

CASE REPORT

CLINICAL AND PATHOLOGICAL ANALYSIS OF INVASIVE MUCINOUS MICROPAPILLARY CARCINOMA OF THE BREAST: A SERIES OF CASE REPORTS

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The aim of the study was to analyse the clinicopathological features of invasive mucinous micropapillary carcinoma (MUMPC), a rare breast cancer variant. We retrospectively reviewed 7 MUMPC cases diagnosed in 2017–2024, evaluating clinical, imaging, histopathological, and immunohistochemical data (including epithelial membrane antigen, hormone receptors, human epidermal growth factor receptor 2, and Kiel 67 antigen). The cohort comprised 7 female patients aged 40–77 years (mean, 53.8 years). Tumour size range was 0.9–6.0 cm (mean, 3.4 cm). Histologically, all tumours exhibited classic features: abundant extracellular mucin containing tumour cells arranged in micropapillary, glandular, and cluster patterns, with characteristic retraction-like spaces. Regarding nuclear grade, 5 cases showed low-to-intermediate-grade nuclei, while 2 cases exhibited high-grade nuclei. Immunophenotypically, 6 cases demonstrated strong positivity for oestrogen and progesterone receptors (50–95% of cells). The human epidermal growth factor receptor 2 status was negative (score 0) in 4 cases, equivocal (2+) in 2 cases (both confirmed to have amplification by fluorescence *in situ* hybridisation), and positive (3+) in 1 case. Two patients had lymph node metastasis; 5 patients remained recurrence-free at 60 months of follow-up, 1 patient died at 17 months, and 1 patient was newly diagnosed. Mucinous micropapillary carcinoma displays distinct clinicopathological and immunophenotypic features, suggesting potentially more aggressive behaviour than pure mucinous carcinoma. Accurate diagnosis and individualised management are essential.

Key words: breast cancer, mucinous carcinoma, diagnosis, prognosis, case report.

Introduction

Pure mucinous carcinoma (PMC) of the breast is characterised by tumour cells with mild morphology, minimal cellular atypia, abundant extracellular mucin, and a favourable prognosis [1]. Extracellular mucin may restrict tumour spread [2]. In contrast, invasive micropapillary carcinomas (IMPC) are highly aggressive, with high-grade nuclei, high rates of lymph node metastasis and vascular invasion, and a poor prognosis [3]. In 2002, Ng [4] first reported cases of PMC with a micropapillary pattern (mucinous carcinomas with micropapillary pattern; or mu-

cinous micropapillary carcinoma – MUMPC), proposing it as a new subtype of mucinous carcinoma. Invasive MUMPCs are rare and exhibit the histological features of both invasive mucinous carcinoma and IMPC. There is currently no consensus on whether tumours with these characteristics should be classified as invasive mucinous carcinoma or IMPC with mucin production. Although MUMPC exhibits histopathological features suggestive of invasive mucinous carcinoma, some studies suggest it should be considered biologically as an IMPC [5].

Due to limited case reports and an insufficient understanding of breast invasive MUMPC, this study re-

ports 7 cases of breast invasive mucinous micropapillary carcinoma (MPMC) analysing, for example, their clinical and pathological features, as well as immunophenotypic characteristics, to improve the diagnostic level for this special type of breast cancer and optimise clinical treatment.

General data

Cases diagnosed at Ningbo Hospital of Integrated Traditional Chinese and Western Medicine, from January 2017 to May 2024, were collected. Clinical information, imaging data, and gross examination findings were obtained from the electronic medical record system. Follow-up data were obtained from the electronic medical record system and telephone communications with clinicians and/or patients. All cases in this study obtained patient-informed consent.

