ORIGINAL RESEARCH PAPER

Oncology

KEY WORDS: Primary Breast Lymphoma, DLBCL, ALCL.

PRIMARY BREAST LYMPHOMA: AN IMMUNOMORPHOLOGIC STUDY OF A RARE ENTITY

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ABSTRACT

Introduction& Objective: Breast lymphomas can be either arising primarily from the breast or as secondary involvement of the breast by primary nodal lymphomas. Primary breast lymphomas are extremely rare and account for 0.04 to 0.5% of all the breast malignancies and 1 to 2% of the extra nodal lymphomas. We studied the clinicopathologic features and subtypes of this rare disease at a regional cancer centre in South India.

Methods: All cases of primary breast lymphomas diagnosed over a period of 11.5yrs, were included in the study. The demographic, morphologic and immunophenotypic features were analysed.

Results: Thirteen cases of primary breast lymphomas were diagnosed and all were females. The mean age at presentation was 42.5yrs, with an age range of 19 to 65yrs. All the patients presented with unilateral breast lesion except one case with bilateral involvement. All 13 cases were Non-Hodgkin lymphomas(NHL), of which eleven cases were B-cell type and two cases were T-cell type (both anaplastic Large cell lymphomas- ALCL) and two cases were immunocompromised. The most common subtype of B-cell lymphoma was Diffuse Large B Cell Lymphoma (DLBCL).

Discussion & Conclusion: DLBCL was the commonest primary breast lymphoma, most of them of the non germinal centre(NGCB) subtype. The two cases of ALCLs in our study were non- implant associated and Anaplastic Lymphoma Kinase(ALK-1) positive. There was one case of plasmablastic lymphoma in a HIV infected individual. High index of suspicion and immunophenotyping play a vital role in the diagnosis of primary breast lymphomas and helps to make appropriate treatment decisions which in turn has prognostic implications.

INTRODUCTION

Lymphomas involving the breast are very rare. They can occur primarily in the breast tissue or as a secondary involvement of the breast as a part of systemic lymphoma. Primary lymphomas of the breast are defined as

1) those confined to one or both the breasts with or without ipsilateral axillary lymph node involvement.
2) with no evidence of disease elsewhere at presentation in a patient without a prior history of lymphoma.[1,2]

Primary breast lymphomas account for 0.04 to 0.5% of all the breast malignancies and 1 to 2% of all the extra nodal lymphomas.[2] It is believed that primary breast lymphomas arise from either the lymphocytes residing in the intra mammary lymph nodes or from the endogenous lymphoid tissue.[3]

This study was undertaken to evaluate the clinicopathological and immunophenotypic features of primary breast lymphomas at our institute which is a tertiary care cancer centre in South India. To the best of knowledge of the authors, this is one of the largest single institutional study of this kind.

METHODS

This is a retrospective observational study of all the cases of primary lymphomas of the breast diagnosed at the department of pathology, over a period of eleven and half years. Lymphomas secondarily involving the breast were excluded from the study. The relevant clinical & demographic details and HIV status were obtained from the case records. Tumor samples received in 10% neutral buffered formalin were processed according to the laboratory protocol and embedded in paraffin. Five micron thick sections were cut and stained with Hematoxylin and Eosin(H&E). H&E stained sections of all the cases were reviewed and histomorphologic typing into different lymphoma subtypes was done. A wide panel of Immunohistochemistry markers were performed based on morphology for exact subtyping by avidin, streptavidin, biotin method and HRP polymer method using Diaminobenzidine (DAB) as a chromogen. Details of antibodies are given in [Table 1].

TABLE 1: Details of the antibodies used in the diagnosis of primary breast lymphoma.

Antibody	Clone	Source	Antigen retrieval	Dilution
LCA	LCA8(Mouse monoclonal)	BioGenex	EDTA buffer, pH-9.0	1:400
Ki67	Mib1 (Mouse monoclonal)	BioGenex	EDTA buffer, pH-9.0	1:100
CD3	PS1 (Mouse monoclonal)	BioGenex	EDTA buffer, pH-9.0	1:100
CD20	L26 (Mouse monoclonal)	BioGenex	EDTA buffer, pH-9.0	1:70
CD10	56C6 (Mouse monoclonal)	BioGenex	EDTA buffer, pH-9.0	1:40
BCL-6	LN22 (Mouse monoclonal)	Biocare	EDTA buffer, pH-9.0	1:100
MUM-1	EAU32(Mouse monoclonal)	Biocare	EDTA buffer, pH-9.0	1:100
BCL-2	3.1 (Mouse monoclonal)	Biocare	EDTA buffer, pH-9.0	1:100
Cyclin D1	EPR224(Rabbit monoclonal)	BioGenex	EDTA buffer, pH-9.0	1:60
CD5	4C7 (Mouse monoclonal)	BioGenex	EDTA buffer, pH-9.0	1:60
ALK-1	SP8 (Rabbit monoclonal)	BioGenex	EDTA buffer, pH-9.0	Ready to use
CD 30	HRS4 (Mouse monoclonal)	BioGenex	EDTA buffer, pH-9.0	1:40
CD138	MI15 (Rabbit polyclonal)	BioGenex	EDTA buffer, pH-9.0	1:60
EMA	E29 (Mouse monoclonal)	BioGenex	EDTA buffer, pH-9.0	1:100
Pan CK	C11 (Mouse monoclonal)	Biocare	EDTA buffer, pH-9.0	1:20

