Correlation between the dynamic contrast-enhanced MRI features and prognostic factors in breast cancer

A retrospective case-control study

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Abstract
This study analyzed the correlation between the dynamic contrast-enhanced MRI (DCE-MRI) features with prognostic factors of breast cancer. Eighty-five breast cancer patients verified by pathology and immunohistochemistry underwent DCE-MRI examination. Spearman correlation analysis was used to analyze the DCE-MRI features (the strengthening types, shape, distribution, edge, internal reinforcement and the time-signal intensity curve (TIC) types) and the 4 immunohistochemical markers (ER, PR, Her-2, and Ki-67) by GraphPad InStat version 6.0 software. The enhanced morphology types, shapes, edge had significant correlation with the expression of ER (P = .001, P = .000, P = .001, respectively), PR (P = .045, P = .015, P = .000, respectively) and Ki-67 (P = .039, P = .000, P = .024, respectively), and no significant correlation with Her-2 expression (P = .906, P = .074, P = .679, respectively) was observed. There was significant correlation between internal enhancement patterns and Ki-67 expression (P = .004), and no significant correlation between internal enhancement patterns and the expression of ER, PR, and Her-2 (P = .208, P = .689, P = .437, respectively) was observed. TIC had significant correlation with ER, Ki-67 expressions (P = .022, P = .001, respectively), and no correlation with expressions of PR and Her-2 (P = .128, P = .391, respectively) was observed. The DCE-MRI features of breast cancer were well correlated with the expression of immunohistochemistry, and might also be helpful to evaluate the biological progress and prognosis.

Abbreviations: BI-RADS = the Breast Imaging Reporting and Data System, DCE-MRI = the dynamic contrast-enhanced magnetic resonance imaging, ER = estrogen receptor, Her-2 = human epidermal growth factor receptor 2, Ki-67 = antigen identified by monoclonal antibody Ki-67, PR = progesterone receptor, TIC = the time-signal intensity curve.

Keywords: breast cancer, dynamic contrast-enhanced MRI, prognostic factors

1. Introduction
MRI allows high contrast in soft tissues, multidirectional, multiparameters, and multifunctional imaging, thus it showed a sensitivity of 95% to 99% in various breast cancer examinations.\textsuperscript{1,2} Of all MRI techniques used for assessment of breast cancer, the dynamic contrast-enhanced MRI (DCE-MRI) is particularly sensitive as it demonstrates the morphological and hemodynamic features of tumors more accurately than mammography and ultrasound.\textsuperscript{1,3} The study by Lee et al\textsuperscript{[4]} showed that morphological features like lesion size, number, shape, boundary and internal structure, and hemodynamic features of DCE-MRI in the breast cancer can indirectly reflect the tumor proliferation rate and growth of the tumor state, and then help in the early diagnosis of breast cancer and prognosis evaluation.

As a sex hormone dependent organ, the cell proliferation and development of breast are controlled by estrogen and progesterone hormones.\textsuperscript{[5]} Therefore, estrogen and progesterone are important endocrine factors that not only were involved in the normal development of breast, but also in the induction of breast cancer. Estrogen receptor (ER) and progesterone receptor (PR) play an important biological function role by specifically combining with estrogen and progesterone. Higher positive expressions of ER and PR demonstrated breast cancer with a higher degree of differentiation, low malignant degree, and with better prognosis.\textsuperscript{[6,7]} Human epidermal growth factor receptor 2 (Her-2) is one of the genetic markers that is commonly observed in breast cancer, which is positively correlated with the amplification and higher protein expression and in turn in the progression of breast cancer.\textsuperscript{[8]} Her-2 is rarely expressed in normal tissues, and is expressed positively in poorly differentiated and invasive breast cancers.\textsuperscript{[7,9]} Ki-67 antigen is a nuclear proliferation marker that exists in the cell cycle except in the G0 phase. It begins to express in the G1 phase of cell cycle, expresses increasingly in S and G2 phases, reaches the peak in late M phase quickly and disappears during the late cell division.\textsuperscript{[10]} Ki-67 is closely associated with the degree of differentiation, invasion, metastasis and prognosis of several tumors, and reflects the biological behavior and prognosis of breast cancer to a certain
extent. These in turn demonstrate higher expression of Ki-67, with a higher malignant degree and with worse prognosis of breast cancer.\(^{[11]}\)