All tissue samples were fixed in 4% neutral formalin, embedded in paraffin, sectioned at 3–4 μm , stained with haematoxylin and eosin, and observed under a microscope. Immunohistochemistry was performed using the Envision method to detect proteins in paraffin sections. Antibodies used included epithelial membrane antigen (EMA), E-cadherin, human epidermal growth factor receptor 2 (HER2), oestrogen receptor (ER), progesterone receptor (PR), androgen receptor (AR), Kiel 67 antigen (Ki-67), cytokeratin 5/6, tumour protein p63, neuron-specific enolase, chromogranin A (CGA), and synaptophysin (SYN). The human epidermal growth factor receptor 2 antibodies were purchased from Roche (Roche Diagnostics International Ltd, Rotkreuz, Switzerland), and all other antibodies were obtained from Fuzhou Maixin Biotechnology Development Co., Ltd. (Fuzhou, Fujian, China). Staining procedures were performed according to the kit manufacturer's instructions. Immunohistochemistry included positive and negative controls. The interpretation criteria were as follows: EMA positivity at the outer edge of micropapillae and loss of E-cadherin expression at the outer edge of micropapillae; nuclear ER, PR, AR, and Ki-67 staining, and the percentage of positive cells was calculated. Neuron-specific enolase, CGA, and SYN positivity were cytoplasmic. The human epidermal growth factor receptor 2 scoring was performed according to the 2018 College of American Pathologists breast cancer HER2 testing guidelines [6]. Cases scored as HER2 (2+) were tested for gene amplification using fluorescence *in situ* hybridisation (FISH). The *human epidermal growth factor receptor 2* gene amplification detection kit was provided by Guangzhou Anbiping Medical Technology Co., Ltd. (Guangzhou, Guangdong, China). The *in situ* hybridisation heater was obtained from Fuzhou Maixin Biotechnology Co., Ltd. (Fuzhou, Fujian, China). Procedures and result interpretation followed the kit manufacturer's instructions.

