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A Case of an Invasive Lobular Carcinoma with Extracellular Mucin: Radio-Pathological Correlation

Shinya Tajima, Keiko Kishimoto, Yoshihide Kanemaki, Ichiro Maeda, Akira Endo, Motohiro Chosokabe, Takafumi Ono, Koichiro Tsugawa and Masayuki Takagi

Abstract

A case of 77-year-old female with an invasive lobular carcinoma with extracellular mucin is presented. She felt palpable mass in her left breast. Then, she came to our hospital for further examination. Mammography of right in full view revealed architectural distortion in left upper portion. And ultrasonography demonstrated low-echoic mass about 2 cm in diameter and invasion of the fat tissue was observed. Hence, malignancy was suspected and magnetic resonance imaging (MRI) was performed. MRI findings showed irregular shaped and margined mass with small T2-high-signal intensity. These findings suggested invasive carcinoma with mucin. Because the cancer lesion was not large, partial mastectomy was performed. Interestingly, pathological diagnosis was invasive lobular carcinoma with extracellular mucin. Extracellular mucinous lesion was concordant with small T2-high-signal intensity. This type of carcinoma was previously reported only in three cases, and rare but important, because the treatment and prognosis might change by histological subtypes. We suggest one of the MRI special features of our case is not only irregular shaped and margined mass but also small T2-high-signal intensity. These MR findings might be one of the valuable findings for the diagnosis and differentiation between this type of carcinoma from other tumors.

Keywords: magnetic resonance imaging, breast, invasive lobular carcinoma, extracellular mucin, E-cadherin

1. Introduction

To discriminate between invasive lobular carcinoma and invasive ductal carcinoma is a big theme for both radiologically and pathologically. The limitation of radiologic imaging in the
detection and evaluation of invasive lobular carcinoma have been recognized for a long time. Whereas, advances in breast radiologic imaging present opportunities to improve the diagnosis of invasive lobular carcinoma.

On mammography, invasive lobular carcinoma is not likely to form calcifications. However, calcifications may be present in benign proliferative lesions such as sclerosing adenosis [1]. The most common manifestations of invasive lobular carcinoma are asymmetric density, irregular, or spiculated mass on mammography [2–4].

On ultrasonography, 60.5% of invasive lobular carcinomas produced “a heterogeneous low-echoic mass with angular or irregular margins and posterior acoustic shadowing.” The remaining tumors had various other sonographic characteristics, including 12% that were “ultrasonographically invisible.” The sensitivity of ultrasonography for tumors measuring less than 1 cm was 85.7%. Invasive lobular carcinoma of common type tended to produce “focal shadowing without a discrete mass,” whereas tumors with pleomorphic histology were seen as “a shadowing mass.” Tumors of the alveolar, solid, and signet-ring variant of invasive lobular carcinoma were most often manifested as a “lobulated, well circumscribed mass” [5]. Ultrasonography is useful and more accurate than mammography in diagnosing invasive lobular carcinoma [5]. However, it is difficult to narrow down the diagnosis of invasive lobular carcinoma.

Breast magnetic resonance imaging (MRI) has an overall sensitivity of 93% for detecting invasive lobular carcinoma, similar to the detection of breast cancers overall (90%). On MRI, tumor of smooth margin, or absence of smooth margin and the distribution of nonmass-like enhancement are the features of invasive lobular carcinoma. Invasive lobular carcinoma may present as ductal, segmental, regional, or diffuse patterns [6]. MR imaging is considered to be a useful tool for detecting invasive lobular carcinoma on radiologically.

Pathologically, the invasive lobular carcinoma includes not only classical type (Foote and Stewart advocated in 1946 [7]) but also variants of solid, alveolar, pleomorphic, tubulolobular, signet-ring, trabecular, and mixed types [8].