ER, PR, Her-2, and Ki-67 as immunohistochemistry indexes of breast cancer are the factors that were associated in the prognostic analysis, and acts as indicators of targeted therapy.\(^{[12-14]}\) However, there were very few studies that analyzed the correlation between DCE-MRI features and prognostic factors of breast cancer. Here, we studied the correlation of DCE-MRI features and the expression of ER, PR, Her-2, and Ki-67, and then predicted the biological behavior and prognosis of breast cancer indirectly from the imaging characteristics of DCE-MRI.

2. Materials and methods

2.1. Study population

This study was approved by the Ethics Committee of the Affiliated Huai’an No.1 People’s Hospital of Nanjing Medical University and First Affiliated Hospital, Soochow University, and informed consent was obtained from all study participants. In our retrospective study, all patients need to comply with the following 3 conditions: every patient must undergo examination of MRI contrast-enhanced imaging of breast before treatment and puncture; patients were pathologically proved for breast cancer; every lesion should undergo immunohistochemistry examination. From September 2014 to October 2017, 85 female patients (mean age, 45.7 years; 24–63 years) with breast cancer were included in our study. All patients underwent DCE-MRI examination before therapy and were pathologically proved to have breast cancer and immunohistochemistry after surgery. Two pathologists have reviewed the patients’ pathological and immunohistochemistry reports.

2.2. MRI equipment and scanning methods

MR imaging is performed with the patient lying in prone position using a 4-channel bilateral breast coil that covers both the breasts with 3.0 T MRI scanner (Verio: Siemens Healthcare, Erlangen, Germany). All the patients were examined by the following sequences: an axial, turbo spin-echo T2-weighted imaging sequence with a TR/TE of 4300/61.0, a flip angle of 80°, 34 slices, a field of view (FOV) of 360 mm, a matrix size of 272 × 320, a number of excitations (NEX) of 1, a slice thickness of 4 mm and an acquisition time of 5 min and 19 seconds; pre- and postcontrast, axial T1-weighted flash 3-dimensional sequences with a TR/TE of 4.67/1.66, a flip angle of 10°, a slice thickness of 1.2 mm, measurement of 6 and an acquisition time of 6 minutes and 10 seconds. The images were obtained before and at 10, 70, 130, 190, 250, and 310 seconds after the rapid bolus injection of GD-DTPA (Magnevist, Wayner, NJ), delivered 0.1 mmol/kg (0.5 ml) over 8 seconds. Postprocessing manipulation included the production of subtraction, multiplanar reconstruction of sagittal image, and maximum-intensity-projection (MIP) images.

It was unknown that menstrual cycle of the premeopausal patients were told to do the breast MRI examination.

2.3. Image descriptors

All MRI images were assessed retrospectively by 2 experienced radiologists in consensus using the Breast Imaging Reporting and Data System (BI-RADS) 5th lexicon.\(^{[15]}\) Based on MR imaging, the tumors were divided into 3 categories: focus, the mass enhancement, and the non-mass-like enhancement. The mass enhanced types were described by 3 aspects such as shape, edge, and internal enhancement pattern. The masses were round, oval, lobular, and irregular in the shape, and smooth, speculated, irregular at the edge. The internal enhancement pattern included homogeneous, heterogeneous, rim enhancement, dark internal septation which in turn enhances the internal septation and central enhancement. The non-mass-like enhanced type contained 2 aspects: the distribution pattern and internal enhancement pattern. The distribution pattern included focal area, linear, ductal, multiple regions, and diffuse distribution. The internal enhancement pattern included the homogeneous, heterogeneous, stipple or punctate, clump, reticular, or dendritic. The internal enhancement pattern was not evaluated with linear distribution. Time signal intensity curve of these lesions are divided into 3 types: type inflow, platform, and outflow. In cases of multifocal and multicentric breast cancers, we evaluated only cancer indices for size measurement. Bilateral breast cancers were treated as individual lesions. All the MR findings, including areas of multifocality/multicentricity, were found and recorded at histological sites.