RESULTS

During the study period of eleven and half years (January 2006-June 2017), thirteen cases of primary breast lymphomas were diagnosed. They constituted 0.17% (13/8079) of all the primary

breast malignancies, 1.28% (13/1017) of all the extra nodal lymphomas and 0.53% (13/2456) of all the Non- Hodgkin Lymphoma (NHLs) at our institute. The patient's age at presentation ranged from 19 to 65 years, with a mean age of 42.5 years. All the cases studied were female patients and all of them presented with unilateral breast lump except one case with bilateral involvement. Peripheral blood and bone marrow were not involved in any of the cases at presentation.

All the thirteen cases were NHLs, of which eleven cases (85%) were of B- cell type and two cases (15%) were of T- cell type. Among the B- NHLs, ten cases (91%) were Diffuse Large B- Cell Lymphomas (DLBCL) (Fig. 1A) and one case was Plasmablastic Lymphoma (PBL) (Fig. 2A). Both the cases of T- NHL were Anaplastic Large Cell Lymphomas (ALCL) (Fig. 3A). Two of the thirteen cases were HIV- positive, of which one case was a DLBCL and the other one was a PBL. Histomorphologically, the DLBCLs were categorised as centroblastic (40%), immunoblastic (10%), anaplastic (10%) and indeterminate (10%) types. Three cases (30%) of DLBCL could not be further categorised because of poor morphology. Immunohistochemically, the tumor cells were negative for cytokeratin and positive for LCA. Among the ten cases of DLBCL, five cases (50%) were Non- Germinal Centre B cell type (NGCB) being positive for CD20, BCL6& MUM1(Fig. 1B) and negative for CD10 and two cases (20%) were Germinal Centre B-cell type (GCB) being positive for CD20, CD10& BCL6 and negative for MUM1, according to Han's algorithm. Three cases of DLBCL could not be further categorised based on Han's algorithm, due to poor preservation of the tissue.The case of PBL was positive for MUM1& CD138 (Fig. 2B) and negative for CD20. Both the cases of ALCL were positive for CD3, ALK-1 (Fig. 3B), EMA & Cd30.

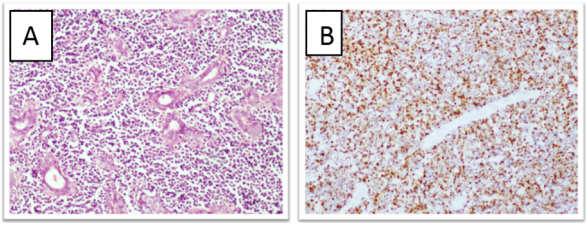

FIG.1: A: Diffuse Large B Cell Lymphoma (DLBCL), with neoplastic lymphoid cells around ducts and lobules. (H&E stain; original magnification x40); **B:** DLBCL NGCB type showing nuclear MUM1 expression. (IHC; original magnification x40).

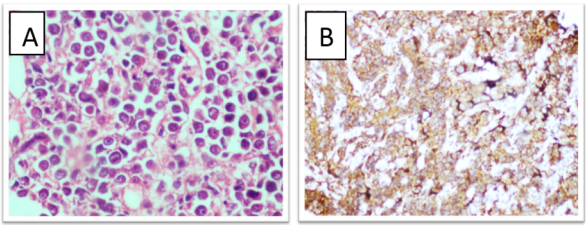

FIG.2: A: Plasmablastic Lymphoma (PBL). (H&E stain; original magnification x400); **B:** Shows CD138 expression by neoplastic cells (IHC; original magnification x100).

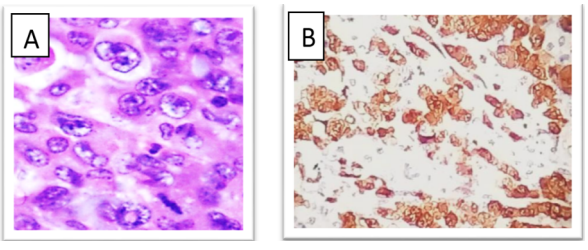

FIG.3: A: Shows hallmark cells in ALCL (H&E stain; original magnification x400); **B:** Shows nuclear and cytoplasmic ALK expression (IHC; original magnification x100).

The clinical details of the patients are given in [Table 2].

TABLE 2: Clinical and therapy details of the individual patients.