We encountered a tumor of invasive carcinoma coexisting with mucinous carcinoma-like lesion. At first, the differential diagnoses of this tumor are (i) mixed mucinous-ductal carcinoma, (ii) mucinous carcinoma with neuroendocrine feature, (iii) mucinous papillary neoplasms, and (iv) carcinoma of mixed type (lobular and ductal carcinoma). Lobular carcinoma has been considered a variant of mucin-secreting carcinoma with only intracytoplasmic mucin [9–11]. In common practice, a diagnosis of mucinous carcinoma or ductal carcinoma with mucinous features is often made in the presence of extracellular mucin, without immunohistochemical confirmation of the ductal phenotype [10]. However, final diagnosis was “invasive lobular carcinoma with extracellular mucin” which has been reported only in three cases in the English medical literature [9–11]. Accordingly, the current report is the fourth documented case in pathology, and in the viewpoint of radiology, this is the first case.
Taking into consideration of the above information, we will discuss the unique variant of “invasive lobular carcinoma with extracellular mucin” with radiopathological correlation.

2. Materials and methods

2.1. MR imaging protocol

MR imaging was performed using a 1.5T MR system (Achieva 1.5T, Philips Healthcare Nederland, Eindhoven, Netherland), and a synergy breast coil. Routine breast MR images were acquired as follows in the prone position: axial T2-weighted turbo spin echo (TSE) with STIR images (repetition time (TR)/echo time (TE) = 5175/60 ms, section thickness = 5 mm, FOV = 280 mm, matrix = 480 × 480), and axial diffusion-weighted (DW) images using a single-shot EPI (FOV = 280 mm, matrix = 128 × 128), with MPG pulse applied along three directions (x, y, and z axes) with TR/TE of 5532/80 ms and b-factors of 0, 1000, and 2000 s/mm². Apparent diffusion coefficient (ADC)-maps were automatically generated on a postprocessing workstation. After axial T1-weighted SE images, [TR/TE = 468.8/12 ms, flip angle = 90, section thickness = 5 mm, FOV = 280 mm, matrix = 128 × 128] were obtained, axial 3D dynamic contrast enhanced T1-weighted images with SPAIR using eTHRIVE sequence [TR/TE = 5.9/2.8 ms, flip angle = 10, section thickness = 5 mm, FOV = 280 mm, matrix = 512 × 512] were performed in precontrast, double arterial phase, and delayed phase.

2.2. Image interpretation

For image interpretation of mammography, ultrasonography, and magnetic resonance imaging, we used the breast imaging and reporting data system (BI-RADS) lexicon and associated report of Tozaki et al. [12, 13].

2.3. Hematoxylin and eosin staining

Operation samples obtained from the partial mastectomy were fixed at least 8 h in 10% neutral phosphate buffered formalin, then, embedded in paraffin, and sectioned at 3–4 μm. The paraffin-embedded specimens were stained with hematoxylin and eosin (HE) for light microscopic examination.

2.4. Immunohistochemistry

Paraffin-embedded sections measuring 3–4 μm thick were deparaffinized. Antigen retrieval was then performed for 20–40 min in boiling water (95°C) containing citric acid solution (pH 6.0) or Target Retrieval Solution (pH 9.0; cat. no. 415211; Nichirei, Tokyo, Japan), and (pH 6.0; cat. no. S2031; Dako, Glostrup, Denmark). Sections were pretreated with 0.3% peroxide and reacted with primary monoclonal antibodies against estrogen receptor (ER), progesterone
receptor (PgR), HER2, Ki67, E-cadherin, Synaptophysin, MUC1, and MUC3 are shown in Table 1. Subsequently, specimens were incubated with a secondary antibody (Histofine simple stain MAX-PO; cat. no. 424154; Nichirei). The chromogenic substrate was 3,3′-diaminobenzidine (DAB), and the slides were counterstained with hematoxylin.

<table>
<thead>
<tr>
<th>Antibody</th>
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</table>

Table 1. Primary monoclonal antibodies against ER, PgR, HER2, Ki67, E-cadherin, synaptophysin, MUC1, and MUC3.