2.4. Immunohistochemistry analysis

The expressions of ER, PR, HER-2, and Ki-67 were evaluated using immunohistochemistry S-P method. According to the international practice,\(^{[16]}\) ER and PR positivity were defined using the cut-off values of 1% and 10%,\(^{[15]}\) Therefore, positive cells of ER or PR rate < 1 % were considered as negative, marked as “0,” 1%–10% as low expression, marked as “1+,” > 10% were positive and marked as “2+.” Her-2 positive is expressed in cell membrane, and the results of Her-2 were calculated by the percentage of tumor cell membrane which showed a completely shading and tinting strength.\(^{[17]}\) The negative staining results and the cells whose cell membrane were stained less than 10% of tumor cells were marked as “0”; “1+” was marked for the tumor cells > 10% and ≤ 30% where the cell membrane staining intensity showed light and barely visible performance cells, and the cell membranes were partly stained. “2+” was given for weak-medium strength of intact cell membrane performance for >10% and ≤ 30% of the tumor cells. “3+” showed a strong complete membrane staining intensity in >30% of tumor cells. Ki-67 positive was expressed in the nucleus. According to the breast cancer working group on international recommended guidelines in 2011,\(^{[17]}\) Ki-67 can be divided into 3 groups: positive cell rate ≤14% was marked as “0”, positive cell rate between 14 % and 20% was “1+”; and positive cell rate > 20% was “2+.”

2.5. Statistical methods

GraphPad Prism 6 for Windows (Graphpad Software, San Diego, CA) was used to perform statistical analysis. Spearman correlation analysis was performed with α=0.05 for statistically significant. \(P<0.05\) was considered to be statistically significant.

3. Results

3.1. Pathological types of breast cancer

There were 85 cases of biopsy-proven unifocal breast cancer. Of these, 65 cases were with infiltrating ductal carcinoma, 17 cases were with ductal carcinoma in situ, 2 cases with mammary gland mucous carcinoma, and 1 case of medullary carcinoma.
3.2. DCE-MRI features of lesions

Of the 85 cases with breast cancer, 58 cases had mass enhancement and 27 cases (Fig. 1A) had non-mass-like enhancement (Fig. 2A). There were 17 cases with oval, 29 with lobulated, and 12 with irregular in mass enhancement according to their strengthening shape. The mass enhancement was divided into 4 cases of smooth, 40 cases of burr, and 14 cases were irregular by the edge of the tumor. These were grouped into 4 cases of uniform, 43 cases of uneven reinforcement, and 11 cases of circular in the light of the internal reinforcement characteristics. There were 8 cases of duct distribution and 19 cases of segmental distribution in the non-mass-like enhancement, which were further divided into 13 cases of reinforcement cluster and 14 cases of uneven strengthening according to the internal reinforcement characteristics. The time intensity curve (TIC) of DCE-MRI was divided into 4 cases of flow-in type (Fig. 1B) with mammary gland mucous carcinoma in 2 cases and the 2 cases of ductal carcinoma in situ, 10 cases of platform type all with infiltrating ductal carcinoma, and 71 cases of flow-out type (Fig. 2B) with ductal carcinoma in situ in 15 cases, infiltrating ductal carcinoma in 55 cases, and medullary carcinoma in 1 case.

3.3. Immunohistochemistry of lesions

MRI strengthening characteristics of the lesions and their corresponding immunohistochemical results of ER, PR, Her-2, Ki-67 (Figs. 1B–F and Figs. 2B–F) were shown in Table 1.
3.4. The CE-MRI features correlates with the expression of immunohistochemistry

Spearman correlation analysis results presented the MRI strengthening types, shape or distribution and edge of the breast cancer which showed significant differences with the expression of ER (P = .001, P = .000, P = .001, respectively), PR (P = .045, P = .015, P = .000, respectively) and Ki-67 (P = .039, P = .000, P = .024, respectively), and no significant differences were observed with Her-2 expression (P = .906, P = .074, P = .0679, respectively). There was significant difference between the internal reinforcement patterns and Ki-67 expression (P = .004), and no significant differences between signal intensity-time curve and expression of ER, PR, and Her-2 (P = .208, P = .682, P = .437, respectively). Time-signal intensity curve demonstrated significant differences with ER, Ki-67 expression (P = .022, P = .001, respectively), and no significant differences were observed with the expression of PR and Her-2 (P = .128, P = .391, respectively), (Table 2, Figs. 3–8).