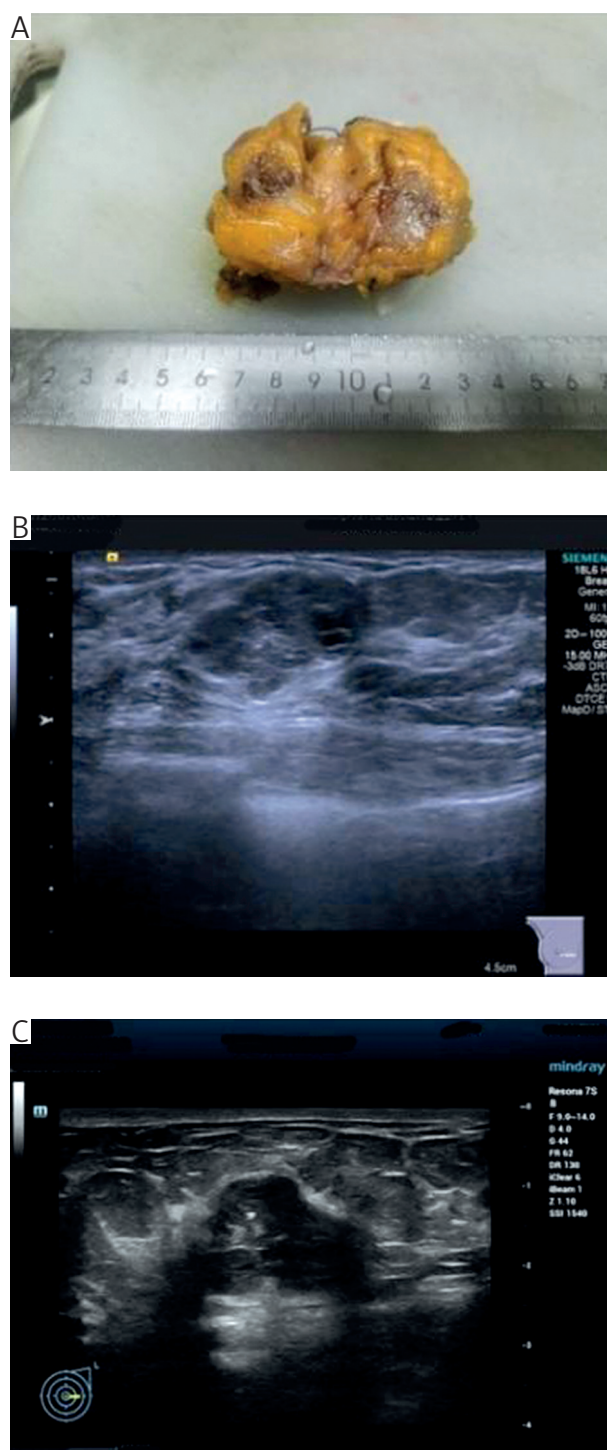


Figure 1. A) Gross specimen of a resected tumour. The cut surface appears translucent, gelatinous, and soft, with a greyish-white to greyish-pink colour, which is characteristic of mucin-rich neoplasms. B) Typical breast B-ultrasound image of mucinous micropapillary carcinoma (MUMPC). The tumour presents as a well-defined, oval-shaped, hypoechoic nodule with heterogeneous internal echogenicity and anechoic areas corresponding to mucin pools. No calcification is observed. C) Breast ultrasound image of a more aggressive MUMPC. The tumour exhibits irregular margins, an irregular shape, parallel orientation, heterogeneous internal echogenicity, visible calcifications, mixed posterior acoustic enhancement, and peripheral vascularity, and is suspicious for axillary lymph node metastasis.

Results

Clinical results: all 7 patients were women, with an average age of 53.8 years (ranging 40–77 years). Three tumours were located in the left breast, and 4 in the right breast. All cut surfaces were translucent, gelatinous, greyish-white, greyish-pink, and soft (Figure 1A). Five cases had clear boundaries, appearing encapsulated. Two cases had unclear boundaries and irregular shapes. Breast ultrasound showed 5 tumours as well-circumscribed hypoechoic nodules, without thickened peripheral hyperechoic zones, having mixed cystic and solid components, with internal anechoic areas representing fluid or mucin, posterior acoustic shadowing, and no blood flow signals (Figure 1B). Two tumours had ill-defined margins, irregular shapes, parallel growth, heterogeneous internal echogenicity, mixed posterior acoustic enhancement, peripheral blood flow signals, and suspicious axillary lymph nodes (Figure 1C). Calcifications were seen in 3 cases.

The magnetic resonance imaging findings were as follows: MUMPC mostly showed washout enhance-

ment patterns. The tumour centre showed low signal intensity on T1-weighted images and high signal intensity on T2-weighted images.

Postoperative follow-up yielded the following: five cases showed no evidence of recurrence or metastasis within a 60-month follow-up period; 1 case died after 17 months of follow-up, and 1 case was newly diagnosed.

Microscopic observations showed the following: in a rich, extracellular mucin pool, tumour cells were arranged in clusters and nests, were pseudoglandular, and had micropapillary patterns, separated by delicate fibrous and capillary septa into nodular formations, resembling a mucin-rich fibroadenoma (Figure 2A). Cancer cells showed infiltrative growth with unclear boundaries to the surrounding tissue. Micropapillary structures varied, appearing as morula-like, clustered, or pseudoglandular, lacking fibrovascular cores and infiltrating fibromucinous stroma. Micropapillary arrangements varied in size; large papillae consisted of 10–20 cells (Figure 2B), while small papillae consisted

