Kozonoe *et al.*
Surgical and Experimental Pathology (2024) 7:13
https://doi.org/10.1186/s42047-024-00156-2

Surgical and Experimental Pathology

RESEARCH Open Access

Mismatch repair deficiency in bilateral breast cancer

Milena Massumi Kozonoe[1,2], Jacqueline Justino Nabhen[3], Bruno Ribeiro Batista[1], Lucas Novello[2], Edenir Inêz Palmero[4], Sérgio Ossamu Ioshii[1,2] and Júlia Costa Linhares[2*]

Abstract

Background Since the FDA approved immune-enhancing therapies for patients with high microsatellite instability (MSI-H) and/or mismatch repair deficiency (dMMR), recognizing these biomarkers in solid tumors has gained clinical importance. Although MSI-H and dMMR are considered uncommon in breast cancer, previous studies on bilateral breast cancer (biBC) identified a surprisingly high frequency of MSI.

Methods In this study, we aimed to describe the prevalence of dMMR and its association with clinicopathologic parameters in biBC. We performed immunohistochemistry with anti-MMR proteins on tissue microarrays (TMAs) with 58 bilateral breast cancer cases. The biomarkers used were MLH1, PMS2, MSH2, MSH6, ER, PR, HER2 and Ki67. SPSS was used for data analysis.

Results Four (6.9%) cases showed dMMR on TMAs. Three (75%) of the dMMR cases were luminal and one (25%) was triple negative. Two biBC cases presented unilateral dMMR. No association between dMMR status and clinicopathologic parameters was found.

Conclusions This work highlights a noticeable frequency of dMMR in bilateral breast cancer and builds upon previous research in this area, suggesting routine MMR protein testing as part of the immunohistochemical panel for biBC to identify candidates for immune-enhancing therapies.

Keywords Breast cancer, Bilateral breast cancer, Microsatellite instability, Mismatch repair deficiency, Mutational load, Immunotherapy

Background

Microsatellite instability (MSI) is a hypermutation phenotype that results from impairment in the DNA mismatch repair (MMR) system. Both MMR deficiency (dMMR) and MSI are established as biomarkers for routinely testing in colorectal cancer (CRC) and other solid tumors (Alexandrov 2013; Allison et al. 2020). In clinical practice, testing is performed either by molecular diagnostic methods, such as polymerase chain reaction (PCR) of specific microsatellite sequences, or by immunohistochemical (IHC) detection of loss in MMR proteins (MLH1, MSH2, MSH6, PSM2) (Allison et al. 2020).

For interpretation and standardization purposes, the College of American Pathologists (CAP) has introduced a template for reporting the results of DNA mismatch repair testing. The template provides options for recording intact or lost nuclear expression of these proteins and allows for interpretation regarding the presence or absence of deficient mismatch repair. Furthermore, it outlines interpretations for microsatellite instability (MSI), distinguishing between MSI-Stable (MSS), MSI-Low (MSI-L), and MSI-High

*Correspondence:
Júlia Costa Linhares
juliaclinhares@yahoo.com.br
[1] Pontifical Catholic University of Paraná, Curitiba, PR, Brazil
[2] Department of Pathology, Erasto Gaertner Hospital, Curitiba, PR, Brazil
[3] Federal University of Paraná, Curitiba, PR, Brazil
[4] Instituto de Pesquisa Pelé Pequeno Príncipe, Curitiba, PR, Brazil

(MSI-H) categories, with implications for further testing for Lynch syndrome and genetic counseling (Barzaman et al. 2021; Bates 2018).

The relevance of MSI detection is based on the rationale that genomes of MSI-H) tumors present a high load of somatic mutations, some encoding potential immunogenic neoantigens. This molecular characteristic likely explains why such tumors are highly sensitive to immune-enhancing therapies irrespective of their anatomic origin (Boland 1998; Bonneville 2017). In 2017, the US Food and Drug Administration (FDA) approved the immune checkpoint inhibitor pembrolizumab for solid tumors showing MSI-H and/ or dMMR. Since then, recognition of the MSI-H phenotype in solid tumors other than colorectal cancer became of great interest in clinical practice and a growing body of data has described the MSI-H phenotype in several tumor types (Cheng et al. 2019; Meltem 2012). A recent meta-analysis estimates that around 14% of solid tumors present with MSI-H (Cohen et al. 2015), although this percentage largely varies across malignancies (Cheng et al. 2019; Meltem 2012; Cohen et al. 2015).

Breast cancer (BC) is not traditionally considered a tumor capable of eliciting a robust immune response within the organism (Contegiacomo 1995; Curigliano 2017). However, given its mixture of different molecular subtypes, recent years have seen investigations into potential new therapies as well as the immunogenicity of several subtypes (Curigliano 2017; Davies et al. 2017; Denkert 2014). Considerable attention has been directed towards molecular reclassification of BC subtypes based on immunity-related genes alongside conventional intrinsic subtypes (Curigliano 2017; Davies et al. 2017; Denkert 2014).

