessing the response of BC to NAC. In addition, the diagnostic OR estimated for CEUS was 36 (95% CI: 18–70). This benign high-DOR value indicated that CEUS could monitor response in NAC accurately.

Various conventional imaging modalities are used in the preoperative setting, including mammography, ultrasound, and magnetic resonance imaging (MRI). A common potential limitation of mammography, ultrasound, and MRI imaging is their inability to distinguish viable tumor tissue from fibrotic scar tissue; thus, they are incapable of accurately predicting response to NAC.

Figure 2. Forest plot of the association of CEUS with the neoadjuvant chemotherapy response. CEUS = contrast-enhanced ultrasound, NAC = neoadjuvant chemotherapy.

Figure 3. Forest plots of sensitivity and specificity for CEUS predicting NAC response. CEUS = contrast-enhanced ultrasound, NAC = neoadjuvant chemotherapy.
response. Response as assessed by a reduction in tumor size often manifests later than changes in underlying tumor characteristics, such as vascularization and vascular permeability, cellularity, metabolism, and biochemistry. Thus, imaging modalities, such as CEUS, dynamic-contrast enhanced MRI (DCE-MRI), and fluorodeoxyglucose positron emission tomography, and computed tomography (FDG-PET/CT), which can quantify tumor functions, are becoming increasingly important in the evaluation and prediction of therapy response. Among different approaches, DCE-MR is especially promising due to its ability to quantitatively measure kinetic parameters related to perfusion and permeability of tumor. Several clinical studies in the NAC setting have demonstrated that tumor reduction measured by DCE-MRI is in concordance with pathologic response, and the measurement can be a prognostic indicator of survival. However, a recently published

![Figure 4. Summary ROC curve for the nine included studies. Numbers in brackets are 95% CIs. CEUS = contrast-enhanced ultrasound, NAC = neoadjuvant chemotherapy, AUC = area under ROC curve, SENS = sensitivity, SPEC = specificity.](image)

| Table 2 | Subgroup analysis. |
|---------|-------------------|
| Number of studies | OR (95% CI) | $P$ | $\hat{P}$ ($P$ value) |
| Language | | | |
| English | 4 | 58.40 (17.72–192.44) | <.001 | 0.0% (.396) |
| Chinese | 5 | 22.86 (10.45–50.00) | <.001 | 0.0% (.647) |
| Pathological response | | | |
| pCR | 6 | 32.19 (14.26–72.71) | <.001 | 4.4% (.388) |
| Response | 3 | 31.04 (11.06–87.13) | <.001 | 0.0% (.399) |
| Sample size | | | |
| ≥50 | 5 | 34.99 (15.83–77.35) | <.001 | 0.0% (.579) |
| <50 | 4 | 26.37 (8.75–79.45) | <.001 | 27.6% (.246) |
| Country | | | |
| China | 7 | 29.98 (14.78–60.79) | <.001 | 23.1% (.254) |
| Non-China | 2 | 40.90 (8.45–198.02) | <.001 | 0.0% (.453) |

CI = Confidence interval, OR = odds ratios, pCR = pathological complete response.
systematic review has shown that DCE-MRI has a high specificity (50%–97%) versus only moderate sensitivity (25%–100%) in the prediction for pCR. FDG-PET/CT is correlated with increased glucose metabolism in BC. Metabolic reduction detected between baseline and the early phase of NAC can provide early information on the potential BC response. By contrast, FDG-PET/CT has a high sensitivity (86%–90%) versus only moderate specificity (40%–85%) in pCR prediction.[31–33] The contrast agents used in CEUS remain only within the intravascular bed and do not diffuse into the interstitial space; hence, the reliability of CEUS is high. CEUS is widely available and can be performed in patients who cannot undergo DCE-MRI. Moreover, DCE-US can directly visualize perfusion status in the tumor. Notably, for deep-seated tumors and tumors with low vascularity, CEUS cannot delineate microvasculature and microcirculation features and monitor BC responses to NAC.[34] However, the varying results in the separate studies showed that the usefulness of the various imaging parameters in predicting the response to NAC in BC was still not clearly defined. Nevertheless, each modality offers unique and complementary information on several clinically relevant tumor characteristics.