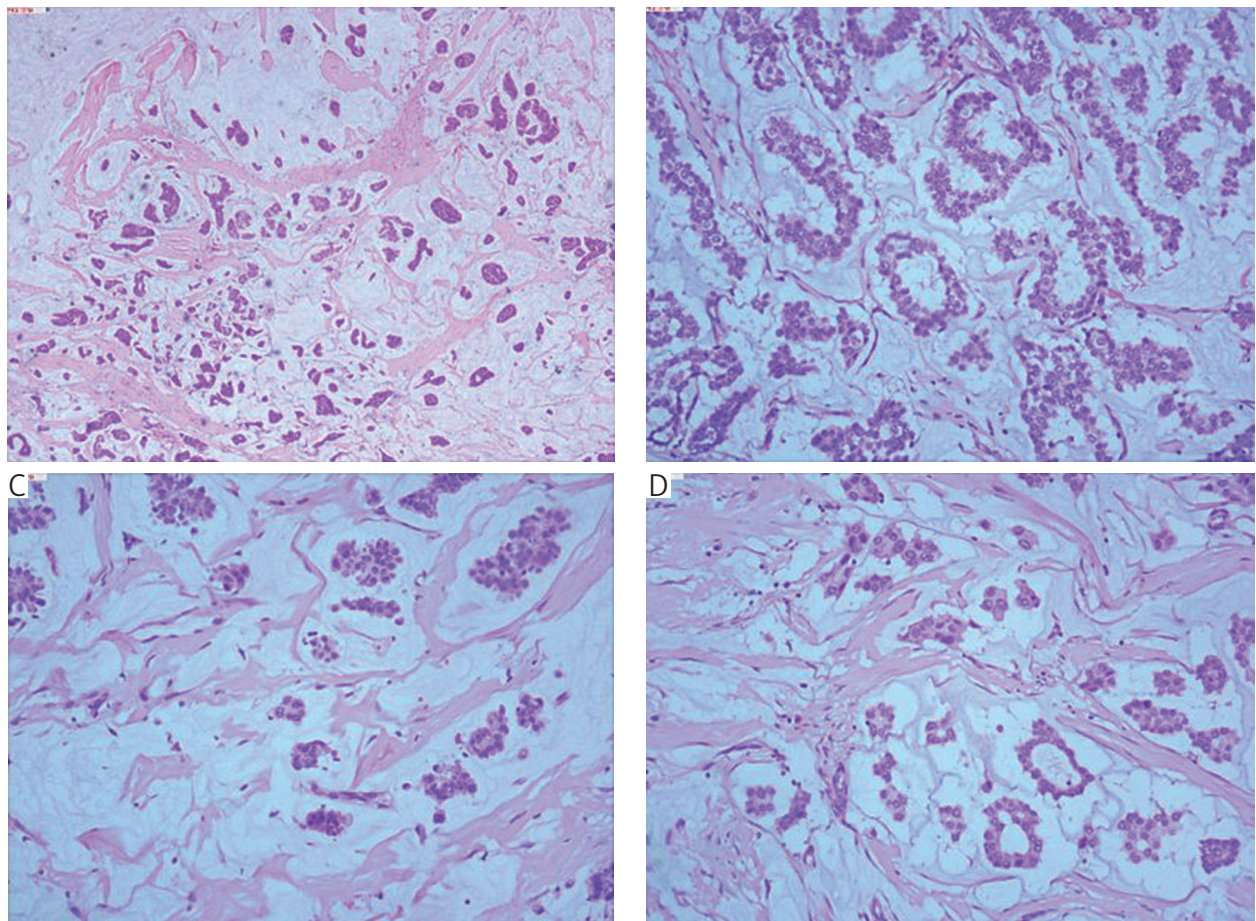


Figure 2. Histopathological characteristics of invasive mucinous micropapillary carcinoma. **A)** Low-power view. In a rich extracellular mucin pool, tumour cells are arranged in clusters, nests, pseudoglandular, and micropapillary patterns, separated by delicate fibrous septa into nodular formations. Resembling a mucin-rich fibroadenoma. **B)** Cancer cells arranged in micropapillary patterns of varying sizes. Large papillae consist of 10–20 cells. **C)** Tumour cells arranged in micropapillary patterns of varying sizes. Small papillae consist of 4–10 cells. **D)** Tumour cell nuclei are small, centrally located, hyperchromatic; low-intermediate grade nuclei, mitotic figures rare (medium-power magnification, HE)