Our understanding of the intricate interplay between the immune system and BC is evolving. It is increasingly evident that the complexity of immunology necessitates a multifaceted approach to treatment, as no singular therapy is likely to be universally effective. The challenge for researchers lies in devising strategies and methods to harness an effective immune response against BC. Immunotherapeutic strategies in BC, although still in their early stages, show potential for treating specific BC subtypes (Curigliano 2017).

In agreement with that, the frequency of dMMR and MSI-H in BC was consistently reported as low, ranging from 1,7% to a maximum of 3,8% (Cheng et al. 2019; Meltem 2012; Fitzgibbons 2021; Fitzgibbons et al. 2023; Hause 2016). However, previous studies on bilateral BC (biBC) identified a surprisingly higher occurrence of the MSI-H phenotype in this subgroup compared to unilateral BC (Holm et al. 2014; Ibrahim et al. 2015). Since the MSI-H status has a predictive value, routine MSI testing in biBC might be useful in clinical practice (Imyanitov and Kuligina 2019).

When discussing biBC, scholars vary in their definitions. Some define biBC as the occurrence of tumors in both breasts either simultaneously or within a relatively short timeframe, typically around six months (Imyanitov et al. 2000; Kheirelseid et al. 2010; Kuligina 2006; Le 2015)to one year (Lee et al. 2020; Li 2020; Liang et al. 2013; Lorenzi et al. 2020). One can label them as synchronous if the tumors are detected within a shorter period (six months or one year), and metachronous if the interval extends beyond this period.

Given the scant of prior investigations into the role of IHC detection of DNA MMR in biBC, our study contributes to the emerging field by further elucidating the prevalence of dMMR in biBC. This research addresses a critical gap in existing knowledge, seeking to enhance our understanding of the molecular characteristics in biBC.

By specifically focusing on the role of IHC detection of dMMR, this study aims to evaluate the expression of proteins related to DNA mismatch repair in biBC, correlating them with various clinical parameters, including hormonal profile, HER2 status, Ki67 proliferative index, age, and histological types of cancer. The primary objective was to analyze the expression of key DNA repair proteins—MLH1, PMS2, MSH2, MSH6—utilizing IHC in women diagnosed with biBC at an oncology hospital. The use of IHC facilitates cost-effective and accessible detection of protein expression, aiding in the selection of eligible patients for subsequent molecular studies. Through comprehensive analysis of these biomarkers, this research seeks to enhance understanding of dMMR in biBC and its clinical implications.

Methods

We conducted a single-institution, observational, retrospective study at the Erasto Gaertner Hospital — a prominent cancer center situated in Curitiba, Paraná, Brazil. Recognized as Paraná's largest public oncologic hospital. Cases of biBC managed at the institution from 2005 to 2020 were retrieved from the pathology laboratory. We included samples of patients with a histopathological diagnosis of BC in both breasts and we excluded samples of patients diagnosed with carcinoma "in situ" and samples in which the tumor area was insufficient for analysis. For this study, synchronous biBC cases were defined as a time interval between both diagnoses less than one year (Lee et al. 2020; Li 2020; Liang et al. 2013; Lorenzi et al. 2020).

For each patient, formalin-fixed paraffin-embedded (FFPE) tissue samples from biopsies and/ or surgical specimens of both breasts were used for analysis, according to the disponible material for each patient. For each

donor FFPE, two to four samples from the tumor core were collected to construct tissue microarrays (TMAs) (Fig. 1). A skin punch instrument was used to remove tissue cylinders from each tumor from the formalin-fixed paraffin-embedded tissue samples.

Representative 4-um-thick array sections were cut from the TMA blocks to perform immunohistochemical analysis of the hormonal profile, the HER2 and Ki67 statuses, and the MMR status on the specimens. The antibodies used were: MLH1 (clone M1), PMS2 (clone A16-4), MSH2 (clone G219-1129), MSH6 (clone SP93), ER (clone SP1), PR (clone 1E2), HER2 (clone 4B5) and Ki67 (clone 30–9). More details about antibodies used, concentrations, antigen retrieval, as well as summarized information about cutoffs and interpretation are shown in Table 1.

Fig. 1 Two to four samples from the tumor core were collected from each FFPE to the TMAs. Source: The authors

Diagnosis of dMMR by IHC requires antibodies against the 4 repair proteins: MLH1, PSM2, MSH2 and MSH6. As they act as heterodimers, analysis has to be done in pairs (MLH1 with PMS2 and MSH2 with MSH6). If no repair protein is lost, the tumor is classified as MMR-proficient (pMMR) (Fig. 2); if a pair of protein is lost or one of protein (PMS2 or MSH6) is lost, the tumor is classified as dMMR.