4. Discussion

At present, MRI has the highest soft tissue resolution in all the imaging examinations. The DCE-MRI demonstrated the strengthening shape or distribution, edge, internal reinforcement of breast cancer and the relation between the nidus and the structures around the cell membrane clearly to aid greatly in the accurate diagnosis of breast cancer. Research results of Dogan et al[19] showed that the sensitivity of breast cancer was 100% by DCE-MRI, but the specificity was very low. Another research study by Kul et al[20] showed that the DCE-MRI specificity of breast cancer was 72%. The reason for its relatively low specificity was due to the diverse and complex manifestations of MRI in breast cancer. This was determined by the way of tumor growth and was closely related to the genes that control tumor growth.[21] It is believed that the expressions of ER, PR, Her-2, Ki-67 in breast cancer reflect the biological behavior to a certain extent.[22] Our research investigated whether there was a correlation between the expression of immunohistochemical parameters such as ER, PR, Her-2, Ki-67 with the DCE-MRI features of breast cancer from the strengthening types, shape, distribution, edge, internal reinforcement, and the TIC types according to the norm of BI-RADS.

According to the BI-RADS MRI criteria,[15] breast cancer had 2 kinds of MRI strengthening types: the mass reinforcement and the non-mass-like enhancement. These had a correlation with the expression of ER, PR, Ki-67, but no correlation with the expression of Her-2. In our research, the mass reinforcement showed higher positive expression of ER and PR, and lower positive expression of Ki-67 than non-mass-like enhancement in breast cancer (Fig. 3). The results demonstrated that the mass reinforcement had a better prognosis than the non-mass-like enhancement. But some studies indicated that the degree of

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| Table 1 | DCE-MRI features and the corresponding immunohistochemical indexes. |
|---------|-------------------------------------------------------------|
| MRI strengthening characteristics | ER | PR | Her-2 | Ki-67 |
| | 0 | 1+ | 2+ | 0 | 1+ | 2+ | 0 | 1+ | 2+ | 0 | 1+ | 2+ | Total |
| The mass enhancement | 8 | 13 | 37 | 11 | 13 | 34 | 14 | 14 | 20 | 10 | 17 | 15 | 26 | 58 |
| The non-mass-like enhancement | 10 | 10 | 7 | 8 | 10 | 9 | 11 | 2 | 6 | 8 | 3 | 6 | 18 | 27 |
| The mass enhancement | Shape | Oval | 1 | 2 | 14 | 2 | 3 | 12 | 6 | 6 | 3 | 2 | 11 | 5 | 1 | 17 |
| | | Lobulated | 5 | 7 | 17 | 6 | 7 | 16 | 8 | 6 | 11 | 4 | 6 | 6 | 17 | 29 |
| | | Irregular | 2 | 4 | 6 | 3 | 3 | 6 | 0 | 2 | 6 | 4 | 0 | 4 | 8 | 12 |
| | Edge | Smooth | 0 | 0 | 4 | 0 | 0 | 4 | 0 | 2 | 2 | 0 | 4 | 0 | 0 | 4 |
| | | Burr | 3 | 9 | 28 | 3 | 11 | 26 | 10 | 10 | 14 | 6 | 11 | 11 | 18 | 40 |
| | | Irregular | 5 | 4 | 5 | 8 | 2 | 4 | 4 | 2 | 4 | 4 | 2 | 4 | 8 | 14 |
| Internal reinforcement | Uniform | 0 | 2 | 2 | 1 | 1 | 2 | 0 | 2 | 2 | 0 | 3 | 1 | 0 | 4 |
| | | Uneven | 6 | 8 | 29 | 9 | 8 | 26 | 12 | 12 | 11 | 8 | 8 | 12 | 23 | 43 |
| | | Circular | 2 | 3 | 6 | 1 | 4 | 6 | 2 | 6 | 1 | 2 | 6 | 2 | 3 | 11 |
| The non-mass-like enhancement | Distribution | Duct | 3 | 2 | 3 | 1 | 4 | 3 | 7 | 0 | 0 | 1 | 2 | 4 | 2 | 8 |
| | | Segmental | 7 | 8 | 4 | 7 | 6 | 6 | 4 | 2 | 6 | 7 | 1 | 2 | 16 | 19 |
| | Internal reinforcement | Cluster | 4 | 5 | 4 | 4 | 2 | 7 | 7 | 2 | 2 | 2 | 2 | 3 | 8 | 13 |
| | | Uneven | 6 | 5 | 3 | 4 | 8 | 2 | 4 | 0 | 4 | 6 | 1 | 3 | 10 | 14 |
| Flow-in | 0 | 0 | 4 | 0 | 0 | 4 | 2 | 0 | 2 | 0 | 4 | 0 | 0 | 4 |
| platform | 1 | 2 | 7 | 1 | 4 | 5 | 0 | 2 | 6 | 2 | 5 | 2 | 3 | 10 |
| flow-out | 17 | 21 | 33 | 18 | 19 | 34 | 23 | 14 | 18 | 16 | 11 | 19 | 41 | 71 |