of 4–10 cells (Figure 2C). The distribution of micropapillary structures in the mucin lakes was uneven. Some areas had abundant mucin with no visible tumour cells. Some areas resembled cell-rich mucinous carcinoma. Often, gaps existed between micropapillary structures and mucin pools; some gaps appeared ring-like, while others were formed by connected vacuoles. A characteristic feature was tumour cell clusters surrounded by retraction artifact-like gaps filled with mucin, resembling lymphatic or vascular spaces but lacking an endothelial cell lining. The cytoplasm of cancer cells was eosinophilic, cuboidal or columnar, relatively uniform in morphology and size, with indistinct borders, and some appeared syncytial-like. In five cases, tumour cell nuclei were small, centrally located, and hyperchromatic, corresponding to low-intermediate grade nuclei; small nucleoli were sometimes visible, and mitotic figures were rare. Tumour necrosis was absent (Figure 2D). In 2 cases, cancer cells had high-grade nuclei and were large and irregular in shape, with thick nuclear membranes, and some chromatin appeared coarse-granular. The cytoplasm was typically amphophilic. Features resembling apocrine secretory protrusions (seen in tubular carcinomas) were also observed. Some nuclei were compressed to one side by mucin, giving them a signet-ring appearance. Some tumour cells had cytoplasmic protrusions at their periphery; after detachment, these protrusions would become naked nuclei.

Immunohistochemical staining and FISH results: among the 7 tumours, 6 were ER and PR positive (50–95%); 1 case showed weak ER and PR positivity (5–10%); 6 cases were strongly AR positive, and 1 case was negative. Four cases were HER2 (0), 2 were

HER2 (2+), and 1 was HER2 (3+). The Kiel 67 antigen positive expression rate was 10–45%. A significant proportion of MUMPC showed neuroendocrine differentiation: 2 cases were CGA positive, and 3 cases were SYN positive. Epithelial membrane antigen positivity was observed at the outer edge or stromal side of micropapillae or pseudoglands, in contrast to the expression pattern in adenocarcinoma, showing inverted polarity (Figure 3A). E-cadherin was primarily expressed at the intercellular junctions of micropapillae or pseudoglands, with markedly reduced or absent expression on the stromal-facing side (Table I). Both cases with HER2 immunohistochemistry result (2+) underwent FISH testing, and both showed gene amplification (Figure 3B).

Discussion

Invasive MPMC is clinically rare and is a special type of breast mucinous carcinoma, defined as PMC containing micropapillary components [7]. Patients with MUMPC tend to be younger than those with general mucinous carcinoma. Ginter *et al.* [8] suggested that MUMPC onset tends to be in younger individuals; in their study of 14 MUMPC cases, 43% (6/14) were diagnosed below the age of 45 years old, significantly younger than the median age of all invasive mucinous carcinomas. As 45 years is a standard for age stratification in breast cancer and often considered an indicator of genetic risk, patients younger than 45 years may have a degree of genetic risk [9]. In our study, 6 out of 7 tumours were observed in patients aged 40–50 years, with one at 77 years, consistent with literature reports. Yang *et al.* [10] described the imaging