We adhered to the guidelines provided by the CAP for reporting DNA MMR testing results. Specimens were processed and analyzed according to standard protocols, with IHC performed to assess the expression of MMR proteins MLH1, MSH2, MSH6, and PMS2. Interpretation of IHC staining was conducted to determine the presence or absence of nuclear expression for each protein. MSI status was determined based on the CAP-approved criteria (Barzaman et al. 2021).

HR and HER2 testing were conducted following the guidelines outlined by the American Society of Clinical Oncology (ASCO) and the CAP. IHC was primarily employed to determine HR status, with positive staining defined as nuclear staining. Reporting guidelines recommended by ASCO and CAP were followed, classifying cases with at least 1% positive cells as hormone receptor-positive, with specific criteria outlined for reporting (Luchini et al. 2019; Marabelle et al. 2020).

HER2 expression was evaluated through IHC, with scoring systems ranging from 0 to 3+, with 0 and meaning a negative result, 1+meaning a negative result with low HER2 membrane expression, 2+meaning an equivocal result that needs a complementary in situ hybridization technique and 3+meaning a HER2 overexpression.

Table 1 IHC details

Antibody	Clone	Source	Concentration	Antigen retrieval	Interpretation/cut-offs
Anti-MLH1	M1 Roche	Rabbit Monoclonal	1 µg/mL	Cell conditioning 1 (CC1) 92 min, 100 °C	Nuclear expression (Presence/absence)
Anti-PMS2	A16-4 Roche				
Anti-MSH2	G219-1129 Roche			CC1 92 min	
Anti-MSH6	SP93 Roche				
Anti-ER	SP1 Roche			CC1	Nuclear expression (> 1% nuclear staining)
Anti-PR	Clone 1E2 Roche				
Anti-HER2	Clone 4B5 Roche		6 µg/mL	ULTRA CC1 36 min, 95 °C	Membrane expression (0; 1+; 2+; -3+)
Anti-Ki67	Clone 30–9 Roche		2 µg/mL	CC1, 64 min	Nuclear expression (0–20%; > 20%)

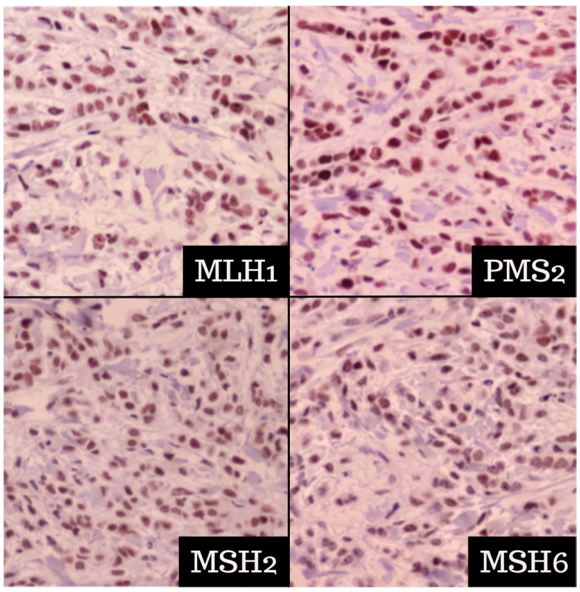

Fig. 2 Figure demonstrating intact nuclear expression on a lobular breast carcinoma. (immunohistochemistry, light microscopy, magnification 400x)

Ki-67 proliferation index was assessed by the percentage of positively stained nuclei, with a cutoff value of 20% (Luchini et al. 2019; Nádorvári 2024).

The St. Gallen International Expert Consensus Conference on the Primary Therapy of Early Breast Cancer 2017 recommendations offer valuable information into determining molecular subtypes using surrogate markers. These classifications, rooted in routine histopathology, are recognized by the panel as clinically valuable (Nádorvári 2024).

For instance, the triple-negative group, characterized by negative expressions of ER, PR, and HER2, represents a distinct subtype. Within the spectrum of luminal tumors, Luminal A-like tumors typically exhibit low-grade features, strong ER/PR positivity, negativity for HER2, and a low proliferative *index*. Conversely, Luminal-B-like tumors, while ER-positive, may demonstrate varying degrees of ER/PR expression, higher grades, and elevated proliferative *indices* (Nádorvári 2024).

All analyses were conducted in accordance with institutional protocols and ethical guidelines and all IHC slides were analyzed separately by two independent pathologists (MK and SI) and submitted to discussion when results were discordant. Data from electronic medical records (EMR) from the dMMR patients was analyzed for first-degree family history of BC and personal history of primary tumors other than BC.

This study was approved by the institutional ethical committee registered with the Certificate of Ethical Appreciation and Authorization (CAAE) number 09593219.6.0000.0098. All information collected from the participants was treated with strict confidentiality and is the responsibility of the researchers.