We believe the conclusions drawn from this study are important but should be interpreted with caution because of several limitations. First, in a meta-analysis of published studies, publication bias is an inevitable problem. Second, the analysis used pooled data (individual data were not available), which restricted us from performing a more detailed relevant analysis and obtaining more comprehensive results. Third, the sample sizes of comparative studies available in the literature are relatively small, which may contribute to an overestimation of diagnostic accuracy. Fourth, most of the included studies were conducted in China and published in unknown magazines, and this may lead to limited generalizability.
In conclusion, the findings of our study demonstrated that CEUS modality holds a relatively high sensitivity and specificity in the evaluation and prediction of the response of BC to NAC. Nonetheless, a variety of issues should be considered when assessing CEUS techniques for estimating BC responses to NAC, and large-scale and well-designed clinical trials are needed to assess the technique’s diagnostic value.

**Author contributions**

**Conceptualization:** Kun Jia, Li Li.
**Data curation:** Li Li, Xiao Jing Wu, Mei Jin Hao, Hong Yuan Xue.
**Methodology:** Mei Jin Hao.
**Software:** Hong Yuan Xue.
**Writing – original draft:** Xiao Jing Wu.
**Writing – review & editing:** Kun Jia.

**References**

[1] Torre LA, Bray F, Siegel RL, et al. Global cancer statistics, 2012. CA Cancer J Clin 2015;65:87–108.

[2] Kaufmann M, von Minckwitz G, Mamounas EP, et al. Recommendations from an international consensus conference on the current status and future of neoadjuvant systemic therapy in primary breast cancer. Ann Surg Oncol 2012;19:1508–16.

[3] Kuerer HM, Newman LA, Smith TL, et al. Clinical course of breast cancer patients with complete pathologic primary tumor and axillary lymph node response to doxorubicin-based neoadjuvant chemotherapy. J Clin Oncol 1999;17:460–9.

[4] von Minckwitz G, Untch M, Blohmmer JF, et al. Definition and impact of pathologic complete response on prognosis after neoadjuvant chemotherapy in various intrinsic breast cancer subtypes. J Clin Oncol 2012;30:1796–804.

[5] Ko EY, Lee SH, Kim HH, et al. Evaluation of tumor angiogenesis with a second-generation US contrast medium in a rat breast tumor model. Korean J Radiol 2008;9:243–9.

[6] Tranquart F, Claudon M, Correas JM. Guidelines for the use of contrast agents in ultrasound. J Radiol 2005;86:1047–54.

[7] Amioka A, Masamoto N, Gouda N, et al. Ability of contrast-enhanced ultrasonography to determine clinical responses of breast cancer to neoadjuvant chemotherapy. Jpn J Clin Oncol 2016;46:303–9.

[8] Cui RR, Xu C, Lu XL, et al. The accuracy of functional magnetic resonance imaging and contrast-enhanced ultrasound on evaluation of breast cancer patients after neoadjuvant chemotherapy. Chin J Postgrad Med 2014;37:24–7.

[9] Guo L, Hao T, Zhang YH, SLL, et al. Application value of contrast-enhanced ultrasound in evaluating the effect of neoadjuvant chemotherapy for breast cancer. J Xiangjia Med Univ 2015;38:1553–5.

[10] Han YF, Zou SW, Wang BH. Application of ultrasound elastography in various intrinsic breast cancer subtypes. J Clin Oncol 2012;30:1796–804.

[11] Jia WR, Tang L, Wang DB, et al. Three-dimensional contrast-enhanced ultrasound of the breast: Is it feasible in malignant risk assessment of breast lesions? Eur J Radiol 2018;103:118–23.

[12] Lea SC, Grant F, Sheth P, et al. Accuracy of contrast-enhanced ultrasound compared with magnetic resonance imaging in assessing the tumor response after neoadjuvant chemotherapy for breast cancer. J Ultrasound Med 2017;36:901–11.

[13] Li L, Li YM, Li HB, et al. Value of ultrasonic contrast in evaluation of neoadjuvant chemotherapy effects on patients with breast cancer. Med J Natl Defending Forces Southwest China 2015;25:737–40.

[14] Wan CF, Liu XS, Wang L, et al. Quantitative contrast-enhanced ultrasound evaluation of pathological complete response in patients with locally advanced breast cancer receiving neoadjuvant chemotherapy. Eur J Radiol 2018;103:118–23.