3. Results

A case of 77-year-old female is presented. She felt palpable mass in her left breast since 1 month and came to our hospital for further examination.

Physical examination revealed a 2.7 cm palpable mass in left upper inner and outer quadrants without nipple discharge and lymphadenopathy. No familial history of breast cancer was confirmed.

On mammography, the breast appeared dense. However, mediolateral oblique (MLO) view of the mammography demonstrated architectural distortion in left upper portion (Figure 1). Hence, this lesion was of four categories. Right MLO view revealed no evidence of malignancy.

Ultrasonography revealed a low-echoic mass about 2 cm in diameter with posterior acoustic shadowing, and disruption of the anterior border line (the anterior border line means boundary line between fat tissue layer and breast parenchymal layer) of the mass was observed in her left breast (Figure 2). There was no lesion suggesting malignancy in her right breast. No lymph node swelling was seen.

These findings suggested the possibility of malignancy. Therefore, magnetic resonance imaging (MRI) of 1.5T (Philips) was performed. On MRI, diffusion-weighted image of b-value 1000 s/mm² showed mass-like lesion with high signal intensity (Figure 3A). Apparent diffusion coefficient (ADC)-map revealed diffusion limitation of the water molecule (Figure 3B). Dynamic contrast-enhanced T1 weighted images demonstrated early enhancement and delayed washout pattern of irregular shaped and margined mass about
2 cm in diameter (Figure 3C and D). The mass included a small high signal intensity area on fat-saturated T2 weighted image, which was confirmed pathologically as extracellular mucin. (Figure 3E). Dynamic-curve of the contrast-enhanced MRI showed rapid wash-out pattern (Figure 3F). ADC value was $0.577 \times 10^{-3} \text{mm}^2/\text{s}$; hence, the existence of water restriction was suggested.

Neoadjuvant chemotherapy was not performed. Then, partial mastectomy was performed because of the imaging findings. Sentinel lymph node was negative for metastatic cancer lesion. Further, axillary lymph node resection was not performed. Histopathological findings revealed low-grade tumor with little or no nuclear atypia and inconspicuous nucleoli. Besides, the tumor was composed of single cell with moderate pleomorphism and lack of cohesion. The neoplastic cells were arranged in files or cords infiltrating the stroma. Partially, trabecular type lesion was observed (Figure 4A). Within the tumor, there were areas showing signet-ring cells floating in a pool of mucin (Figure 4B). The presence of the extracellular mucin is not a characteristic of lobular carcinomas. Therefore, immunohistochemical stain for E-cadherin was performed. However, the lesions were negative for E-cadherin immunostaining (Figure 4C). Besides, neuroendocrine marker of synaptophysin was completely negative. MIB1 (Ki67)-index was about 10%. Estrogen receptor (ER), progesterone receptor (PgR), and human epidermal growth factor receptor 2 (HER2) status were 99, 0%, and negative, respectively. Additionally, special stainings for mucin as MUC1 and MUC3 were performed, and both stainings were positive of both infiltrating cells and signet-ring cells floating in a pool of mucin (Figure 4D and E).
Figure 2. Ultrasonography reveals a low-echoic mass, posterior acoustic shadowing, and disruption of the anterior border line is observed.

Figure 3. (A) Diffusion-weighted image of b=1000 shows mass like high signal intensity. (B) Apparent diffusion coefficient (ADC)-map reveals diffusion limitation of the water molecule. (C) Contrast-enhanced T1 weighted images demonstrate early enhancement and (D) delayed washout pattern. (E) The mass included a small high-signal intensity on fat-saturated T2 weighted image, which is confirmed pathologically as extracellular mucin (arrow). (F) Dynamic-curve of the MRI shows rapid washout pattern.
4. Discussion

The major types of invasive carcinomas are categorized as ductal carcinoma. Invasive lobular carcinoma is the second most common histological type of breast carcinoma, accounting for about 5–15% of all invasive breast cancers \[14, 15\]. Hence, accurate diagnosis of invasive lobular carcinoma is thought to be important. We will discuss about general radiological findings of invasive lobular carcinoma and correlation of our case.