Table 2 Pathological features of the tumors included in the study

	Parameter	58 (n)	100%
Histological subtype	Carcinoma of no special type (bilateral)	28	48.3%
	Lobular (bilateral)	6	10.3%
	Discordant histology	24	41.4%
Hormone receptor status	Positive (bilateral)	45	77.6%
	Positive (unilateral)	6	10.3%
	Negative (bilateral)	7	12.1%
HER2	Positive (bilateral)	5	8.6%
	Positive (unilateral)	6	10.3%
	Equivocal (bilateral)	1	1.7%
	Equivocal (unilateral)	3	5.2%
	Negative (bilateral)	43	74.1%
Ki67	> 20% (bilateral)	24	41.4%
	> 20% (unilateral)	14	24.1%
	< 20% (bilateral)	20	34.5%

Statistical analyses

The SPSS® 16.0 software package (SPSS Inc., Chicago, IL, USA) was used for statistical analysis. Correlation of age at diagnosis of the first tumor and the dMMR status was performed using the chi-squared test. Assessment of associations between tumor histopathological characteristics and the dMMR status considering all tumors was performed using fisher's exact test.

Results

Fifty-eight biBC cases were included, of which twenty-nine (50%) were synchronous. The description of the pathological features of the study population is found in Table 2. In terms of histological subtype, carcinoma of no special type was predominant in bilateral cases, accounting for 48.3%, while lobular subtype constituted 10.3%. Discordant histology was observed in 41.4% of cases.

The analysis of HR status revealed that 77.6% of bilateral cases were positive, compared to only 10.3% in unilateral cases, with 12.1% showing negative status bilaterally. Regarding HER2 status, 8.6% of bilateral cases were positive (3 + score), along with 10.3% in unilateral cases. Equivocal results were found in 1.7% and 5.2% of bilateral and unilateral cases, respectively, while the majority, 74.1%, exhibited negative HER2 status in bilateral cases. Ki67 expression varied, with 41.4% of bilateral cases showing Ki67 levels greater than 20%,

compared to 24.1% in unilateral cases. Conversely, 34.5% of bilateral cases and 34.5% of unilateral cases exhibited Ki67 levels less than 20%.

Out of the 58 biBC patients, four (6.9%) presented with an MMR-deficient tumor in at least one breast with three cases being synchronous tumors. Three (75%) of the dMMR cases were luminal and only one (25%) was triple negative. Three cases had paired MLH1 and PMS2 losses and one case had PMS2 loss only. In two biBC cases the dMMR was present in both breasts, and in both of them concordance was seen as of which proteins were lost. Detailed analysis of each dMMR case is shown in Table 3.

Data from EMR revealed that none of the patients had a first-degree relative with cancer nor a personal history suggesting hereditary syndromes. The chi-squared test found no association between MMR status and age at diagnosis of the first tumor. Fisher's exact test found no association between tumor histopathological characteristics and MMR status.

At diagnosis, metastatic disease was evident in three patients, each exhibiting distinct characteristics. The first, aged 48, presented with synchronous lobular biBC and exhibited metastases in both the bones and pleura. Another patient, aged 52, diagnosed with synchronous lobular carcinoma and characterized as triple-negative by immunophenotype, demonstrated metastasis primarily in the central nervous system, accompanied by pleural and pericardial effusion resulting from tumor infiltration.

Additionally, a patient with metachronous disease exhibited axillary lymph node involvement. Subsequently, the first patient passed away three years after diagnosis, while the other two were lost to follow-up in the hospital's oncology department. Furthermore, a 62-year-old patient with synchronous disease and unilateral dMMR (Fig. 3) had negative sentinel lymph nodes and had survived disease-free for eight years, now presenting with signs suggesting bone metastasis.

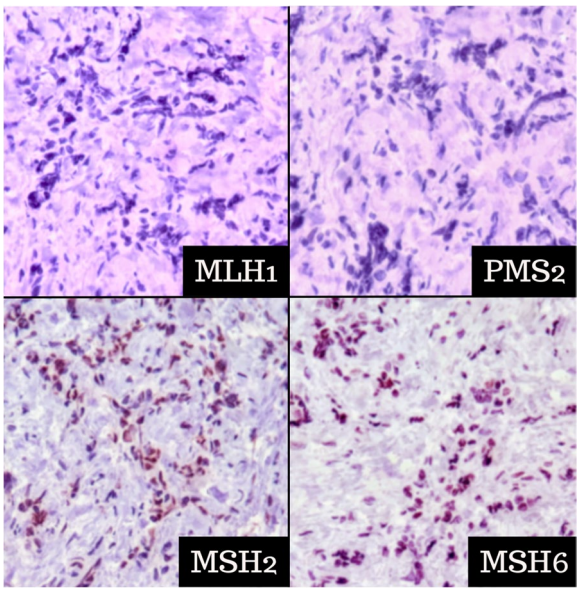

Fig. 3 Figure illustrating repair proteins with loss of expression of MLH1 and PMS2, and without loss of expression of MSH2 and MSH6 (immunohistochemistry, light microscopy, magnification 400x). Source: The authors