ER = estrogen receptor, Her-2 = human epidermal growth factor receptor 2, Ki-67 = antigen identified by monoclonal antibody Ki-67, PR = progesterone receptor.

| Table 2 | Correlation between the DCE-MRI features and the expression of immunohistochemistry. |
|---------|-------------------------------------------------------------|
| | Strengthening types | Strengthening shape/distribution | Strengthening edge | Internal reinforcement | TIC |
| | r | P | r | P | r | P | r | P | r | P |
| ER | −0.346 | .001 | −0.361 | .000 | −0.405 | .001 | −0.125 | .208 | −0.233 | .022 |
| PR | −0.208 | .045 | −0.288 | .015 | −0.443 | .000 | −0.040 | .682 | −0.155 | .128 |
| Her-2 | −0.012 | .906 | 0.162 | .074 | 0.040 | .679 | −0.074 | .437 | −0.084 | .391 |
| Ki-67 | 0.214 | .039 | 0.405 | .000 | 0.277 | .024 | 0.282 | .004 | 0.350 | .001 |

DCE-MRI = dynamic contrast-enhanced magnetic resonance imaging, ER = estrogen receptor, Ki-67 = antigen identified by monoclonal antibody Ki-67, PR = progesterone receptor, TIC = time-signal intensity curve.
malignancy of mass reinforcement was higher than the non-mass-like enhancement which may be associated with the sample types and numbers.\(^{23,24}\)

In our study, there was oval, lobulated shape, and irregular shapes in the mass reinforcement, while there was conduit reinforcement and lobular segment reinforcement in the non-mass-like enhancement. The shape and distribution in the 2 types had a correlation with the expression of ER, PR, Ki-67, but no correlation observed with the expression of Her-2. The results in Figure 4 demonstrated oval lesions which had higher positive expression of ER, PR, and lower positive expression of Ki-67 than lobulated and irregular lesions. Peng’s et al\(^{25}\) study has shown that lobulated tumor was closely related to the expression of Ki-67, which showed stronger invasion, and higher degree of malignancy. In Shu’s et al\(^{15}\) study, the lobulated tumor was closely related to the expression of Ki-67. These standpoints were partly consistent with our study. Figure 5 demonstrated duct lesions that had higher positive expression of ER, PR and lower positive expression of Ki-67 than lobular segmental lesions in the non-mass-like enhancement. Duct reinforcement lesions had higher degree of differentiation, lower degree of malignancy than lobular segment lesions. The size of duct reinforcement lesions was smaller than the lobular segmental lesions. Yu’s et al\(^{23}\) study demonstrated that bigger is the size of the lesion, higher is degree of malignancy and our results were consistent with it.