Sl. No	Age	Presentation	Laterality	Diagnosis	Therapy	Follow-up
1.	52y	Breast lump	Left	DLBCL	Surgery+ Chemo	Lost to follow-up.
2.	62y	Breast lump	Right	ALCL (ALK+)	Surgery+ Chemotherapy	Lost to follow-up.
3.	43y	Breast lump	Left	DLBCL	Surgery+ Chemotherapy	Lost to follow-up.
4.	36y	Breast lump	Left	DLBCL	Surgery+ Chemotherapy	No evidence of disease since 2yrs.
5.	27y	Breast lump	Right	DLBCL	Surgery+ Chemotherapy	Lost to follow-up.
6.	30y	Breast lump	Left	DLBCL	Surgery+ Chemotherapy	Lost to follow-up.
7.	64y	Breast lump	Left	DLBCL	Sugery+ Chemotherapy	On follow-up. Disease free since 6months.
8.	31y	Breast lump	Bilateral	PBL	Chemotherapy	Undergoing treatment.
9.	60y	Breast lump	Left	DLBCL	Surgery+ Chemotherapy	Succumbed to the disease.
10.	38y	Breast lump	Right	DLBCL	Chemotherapy	No evidence of disease since past 3months.
11.	65y	Breast lump	Right	DLBCL	Surgery+ Chemotherapy	Undergoing treatment.
12.	45y	Breast lump	Left	DLBCL	Chemotherapy	Undergoing treatment.
13	19y	Breast lump	Right	ALCL(ALK+)	Chemotherapy	Disease free since past 2 yrs

DISCUSSION

Breast is one of the least common sites to give rise to lymphomas, probably because of the sparse endogenous lymphoid tissue[3]. Primary lymphomas of the breast mostly occur in the middle-aged or elderly women[3, 4] with exceptionally rare reported cases in males[5]. They are also rarely seen in young, pregnant or lactating women.[3,4,6] The youngest patient in our study was a 19year old woman who was neither pregnant nor lactating. Around 10% of the cases occur bilaterally[4]. The case of plasmablastic lymphoma in our study presented with bilateral breast involvement. Most of the cases reported in the literature presented clinically with breast lumps as was in our study.[6]

HIV infected individuals are associated with an increased risk of

extra nodal lymphomas (ENL), but breast is an uncommon primary site.[7] The common sites of extra nodal lymphomas in HIV infected individuals are gastrointestinal tract, oral cavity and central nervous system and DLBCL is the most common NHL in HIV infected individuals[8].

In most reported series of primary breast lymphomas, DLBCL was the most common lymphoma, accounting for 60-70% of the cases.[3,9,10] It was also the commonest breast lymphoma in a large series of primary breast lymphomas published recently by Perez et al, followed by marginal zone lymphoma and follicular lymphoma.[9] Most DLBCLs of the breast are of non-germinal center/ activated B cell type on immunophenotyping, classified according to Hans algorithm (CD10 -, Bcl6+/-, Mum1+), which is consistent with our study findings.[6]

HIV infected individuals are associated with an increased risk of extra nodal lymphomas (ENL)[7], however, breast is an uncommon primary site. Extra nodal plasmablastic lymphoma occurs more specifically in HIV- infected individuals, with typical occurence in the oral cavity[8]. Other reported sites of occurrence of plasmablastic lymphoma in these individuals are the mucosa of the sinonasal & gastrointestinal tracts, skin, soft tissue and the lymph nodes[11, 12]. Pan- B cell markers like CD20 and PAX-5 are usually not expressed in plasmablastic lymphoma[13], but they express terminal B cell differentiation markers like MUM1 and CD138, as was in our case.

T NHL presenting primarily in the breast is extremely rare and accounts for 2 to 3% of primary breast lymphomas[10]. ALK- positive ALCLs are more commonly extra nodal than ALK- negative ALCLs[12]. ALK- negative ALCLs are seen in association with breast implant, which is a recently described entity[14]. Both the cases in our study are non-implant associated ALCLs and are ALK- positive. The reported literature also shows that, non- implant associated primary ALCL of the breast is very unusual with a very few case reports. [14,15,16,17]

T-NHLs of the breast are reported to behave more aggressively than B-NHLs, with the exception of implant associated ALCL, which has an excellent prognosis[10].

Primary breast lymphomas are essentially treated with chemotherapy or radiotherapy or combination therapy, based on the histological subtype [18,19]. Jennings et al reported that mastectomies offer no benefit in the treatment[19]. However, few of our patients underwent surgical treatment at a primary care level and were referred to our center for further management.

CONCLUSION

Primary breast lymphomas are rare neoplasms of the breast. Their diagnosis can be challenging unless there is a high index of suspicion, as they can mimic other primary breast lesions. Immunohi stochemistry plays a major role in the diagnosis and subtyping of the breast lymphomas and the correct diagnosis has an impact on treatment decisions and thereby, the survival of the patient.

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