Discussion

BiBC is a rare disease (Orr 2022). Although some biBC cases are associated with hereditary cancer syndromes, most of them seem to be sporadic, and their natural history is poorly understood. Unlike colorectal and endometrial cancers, MSI-H and dMMR are very uncommon in BC (Cheng et al. 2019; Meltem 2012; , Fitzgibbons 2021; Fitzgibbons et al. 2023; Hause 2016). However, a previous work from Kuligina et al (Ibrahim et al. 2015) identified through molecular testing that the MSI-H phenotype was present in a subset of biBC, but not in

Table 3 Description of each dMMR case

Age at diagnosis (yr)	Chronicity	MMR proteins lost		Histology		Molecular subtype according to the immunophenotype
		Right breast tumor	Left breast tumor	Right breast tumor	Left breast tumor	
48	Synchronous	PMS2	PMS2	Lobular	Lobular	Luminal B
45	Metachronous	MLH1 PMS2	MLH1 PMS2	NOS	NOS	Luminal B
52	Synchronous	None	MLH1 PMS2	Lobular	Lobular	Triple negative
62	Synchronous	None	MLH1 PMS2	NOS	Other	Luminal A

unilateral BC. This finding led experts to suggest testing for MSI in biBC, since MSI-H tumors can be treated with immune-enhancing therapies (Boland 1998; Bonneville 2017; Cheng et al. 2019). However, no previous study accessed the MMR status in bilateral BC. This is important because dMMR can be detected using IHC, which is a routinely used diagnostic method in BC.

Guidelines outlining the assessment of instability biomarkers in colorectal cancer and other solid tumors emphasize the importance of utilizing various methods, including molecular and IHC approaches. Notably, dMMR and its associated MSI-H characterize a specific subset of cancers, potentially responsive to immune checkpoint inhibitor immunotherapy. This subset, defined by genetic mutations in DNA mismatch repair genes, exhibits elevated tumor mutational burden (TMB), rendering it sensitive to immune checkpoint blockade (Alexandrov 2013; Allison et al. 2020; Barzaman et al. 2021).

Mutations in genes such as *MLH1*, *MSH2*, *MSH6*, and *PMS2*, along with promoter methylation-induced inactivation, contribute to dMMR and MSI. The identification of MSI-H/dMMR tumors presents opportunities for personalized treatment strategies, particularly with the emergence of immune checkpoint inhibitors. Typical patterns of MMRd include concurrent loss of *MLH1*/*PMS2* or *MSH2*/*MSH6*, indicating abnormalities in *MLH1* or *MSH2*, respectively, and isolated loss of *PMS2* or *MSH6*, suggesting abnormalities in PMS2 or *MSH6* (Barzaman et al. 2021; Özer et al. 2002; Peshkin 2010).

While typical patterns of MMRd often involve complete loss of staining for specific MMR proteins, such as MLH1, PMS2, MSH2, and MSH6, in the tumor, atypical patterns, such as primary clonal loss, can complicate interpretation (Barzaman et al. 2021; Ibrahim et al. 2015; Özer et al. 2002). Recent studies have highlighted primary clonal loss as a unique phenomenon associated with specific genetic abnormalities and MSI-H. Primary clonal loss refers to distinct loss of MMR proteins. This phenomenon, detectable through MMR IHC, involves discernible regional loss within the tumoral cells with reliable internal control, indicating true clonal loss. These findings suggest that primary clonal loss may signal underlying MMR gene abnormalities, emphasizing the importance of further genetic evaluation, particularly for Lynch syndrome detection.

However, it's important to distinguish true clonal loss from staining variations resulting from technical issues. These observations highlight the complexity of MMR evaluation and suggest the need for comprehensive guidelines to interpret IHC results accurately. Integrating these insights into clinical practice can enhance the identification of MSI-H/dMMR tumors and improve patient outcomes through tailored therapeutic interventions, including immunotherapies (Barzaman et al. 2021; Özer et al. 2002; Peshkin 2010; Schwentner et al. 2012).

Our study found dMMR in four biBC cases (6.19%) (Table 1), a higher frequency compared to unilateral BC (Cheng et al. 2019; Meltem 2012; Fitzgibbons 2021; Fitzgibbons et al. 2023; Hause 2016). In three cases, loss of expression occurred in pairs, meeting the criteria to the immunohistochemical diagnosis of dMMR (Sepulveda et al. 2017). In one case, PMS2-isolated deficiency was found. Unlike reports on unilateral BC (Vuoto 2010), we did not find a correlation between the histological subtype and dMMR. This suggests that there are, up until now, no patterns that can predict dMMR in biBC.