In mammography, Berg et al. \[16\] examined the performance of mammography as a function of both tumor type and breast density. Mammographic sensitivity was 81% for invasive ductal carcinoma compared with 34% for invasive lobular carcinoma; when only those patients with dense breast tissue were considered, sensitivities decreased dramatically to 60 and 11%, respectively \[16\]. Because of these diagnostic interests, it is crucial for breast radiologists to be aware of the asymmetric and subtle mammographic features of invasive lobular carcinoma.

A high-density spiculated or irregular margined mass on mammography means invasive carcinoma. Spicula or irregular margin is thought to be invasion of the breast fat tissue and stromal reaction that disrupts normal breast parenchymal architecture. common mammographic manifestations of invasive lobular carcinoma include spiculated or irregular margined mass or asymmetric density. In our case, these findings are not observed. Apart from spiculated, irregular margined masses and asymmetric densities, the most common mammographic manifestation of invasive lobular carcinoma is architectural distortion, which accounts for about 14–25% of cases of mammographically detected invasive lobular carcinoma \[3, 17, 18\].

Figure 4. (A) The neoplastic cells are arranged in files or cords infiltrating the stroma. Partially, trabecular type lesion is observed. (B) Within the tumor, there are areas showing atypical cells floating in a pool of mucin. (C) All the tumor cells are negative for E-cadherin immunostaining. (D) All the tumor cells and extracellular mucin are MUC1 positive. (E) All the tumor cells and extracellular mucin are MUC3 positive.
Similarly, our case demonstrated architectural distortion on left upper portion in MLO view. We think distortion on mammography means stromal reaction and malignancy.

Ultrasonography of the breast is used primarily as a diagnostic imaging tool. The most common sonographic appearance of invasive lobular carcinoma is a low-echoic mass with posterior acoustic shadowing occurring in up to 60% of cases, however, posterior acoustic shadowing may be lacking in up to 20% of cases [19]. The present case of ultrasonographical findings is thought to be concordant with report of Karen et al. [19].

Here, we will discuss correlation between MRI findings and pathological findings. Our case of mammographic and ultrasonographic findings suggested the possibility of malignancy. Therefore, magnetic resonance imaging (MRI) of 1.5T (Philips) was performed. On MRI, diffusion-weighted image of b-value 1000 s/mm² showed mass-like high signal intensity. Apparent diffusion coefficient (ADC)-map revealed diffusion limitation of the water molecule. Besides, irregular shaped and margined mass was observed in T1 weighted image. These findings suggest malignancy in our case. Furthermore, this finding is similar to the report of Mann et al. of invasive lobular carcinoma [6]. Fat-saturated T2 weighted image of the mass showed small high signal intensity area within the mass. This finding suggests the tumor associated mucinous lesion or coexistence of mucinous carcinoma. However, Rosa et al. [9] reported signet-ring cells floating in a pool of mucinous areas (extracellular mucinous lesion) represented at the most 20% of the tumor. We think this extracellular mucin area is not so large but a small area. If invasive carcinoma and mucinous carcinoma are coexisted as collision tumor, the mucinous area which shows very high signal intensity on T2 weighted image would be larger.