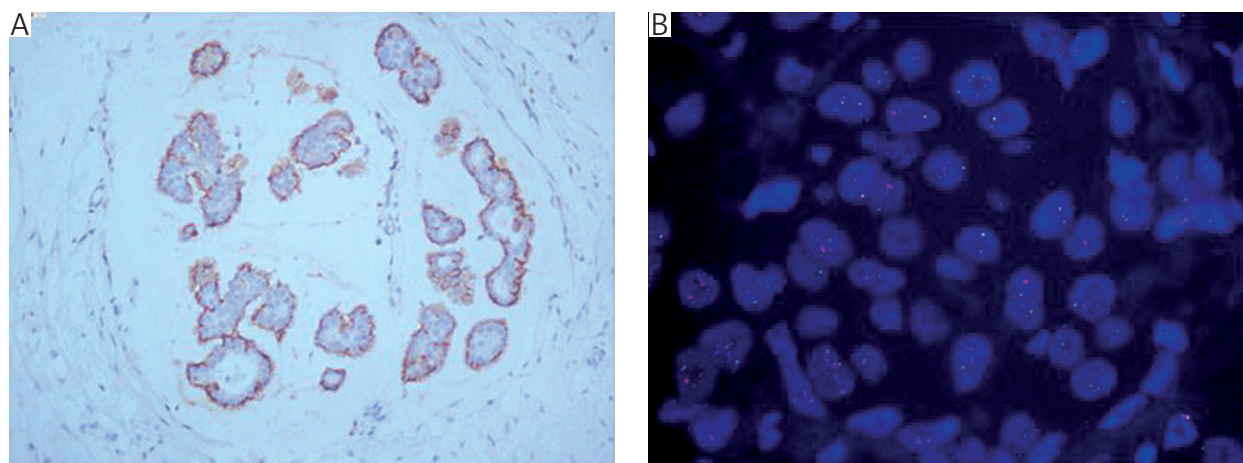


Figure 3. **A)** Immunohistochemistry for epithelial membrane antigen (EMA) (high-power field, HE staining). EMA positivity (brownish-yellow staining) is predominantly localised to the stromal-facing (outer) surface of the tumour cell clusters, demonstrating the characteristic 'inverted polarity' pattern, which is a hallmark feature of micropapillary differentiation. **B)** Fluorescence *in situ* hybridization (FISH) for *human epidermal growth factor receptor 2* (*HER2*) gene amplification (low-power view, FISH method). Red signals represent the *HER2* gene, and green signals represent the chromosome 17 centromere (CEP17). The case shows a high *HER2*/CEP17 signal ratio (> 2.0), with clusters of red signals indicative of *HER2* gene amplification. This corresponds to an immunohistochemistry score of 2+

Table I. Immunophenotype of seven mucinous micropapillary carcinoma tumour cells

CASE	AGE	ER (%)	PR (%)	HER-2	Ki-67 (%)	CGA	SYN	CD56	E-CADHERIN	EMA	AR
1	48	Weak 5	Weak 10	3+	45	–	–	–	+	+	–
2	45	Moderate 80	Strong 90	2+	30	+	+	–	+	+	+
3	42	Strong 95	Strong 70	0	20	–	+	–	+	+	+
4	40	Moderate 70	Moderate 60	2+	25	–	–	–	+	+	+
5	77	Moderate 60	Moderate 50	0	10	+	–	–	+	+	+
6	50	Strong 90	Strong 95	0	15	–	–	–	+	+	+
7	47	Strong 90	Moderate 80	0	12	–	+	–	+	+	+

AR – androgen receptor, CD56 – cluster of differentiation 56, CGA – chromogranin A, EMA – epithelial membrane antigen, ER – oestrogen receptor, HER2 – human epidermal growth factor receptor 2, Ki-67 – Kiel 67 antigen, PR – progesterone receptor, SYN – synaptophysin

features of invasive MPMC, noting that B-ultrasound characteristics were very similar to fibroadenoma with myxoid change: well-circumscribed oval masses, hypoechoic, with a peripheral hyperechoic ring, no definite blood flow signals, and clinically easily misdiagnosed as benign lesions. This is a reminder to clinicians that when B-ultrasound shows a breast fibroadenoma, this type of tumour should be considered. In our group, 5 tumours were well-circumscribed hypoechoic nodules without thickened peripheral hyperechoic zones and no blood flow signals. Two tumours had ill-defined margins, irregular shapes, parallel growth, heterogeneous internal echogenicity, peripheral blood flow signals, and suspicious axillary lymph nodes. Microscopically, mucinous carcinoma shows small clusters of tumour cells floating in mucin lakes, with tumour cells lacking atypia, no mitotic figures, mucinous material in the gaps between tumour cell clusters, and smooth cluster peripheries. Studies have reported the histological characteristics of MUMPC structures as including tumour cell growth patterns similar to IMPC, with variable amounts of mucin occupying retraction gaps around tumour cells, tumour cells arranged in petal-like micropapillary structures, and cancer cells floating in mucin lakes. Tumour cells exhibit an inverted polarity arrangement, which can be demonstrated by the expression pattern of mucin 1/EMA on the cell surface facing the surrounding extracellular mucin [11]. In our study, 7 cases showed infiltrative growth of cancer cells with unclear boundaries to the surrounding tissue. Cancer cells formed various micropapillary structures, some morula-like, some clustered, and some pseudoglandular, lacking fibrovascular cores and infiltrating the fibromucinous stroma. Studies have reported that mucinous carci-