An analysis of our results suggests that the occurrence of dMMR in biBC may be sporadic rather than driven by hereditary syndromes. This inference is supported by several factors. Firstly, a significant majority (75%) of the tumors in our study exhibiting deficiency in the MMR system were of the luminal subtype (Table 3). It is well-established that the most common hereditary genetic defect observed in biBC patients is attributed to mutations in the *BRCA1*/*BRCA2*genes (Wadasadawala et al. 2018; Wolff 2023).

Other factors that can contribute to this assumption include the information that none of the dMMR patients had primary tumors apart from BC nor had a familiar and/ or personal history of cancer associated with hereditary syndromes and half of the dMMR patients presented with this phenotype in only one of their breasts (Table 3). This finding is similar to what was described by Kuligina et al (Ibrahim et al. 2015) that also described unilateral dMMR tumors in biBC patients. However, we acknowledge that the frequency of luminal cancers in our cohort, along with other findings, may not be sufficient to definitively support this assumption.

Interestingly, in both of our patients who presented with unilateral dMMR tumors the time between diagnosis of both tumors classified them as synchronous. Kuligna et al (Ibrahim et al. 2015) found the opposite, since in their study MSI-H preferentially occurred in metachronous tumors. This led the authors to dismiss the role of hereditary factors and to propose that the adjuvant treatment used against the first malignancy was responsible for their findings. Our study does not support this hypothesis because the three dMMR tumors from our cohort were synchronous and it suggests that there might be another acquired factor that led to the dMMR in one of the breasts. Intertumoral pathological heterogeneity (Table1), together with discordant dMMR results between both tumors in two of our cases, reinforces the importance of tumor board discussions in biBC cases to provide the best treatment for each case.

Our study has several limitations. First, it was based on retrospective material. Second, the number of patients included was relatively small, which might explain no association being found between clinicopathologic characteristics and MMR status. Third, it is possible that dMMR prevalence was underestimated in our study, since the use of TMAs may not reflect intratumoral heterogeneity, despite our effort to collect two to four samples of each FFPE block. Thus, dMMR frequency in biBC might be even higher than the one we reported. Studies with larger sample sizes using a whole section from the FFPE blocks are needed to overcome those limitations. Our study encountered constraints regarding the availability and completeness of patient EMR, which hindered the comprehensive analysis of staging, follow-up data, grade, and axillary lymph node status. Consequently, we were unable to conduct a robust statistical evaluation to explore potential associations between mismatch repair (MMR) status and prognostic factors. This limitation underscores the need for more extensive and standardized data collection methods in future studies to facilitate a more thorough understanding of the relationship between MMR status and clinical outcomes in bilateral breast cancer.

Conclusions

While our study reaffirms the observed frequency of dMMR in bilateral BC, it contributes to existing evidence noticed in previous research in the prevalence of dMMR in bilateral BC. Given that dMMR tumors can be treated with immune-enhancing therapies, we suggest routine testing for MMR proteins as a part of the immunohistochemical panel used for biBC.

Abbreviations

ACSO	American Society of Clinical Oncology
BC	Breast cancer
CAP	College of American Pathologists
CRC	Colorectal cancer
dMMR	Mismatch repair deficiency
EMR	Electronic medical records
FDA	Food and Drug Administration
FFPE	Formalin-fixed paraffin-embedded
HER2	Human epidermal growth factor receptor 2
HR	Hormone receptor
IHC	Immunohistochemistry
MSI-H	High microsatellite instability
MSI-L	MSI-Low
MSS	MSI-Stable
PCR	Polymerase chain reaction
SPSS	Statistical Package for the Social Sciences
TMB	Tumor mutational burden

Acknowledgements

Not applicable.

Authors' contribution

MMK and SOI conceptualized and designed the work. MMK and LN performed the histological and immunohistochemical examination, JJN and BRB acquired clinical data for the cases. MMK was a major contributor in writing the manuscript. JCL substantively revised the work and all authors read and approved the final manuscript. All authors have agreed both to be personally accountable for the author's own contributions and to ensure that questions related to the accuracy or integrity of any part of the work, even ones in which the author was not personally involved, are appropriately investigated, resolved, and the resolution documented in the literature.

Funding

There were no fundings involved in this study.

Availability of data and materials

Not applicable.

Declarations

Ethics approval and consent to participate

This study was submitted to and approved by the Research Projects and Studies Ethics Committee (CEP) at the Research and Study Projects Center (CEPEP) of Hospital Erasto Gaertner (HEG), registered with the Certificate of Ethical Appreciation and Authorization (CAAE) number 09593219.6.0000.0098. All information collected from the participants was treated with strict confidentiality and is the responsibility of the researchers.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Received: 27 December 2023 Accepted: 11 June 2024
Published online: 10 August 2024

References

Alexandrov LB, Nik-Zainal S, Wedge DC, Aparicio SAJR, Behjati S, Biankin AV, et al. Signatures of mutational processes in human cancer. Nature. 2013;500(7463):415–21. Available from: https://www.ncbi.nlm.nih.gov/pubmed/23945592.