Figure 3. The corresponding proportions of ER, PR, and Ki-67 in different strengthening types. ER = estrogen receptor, Ki-67 = antigen identified by monoclonal antibody Ki-67, PR = progesterone receptor.

Figure 4. The corresponding proportions of ER, PR, and Ki-67 in different strengthening shapes. ER = estrogen receptor, Ki-67 = antigen identified by monoclonal antibody Ki-67, PR = progesterone receptor.
This research showed that the mass reinforcement edge of breast cancer had a correlation with the expression of ER, PR, Ki-67, but no correlation with the expression of Her-2. Figure 6 indicated lesions of smooth edge which showed higher positive expression of ER, PR and lower Ki-67 positive expression and had higher degree of differentiation, lower degree of malignancy than lesions of the burr edge. Lesions of burr edge had higher positive expression of ER, PR and lower Ki-67 positive expression and had higher degree of differentiation, lower degree of malignancy than lesions of the irregular edge. In Wang’s et al.[21] study, the burr edge was correlated with ER, PR, but not with Her-2. In this regard, this point was in accordance with our study.

Our study manifested the internal strengthening characteristics of breast cancer which were associated with the expression of Ki-67, but not with ER, PR, and Her-2. Figure 7 showed that uniform lesions had lower proportions of Ki-67 positive expression than circular lesions and uneven lesions, and circular lesions had lower proportions of Ki-67 positive expression than uneven lesions in the internal strengthening of the mass reinforcement. In the non-mass-like enhancement, cluster lesions had lower proportions of Ki-67 positive expression than uneven lesions. These in turn indicated the uniform internal strengthening lesions which had the highest degree of differentiation, the lowest degree of malignancy, and the circular strengthening lesions which had lowest degree of differentiation, highest degree

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**Figure 5.** The corresponding proportions of ER, PR, and Ki-67 in different strengthening distributions. ER=estrogen receptor, Ki-67=antigen identified by monoclonal antibody Ki-67, PR=progesterone receptor.

**Figure 6.** The corresponding proportions of ER, PR, and Ki-67 in different strengthening edges. ER=estrogen receptor, Ki-67=antigen identified by monoclonal antibody Ki-67, PR=progesterone receptor.
of malignancy in the mass reinforcement. While in the non-mass-like enhancement, cluster lesions had higher degree of differentiation, lower degree of malignancy than uneven lesions. Shu’s et al. study showed circular strengthening lesions which were not correlated with ER, PR, Her-2, and Ki-67, and partially were not consistent with our study results. This might be due to the different samples and the small sample size.

The DCE-TIC of this research was related to the expression of ER, Ki-67, and were not related to the expression of PR and Her-2. Figure 8 indicated lesions with flow-in DCE-TIC which had higher positive expression of ER, lower Ki-67 positive expression and higher degree of differentiation, and lower degree of malignancy than lesions with DCE-TIC platform. These lesion characteristics were similar to the flow-out DCE-TIC. In Zhang’s study, the DCE-TIC of breast cancer was partly a flow-out platform, while the DCE-TIC of benign lesions was partly flow-in platform. The breast mucinous adenocarcinoma is a special type in pathology, whose DCE-TIC type was flow-in, and has a relatively better prognosis, which was consistent with our result.

In this study, the expression of Her-2 was not correlated with DCE-MRI features. Our study results were consistent with Wang’s et al. study, which demonstrated no correlation of the mass with Her-2 expression. In some papers, the calcification of breast cancer showed correlation with the expression of Her-2. While in our study, there was no calcification observed in the MRI findings of breast cancer.

There are several limitations in our study. Firstly, the number of patients included was relatively small. Secondly, there were no
clinical subtypes and pathological grades in our study. Thirdly, our paper stated the correlation between DCE-MRI features and prognostic factors without further studying the molecular mechanism. Due to these shortcomings such as small sample size and less pathological types, further research is warranted.

5. Conclusions
In summary, this study shows that the diversity of MRI in breast cancer has some correlations with prognostic factors. This study provided a few of perspectives on imaging to evaluate the biological behavior and prognosis of breast cancer.

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