nomas with diffuse micropapillary structures exhibit low-grade nuclei [12, 13], while nuclei of high, intermediate, and low grades have been observed across cases, with the majority being intermediate grade. Our cases also had low, intermediate, and high-grade nuclei in mucinous carcinoma with micropapillary structures observed. Five cases had small, centrally located, hyperchromatic nuclei, corresponding to low/intermediate grade nuclei. Two cases had high-grade nuclei and were large and irregular in shape, with thick nuclear membranes.

Pure mucinous carcinoma is characterised by high hormone receptor expression and low HER2 expression. The immunophenotype of invasive MPMC still needs further investigation. Liu *et al.* [14] reported that, although the majority of invasive MUMPC retained a luminal-like immunophenotype with high ER/PR positivity and low HER2 expression, a subset of cases exhibited HER2 overexpression/gene amplification (11.9%) and occasional PR negativity. In their cohort, HER2 status was identified as an independent predictor of poorer recurrence-free and overall survival in invasive MUMPC patients, suggesting that this immunophenotypic variation carries prognostic significance and distinguishes invasive MUMPC from conventional pure mucinous carcinoma. Immunohistochemically, the EMA positivity in these 7 invasive breast MPMCs was at the outer edge or stromal side of micropapillae or pseudoglands, in contrast to adenocarcinoma. E-cadherin was primarily expressed at the intercellular junctions of micropapillae or pseudoglands, with negative expression on the stromal-facing side. The human epidermal growth factor receptor 2 positive expression pattern was similar to E-cadherin, consistent with ordinary breast IMPC. Among the

7 tumours, 6 showed strong ER and PR positivity (50–95%). One case showed weak ER and PR positivity (5–10%); 4 cases were HER2 (0), 2 cases were HER2 (2+), 1 case was HER2 (3+); Ki-67 (10–45%). The human epidermal growth factor receptor 2 positive expression rate in this study was higher than reported in the literature. Literature reports [15] suggest that MPMC has a higher Ki-67 proliferation index than ordinary mucinous carcinoma, tends towards having a higher histological grade, and the lymph node metastasis rate is proportional to the Ki-67 proliferation index. However, the Ki-67 proliferation index in our 7 patients was relatively low, with lymph node metastasis seen in 2 cases.

The genetic alterations of MUMPC are intermediate between PMC and invasive micropapillary carcinoma (IPMC) [4]. *TP53* and *PIK3CA* are common mutation genes in ER-positive breast cancer, with high mutation frequencies in IPMC, 1q amplification, and 16q deletion. Pareja *et al.* [16] found a 16q deletion in 2 out of 5 MUMPC cases and low *PIK3CA* mutation frequency in 3 cases. Miricescu *et al.* [17] detected 10 MUMPC and 11 PMC samples, as well as alterations in PI3K-Akt, mTOR, and focal adhesion pathways in MUMPC, pathways associated with invasive metastasis in various cancers. Accordingly, MUMPC may be genetically distinct from PMC, and specific genomic alterations may be the cause of its aggressive behaviour.