Allison KH, Hammond MEH, Dowsett M, McKernin SE, Carey LA, Fitzgibbons PL, et al. Estrogen and progesterone receptor testing in breast cancer: ASCO/CAP guideline update. J Clin Oncol. 2020;38(12):1346–66.

Barzaman K, Moradi-Kalbolandi S, Hosseinzadeh A, Kazemi MH, Khorramdelazad H, Safari E, et al. Breast Cancer immunotherapy: Current and Novel Approaches. Int Immunopharmacol. 2021;98:107886.

Bates JP, Derakhshandeh R, Jones L, Webb TJ. Mechanisms of immune evasion in breast cancer. BMC Cancer. 2018;18(1):1–14.

Boland CR, Thibodeau SN, Hamilton SR, Sidransky D, Eshleman JR, Burt RW, et al. A national cancer institute workshop on microsatellite instability for cancer detection and familial predisposition: development of international criteria for the determination of microsatellite instability in colorectal cancer. Cancer Res. 1998;58(22):5248–57. Available from: https://pubmed.ncbi.nlm.nih.gov/9823339/. Cited 2020 Nov 15.

Bonneville R, Krook MA, Kautto EA, Miya J, Wing MR, Chen HZ, et al. Landscape of microsatellite instability across 39 cancer types. JCO Precision Oncol. 2017;2017(1). Available from:https://pubmed.ncbi.nlm.nih.gov/29850653/.

Cheng AS, Leung SCY, Gao D, Burugu S, Anurag M, Ellis MJ, et al. Mismatch repair protein loss in breast cancer: clinicopathological associations in a large British Columbia cohort. Breast Cancer Res Treat. 2019;179(1):3–10.

Baykara M, Ozturk SC, Buyukberber S, Helvaci K, Ozdemir N, Alkis N, et al. Clinicopathological features in bilateral breast cancer. Asian Pac J Cancer Prev. 2012;13(9):4571–5.

Cohen SA, Laurino M, Bowen DJ, Upton MP, Pritchard C, Hisama F, et al. Initiation of universal tumor screening for Lynch syndrome in colorectal cancer patients as a model for the implementation of genetic information into clinical oncology practice. Cancer. 2015;122(3):393–401.