In our case, extracellular mucin area was approximately 10% of the tumor. This small area of high signal intensity on T2 weighted image might be one of the specific features of invasive lobular carcinoma with extracellular mucin. Dynamic-curve of the contrast-enhanced MRI showed rapid washout pattern. Rapid washout pattern is also seen in invasive ductal or lobular carcinoma. ADC value was $0.577 \times 10^{-3}$ mm²/s, hence the existence of water restriction was suggested. From these findings, dynamic-curve, diffusion-weighted image, and ADC-map were suggestive for malignant tumor. And contrast-enhanced T1 weighted image of irregular shaped and margined mass was indicative of invasive carcinoma such as invasive ductal carcinoma or invasive lobular carcinoma. T2 weighted image of small high signal intensity area is thought to be mucinous carcinoma or mucin production. And mucin production is known to be associated with neuroendocrine differentiation. Taking these findings into consideration, other differential diagnoses of this tumor were (i) mixed mucinous-ductal carcinoma, (ii) mucinous carcinoma with neuroendocrine feature, (iii) mucinous papillary neoplasms, and (iv) carcinoma of mixed type (lobular and ductal carcinoma). However, histopathological findings revealed the tumor was composed of single cell with moderate pleomorphism and lack of cohesion. The neoplastic cells were arranged in files or cords infiltrating the stroma. In situ lesion of the invasive lobular carcinoma was not observed. The multifocality of cancer lesion was not demonstrated. Within the tumor, there were areas showing signet-ring cells floating in a pool of mucin. The presence of extracellular mucin is not characteristic of lobular carcinomas. Therefore, immunohistochemical stain for E-cadherin was performed. However, the lesions were completely negative for E-cadherin immunostaining. E-cadherin, a cell-cohesion protein encoded by a gene on chromosome 16q22.1, is the current marker of choice to
help discriminate between lobular and ductal carcinoma [11]. Neuroendocrine marker of syn-
aptophysin immunostaining was also negative. Hence, final diagnosis of our tumor is inva-
sive ductal carcinoma with extracellular mucin. From the results of MRI findings, irregular
shaped and margined mass with small area of fat-saturated T2 high signal intensity might be
one of the special feature of invasive lobular carcinoma with extracellular mucin. Our case is
the fourth case in the English literature. Because the number of cases of these tumors is lim-
ited, it is difficult to comment on the biological behavior and molecular profiles. However, this
diagnosis is important for prognosis and management, and further examination is needed.

In our case, estrogen receptor (ER), progesterone receptor (PgR), and human epidermal
growth factor receptor 2 (HER2) status were 99, 0%, and negative, respectively. Invasive lobu-
lar carcinoma is known to be more commonly estrogen receptor-positive and HER2-negative,
in other words, invasive lobular carcinoma is usually categorized as luminal subtype. The
previous reports of Rosa et al. and Haltas et al. [9, 11] are ER positive of luminal subtype.
Our case is concordant with their results. However, the results of Yu et al. [10] were not only
ER-positive but also HER2 strongly positive. They thought that this type of HER2 status dem-
strate various features.

The spectrum of breast lesions that demonstrate extracellular mucin includes fibrocystic
change with luminal mucin, mucocle-like lesions, papillary lesions with mucin secretion,
and mucinous carcinoma among others [9]. In contrast, lobular neoplasia and invasive lobu-
lar carcinoma demonstrate intracytoplasmic mucin, and no cases of lobular carcinoma with
extracellular mucin have been found except previous three reports [9–11, 20].

Invasive lobular carcinoma comprises 5–15% of invasive breast tumors [14, 15]. They can occur
at any age. The median age at diagnosis is between 45 and 56 years and there is a tendency
to affect older patients compared with ductal carcinoma [8, 9], similar to our results of the
77-year-old. The presenting symptom in most cases is a mass with irregular margins or a breast
thickening with diffuse nodularity [8, 9]. The clinical features of invasive lobular carcinoma are
also different from that of ductal carcinomas. Invasive lobular carcinomas tend to be more often
multifocal and bilateral [8, 9], with a distinct metastatic pattern and a higher frequency of bone,
gastrointestinal tract, uterus, meninges, and diffuse serosal involvement [21]. Hence, discrimi-
nation between lobular carcinoma and ductal carcinoma is important clinically, radiologically,
and pathologically. In our case, multifocality of the tumor was not observed similarly to the
reports of Rosa et al. and Yu et al. [9, 10]. However, the case of Haltas et al. [11] indicated mul-
tifocality. We think this is because the variant of invasive lobular carcinoma with extracellular
mucin might present different behavior compared with classical invasive lobular carcinoma.