A differential diagnosis is necessary to distinguish invasive MUMPC from non-invasive MUMPC, in which the following should be considered:

1. Generally, MUMPC has a better prognosis than IMPC, which is a highly aggressive breast cancer. Cancer cells are arranged in micropapillary structures, often with a fan-shaped or spiky outer edge, and some cancer cells show cytoplasmic protrusions at their periphery, exhibiting a special 'inverted polarity' arrangement. Although MUMPC also has such micropapillary structures, its tumour cells float in mucin lakes, or the retraction gaps around cancer cells contain varying amounts of mucin. In contrast, IMPC generally lacks mucin in the stroma, which supports differentiation;

2. Fibroadenoma with myxoid degeneration: under low magnification, the epithelial cells of a fibroadenoma are separated by fibrovascular tissue into nodular or lobulated patterns, and the stroma is prone to myxoid degeneration, making it difficult to distinguish from MUMPC and easily misdiagnosed during frozen section diagnosis. However, fibroadenomas commonly occur in women of reproductive age, particularly under 30 years old. Tumour cells do not show 'inverted polarity' in EMA immunohistochemical staining. In contrast, MUMPC cancer cells show obvious nuclear atypia and a high nuclear grade, along with high ER expression and HER2-negative expression;

3. Mucinous carcinomas: these tumours commonly occur in postmenopausal women and are characterised

by high ER expression and HER2-negative expression. The morphological characteristic of PMCs is cells clustered and floating in mucin pools, with eosinophilic cytoplasm, uniformly shaped round cells, a low nuclear grade, exhibiting indolent biological behaviour, and no micropapillary structures. If a mucinous carcinoma has micropapillary structures ($\geq 5\%$), it should be diagnosed as MUMPC;

4. Invasive papillary carcinoma: in these tumour types, cancer cells are arranged in papillary structures with fibrovascular cores. Although papillary formation, mucin secretion, and microcalcifications can be seen, it lacks the typical histological features of micropapillary structures. Immunohistochemistry with EMA and CD34 can help distinguish invasive papillary carcinoma from MUMPC. Cancer cells float in abundant extracellular mucin lakes, tumour cells lack atypia, there are no mitotic figures, and they lack the characteristic features of IMPC, with a low axillary lymph node metastasis rate;

5. Breast mucinous cystadenocarcinoma: tumours are often rich in mucin, appearing gelatinous, cystic, and nodular. Microscopically, tumour cells comprise columnar cells that are rich in intracytoplasmic mucin, with nuclei at the base forming cysts of varying sizes. Some cysts contain leaf-like papillae lined by regular columnar mucinous epithelial cells, which can easily be confused with the characteristics of MUMPC. However, the presence of typical micropapillary structures and the 'inverted polarity' expression of tumour cells in EMA staining can help distinguish MUMPC from mucinous cystadenocarcinoma;

6. Metastatic papillary carcinoma of the breast: based on history, histological morphology, and immunohistochemical characteristics, breast invasive MPMC can be distinguished from metastatic ovarian serous papillary carcinoma and metastatic thyroid papillary carcinoma.

Regarding treatment and prognosis, reports suggest that simple local surgical resection does not improve the prognosis of mucinous carcinoma [18], possibly because of a poor prognosis of the special subtype of mucinous carcinoma (MPMC). Therefore, systemic therapy should be an important component of treatment for MPMC. In our group, 2 patients underwent a modified radical mastectomy followed by endocrine therapy + chemotherapy + targeted therapy; 1 patient underwent a modified radical mastectomy followed by chemotherapy + radiotherapy + targeted therapy; 2 patients underwent breast-conserving surgery + sentinel lymph node surgery, followed by endocrine therapy + radiotherapy. After 60 months of follow-up, no recurrence or metastasis was observed. One patient was not treated and died after 17 months of follow-up, and 1 case was newly diagnosed. However, the follow-up time in this group was relatively short; more cases with

long-term follow-up are needed to understand this special type of breast cancer.

Conclusions

Breast MPMC is a type of invasive mucinous carcinoma with aggressive features. It should not be loosely diagnosed as mucinous carcinoma, nor hastily diagnosed as IMPC. It may be a special type of breast cancer with transitional biological behaviour between ordinary mucinous carcinoma and IMPC. Therefore, it is important to correctly recognise and diagnose this type of mucinous carcinoma to enable appropriate clinical treatment.

Disclosures

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4. Conflicts of interest: None.

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