[15] Zhang L, Hao J, Wang LP, et al. Contrast-enhanced ultrasound, color Doppler ultrasound and MRI-PWI for evaluating the response of breast cancer to neoadjuvant chemotherapy. Acta Med Sci Technol Huazhong 2014;43:449–52.

[16] Liberati A, Altman DG, Tetzlaff J, et al. The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate healthcare interventions: explanation and elaboration. BMJ 2009;339:b2700.

[17] Stang A. Critical evaluation of the newcastle-ottawa scale for the assessment of the quality of nonrandomized studies in meta-analyses. Eur J Epidemiol 2010;25:603–5.

[18] Wang Z, Wang J, Wang P. The prognostic value of prognostic nutritional index in hepatocellular carcinoma patients: a meta-analysis of observational studies. PLoS One 2018;13:e0202987.

[19] Yang Y, Gao P, Song Y, et al. The prognostic nutritional index is a predictive indicator of prognosis and postoperative complications in gastric cancer: a meta-analysis. Eur J Surg Oncol 2016;42:1176–82.

[20] Higgins JP, Thompson SG, Deeks JJ, et al. Measuring inconsistency in meta-analyses. BMJ 2003;327:557–60.

[21] Yu L, Wang WP, Jia WR, et al. Three-dimensional contrast-enhanced sonography in the assessment of breast tumor angiogenesis: correlation with microvessel density and vascular endothelial growth factor expression in breast cancer. J Ultrasound Med 2014;33:835–46.

[22] Loo CE, Teertstra HJ, Rodenhuis S, et al. Dynamic contrast-enhanced MRI for prediction of breast cancer response to neoadjuvant chemotherapy: initial results. AJR Am J Roentgenol 2008;191:1331–8.

[23] Huang W, Li X, Chen Y, et al. Variations of dynamic contrast-enhanced magnetic resonance imaging in evaluation of breast cancer therapy response: a multicenter data analysis challenge. Transl Oncol 2014;7:153–66.

[24] Li X, Kang H, Arlinghaus LR, et al. Analyzing spatial heterogeneity in DCE- and DW-MRI parametric maps to optimize prediction of pathologic response to neoadjuvant chemotherapy in breast cancer. Transl Oncol 2014;7:14–22.

[25] Partridge SC, Gibbs JE, Lu Y, et al. MRI measurements of breast tumor volume predict response to neoadjuvant chemotherapy and recurrence-free survival. AJR Am J Roentgenol 2005;184:1774–81.

[26] Heldahl MG, Bather TF, Rydland J, et al. Prognostic value of pretreatment dynamic contrast-enhanced MR imaging in breast cancer patients receiving neoadjuvant chemotherapy: overall survival predicted from combined time course and volume analysis. Acta Radiol 2010;51:604–12.

[27] Hylton NM, Blume JD, Bernreuter WK, et al. Locally advanced breast cancer: MR imaging for prediction of response to neoadjuvant chemotherapy—results from ACRIN 6657/SPY TRIAL. Radiology 2012;263:663–72.

[28] Pickles MD, Manton DJ, Lowry M, et al. Prognostic value of pretreatment DCE-MRI parameters in predicting disease-free and overall survival for breast cancer patients undergoing neoadjuvant chemothera- pty. Eur J Radiol 2009;71:498–503.

[29] Chen L, Yang Q, Bao J, et al. Direct comparison of PET/CT and MRI to predict the pathological response to neoadjuvant chemotherapy in breast cancer: a meta-analysis. Sci Rep 2017;7:8479.

[30] Gu YL, Pan SM, Ren J, et al. Role of magnetic resonance imaging in detection of pathologic complete remission in breast cancer patients treated with neoadjuvant chemotherapy: a meta-analysis. Clin Breast Cancer 2017;17:245–35.

[31] Liu Q, Wang C, Li P, et al. The role of (18)F-FDG PET/CT and MRI in assessing pathological complete response to neoadjuvant chemotherapy in patients with breast cancer: a systematic review and meta-analysis. Biomed Res Int 2016;2016:3746232.

[32] Luo J, Chen JD, Chen Q, et al. Predictive model for contrast-enhanced ultrasound of the breast: Is it feasible in malignant risk assessment of breast imaging reporting and data system 4 lessons? World J Radiol 2016;8:600–9.