Mucin has been classified as membrane-bound mucin, which mediates signal transduction,
and secretory mucin, which are directly secreted into extracellular spaces. In the MUC immu-
nostaining family, the membrane-bound mucins include MUC1, MUC3, MUC4, and the sec-
retary mucins include MUC2, MUC5AC, MUC6 [22]. In our current case, MUC4, MUC5AC,
and MUC6 were negative; however, all the tumor cells revealed cytoplasmic expression of
MUC1. Molecular and biochemical studies have demonstrated that MUC1 is involved in the
inhibition of E-cadherin mediated -cell and cell-matrix adhesion [10, 22–25]. The cytoplasmic
domain of MUC1 molecule has been shown to inhibit the formation of E-cadherin-β-catenin
complex [10, 24]. Therefore MUC1 may play a role in tumor invasion and metastases by disrupting cell adhesions [10]. Similarly, our case of tumor invasion may correlate with the MUC1 cytoplasmic expression.

Additionally, our case of all the tumor cells was positive for MUC3 immunostaining. Rakha et al. and Furuya et al. [26, 27] reported that MUC3 immunostaining is useful for distinguishing between benign lesion and malignant lesion of the breast carcinoma. Our case is concordant with their reports, and membrane-bound mucin of MUC3 may mediate signal transduction correlate with malignancy. Furthermore, Rakha et al. [26] indicated that most breast carcinomas express MUC1, MUC3, and MUC4; however, MUC1 and MUC3 are potential prognostic indicators. Hence, diagnosing invasive lobular carcinoma with extracellular mucin is important on not only pathologically but also radiologically.

Early genomic studies revealed very little overall difference in genomic profiles between low-grade invasive ductal carcinoma and classical invasive lobular carcinoma, implying that classical invasive lobular carcinoma might represent a subtype of low-grade invasive ductal carcinoma [10]. Recent gene expression studies comparing invasive lobular carcinoma and invasive ductal carcinoma have identified two subsets of invasive lobular carcinoma with distinct transcription patterns [10]. Approximately, half of the invasive lobular carcinomas differs from invasive ductal carcinomas in gene expression profiles (“typical” invasive lobular carcinomas), while the remaining invasive lobular carcinomas closely resemble invasive ductal carcinomas in transcription patterns. (“ductal-like” invasive lobular carcinomas) [10]. On the other hand, a recent study on grade- and molecular subtype-matched invasive lobular carcinomas and invasive ductal carcinomas of no special type demonstrated that invasive lobular carcinomas had different transcriptomic profiles in the genes related to cell-to-cell adhesion and signaling, as well as actin cytoskeleton signaling, when compared with grade- and molecular subtype-matched invasive ductal carcinomas [10]. This finding suggested that even though invasive lobular carcinomas and invasive ductal carcinomas might present as a spectrum or form of a family [10]. Taking into consideration of the above Yu et al. reported case, our current case might be in the middle stage of the spectrum between lobular carcinoma and ductal carcinoma. We think existence of extracellular mucin is not definitive for ductal phenotype not only histologically but also genetically.

5. Conclusion

We encountered the distinct variant of invasive lobular carcinoma with extracellular mucin. We described the correlation of its radiological and pathological interest. Radiological findings of invasive lobular carcinoma with extracellular mucin are documented in English literature for the first time. We suggest one of the MRI special features of our case is not only irregular shaped and margined mass but also small T2-high-signal intensity. These findings by the knowledge from radiopathological correlation might be one of the specific features of invasive lobular carcinoma with extracellular mucin. Further examinations are needed to clarify this lesion.
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