Contegiacomo A, Palmirotta R, De Marchis L, Pizzi C, Mastranzo P, Delrio P, et al. Microsatellite instability and pathological aspects of breast cancer. Int J Cancer. 1995;64(4):264–8.
Curigliano G, Burstein HJ, Winer EP, Gnant M, Dubsky P, Loibl S, et al. De-escalating and escalating treatments for early-stage breast cancer: the St. Gallen International Expert Consensus Conference on the Primary Therapy of Early Breast Cancer 2017. Ann Oncol. 2017;28(8):1700–12. Available from: https://www.annalsofoncology.org/action/showPdf?pii=S0923-7534%2819%2932181-7.
Davies H, Morganella S, Purdie CA, Jang SJ, Borgen E, Russnes H, et al. Whole-Genome Sequencing reveals breast cancers with mismatch repair deficiency. Can Res. 2017;77(18):4755–62.
Denkert C. The Immunogenicity of Breast Cancer—molecular Subtypes Matter. Ann Oncol. 2014;25(8):1453–5.
Fitzgibbons PL, Bartley AN, Broaddus RR, Shi CS. Template for reporting results of dna mismatch repair testing with guidance from the CAP cancer and CAP pathology electronic reporting committees. 2021. Available from: https://documents.cap.org/protocols/DNA.MMR.Bmk_1.0.0.2.REL_CAPCP.pdf?_gl=1. Cited 2024 Mar 10.
Fitzgibbons PL, Dillon DA, Alsabeh R, Berman MA, Hayes DF, Hicks DG, et al. Template for reporting results of biomarker testing of specimens from patients with carcinoma of the breast. Arch Pathol Lab Med. 2023;138(5):595–601.
Hause RJ, Pritchard CC, Shendure J, Salipante SJ. Classification and characterization of microsatellite instability across 18 cancer types. Nat Med. 2016;22(11):1342–50. Available from: https://pubmed.ncbi.nlm.nih.gov/27694933/.
Holm M, Tjønneland A, Balslev E, Kroman N. Prognosis of synchronous bilateral breast cancer: a review and meta-analysis of observational studies. Breast Cancer Res Treat. 2014;146(3):461–75.
Ibrahim NY, Sroor MY, Darwish DO. Impact of bilateral breast cancer on prognosis: synchronous versus metachronous tumors. Asian Pac J Cancer Prev. 2015;16(3):1007–10.
Imyanitov EN, Kuligina ESh. Systemic investigations into the molecular features of bilateral breast cancer for diagnostic purposes. Expert Rev Mol Diagn. 2019;20(1):41–7.
Imyanitov EN, Togo AV, Suspitsin EN, Grigoriev MY, Pozharisski KM, Turkevich EA, et al. Evidence for microsatellite instability in bilateral breast carcinomas. Cancer Lett. 2000;154(1):9–17.
Kheirelseid EAH, Jumustafa H, Miller N, Curran C, Sweeney K, Malone C, et al. Bilateral breast cancer: analysis of incidence, outcome, survival and disease characteristics. Breast Cancer Res Treat. 2010;126(1):131–40.
Kuligina ESh, Grigoriev MYu, Suspitsin EN, Buslov KG, Zaitseva OA, Yatsuk OS, et al. Microsatellite instability analysis of bilateral breast tumors suggests treatment-related origin of some contralateral malignancies. J Cancer Res Clin Oncol. 2006;133(1):57–64.
Le DT, Uram JN, Wang H, Bartlett BR, Kemberling H, Eyring AD, et al. PD-1 blockade in tumors with mismatch-repair deficiency. N Engl J Med. 2015;372(26):2509–20. Available from: https://www.nejm.org/doi/full/10.1056/NEJMoa1500596.
Lee A, Moon BI, Kim TH. BRCA1/BRCA2 pathogenic variant breast cancer: treatment and prevention strategies. Ann Lab Med. 2020;40(2):114.
Li K, Luo H, Huang L, Luo H, Zhu X. Microsatellite instability: a review of what the oncologist should know. Cancer Cell Int. 2020;20(1):1–13.
Liang X, Li D, Geng W, Cao X, Xiao C. The prognosis of synchronous and metachronous bilateral breast cancer in Chinese patients. Tumor Biology. 2013;34(2):995–1004.
Lorenzi M, Amonkar M, Zhang J, Mehta S, Liaw KL. Epidemiology of Microsatellite Instability High (MSI-H) and Deficient Mismatch Repair (dMMR) in solid tumors: a structured literature review. J Oncol. 2020;9(2020):1–17.
Luchini C, Bibeau F, Ligtenberg MJL, Singh N, Nottegar A, Bosse T, et al. ESMO recommendations on microsatellite instability testing for immunotherapy in cancer, and its relationship with PD-1/PD-L1 expression and tumour mutational burden: a systematic review-based approach. Ann Oncol. 2019;30(8):1232–43.
Marabelle A, Le DT, Ascierto PA, Di Giacomo AM, De Jesus-Acosta A, Delord JP, et al. Efficacy of pembrolizumab in patients with noncolorectal high microsatellite instability/mismatch repair-deficient cancer: results from the phase II KEYNOTE-158 Study. J Clin Oncol. 2020;38(1):1–10.
Nádorvári ML, István Kenessey, Kiss A, Tamás Barbai, Kulka J, Erzsébet Rásó, et al. Comparison of standard mismatch repair deficiency and microsatellite instability tests in a large cancer series. J Transl Med. 2024;22(1):1–9.
Orr C, Wang C, Canan Firat, Connell LC, Sheehan MR, Efsevia Vakiani, et al. Primary clonal loss of mismatch repair protein on immunohistochemistry: a pattern of abnormality that warrants genetic workup. JCO Precision OncoL. 2022;(6):e2200111.
Özer E, Yuksel E, Kizildag S, Sercan O, Özen E, Canda T, et al. Microsatellite Instability in Early-onset Breast Cancer. Pathol Res Prac. 2002;198(8):525–30.
Peshkin BN, Alabek ML, Isaacs C. BRCA1/2 Mutations and Triple Negative Breast Cancers. Breast Dis. 2010;32(0). Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3870050/.
Schwentner L, Wolters R, Wischnewsky M, Kreienberg R, Wöckel A. Survival of patients with bilateral versus unilateral breast cancer and impact of guideline adherent adjuvant treatment: a multi-centre cohort study of 5292 patients. Breast. 2012;21(2):171–7.
Sepulveda AR, Hamilton SR, Allegra CJ, Grody WW, Cushman-Vokoun AM, Funkhouser WK, et al. Molecular biomarkers for the evaluation of colorectal cancer. J Mol Diagn. 2017;19(2):187–225.
Vuoto HD, García A, Candá G, Zimmermann AG, Uriburu JL, Isetta J, et al. Bilateral breast carcinoma: clinical characteristics and its impact on survival. Breast J. 2010;16(6):625–32.
Wadasadawala T, Lewis S, Parmar V, Budrukkar A, Gupta S, Nair N, et al. Bilateral breast cancer after multimodality treatment: a report of clinical outcomes in an asian population. Clin Breast Cancer. 2018;18(4):e727–37.
Wolff AC, Somerfield MR, Dowsett M, Hammond EH, Hayes DF, McShane LM, et al. Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer: ASCO–College of American Pathologists Guideline Update. J Clin Oncol. 2023;22(41):3867–72.

Publisher's Note