Original Article

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Extra Nodal Lymphoma, An Epidemiological and Histomorphological Trend of Primary Extra Nodal Lymphoma in India with Comparative Review of Articles

Sonali Vijaywargi[1], Syeda Firdos Jamil[2], Supriya Bandil[3], Mansi Faujdar[4] and Shubha Gupta[4]

[1]Camberwell Diagnostic and MR Centre, Jaipur, Rajasthan
[2]Department of Pathology, Bhagwan Mahaveer Cancer Hospital and Research Centre, Jaipur, Rajasthan
[3]Jay Pathology, Firozabad UP
[4]Department of Pathology, Santokbha Durlabhji memorial hospital and Research Centre, Jaipur, Rajasthan

ABSTRACT

Background: Geographically there is considerable variation exists in epidemiology of extra nodal lymphomas. Aim: The aim was to study the epidemiological and histomorphological trends of Primary Extra Nodal Lymphoma (P-ENL) in India.

Material & Methods: The biopsy materials from seventy patients with P-ENL (46 male, 24 female, M: F= 1.92:1), diagnosed over a period of two year (2014-2016), were analyzed and pathologically subclassified according to the 2008 World Health Organization (WHO) classification criteria.

Results: Maximum incidence of P-ENL including both sexes was seen in age group 41-60 years. P-ENL constituted 23.72% and secondary extra nodal lymphomas constituted 6.7% of all lymphomas. 98.5% cases of P-ENL were Non-Hodgkin's Lymphomas and 1.4% cases were Hodgkin's lymphoma. The most common site involved by P-ENL was gastrointestinal tract 48.57%, followed by nasopharynx 12.8%, testis 8.4% and salivary gland 7.1%. DLBCL was the commonest lymphoma (72.8%) at all extranodal sites. B-cell phenotype predominated being 94.3% cases, followed by T-cell phenotype being 5.7% cases of P-ENL.

Conclusions: Gastrointestinal tract was the most common site involved by P-ENL followed by nasopharynx. Diffuse large B-cell lymphoma (DLBCL) was the most common subtype followed by marginal zone lymphoma. Majority of P-ENL cases were seen in immunocompetent hosts having a favorable prognosis.

Keywords: *DLBCL, Hodgkin's lymphoma, Non-Hodgkin's Lymphomas, Primary Extra Nodal Lymphoma*

Introduction

Lymphomas are malignant proliferations originating in lymph nodes. These malignant lymphoma cells proliferate and infiltrate in organ other than lymph node is designated as extra nodal lymphomas. Invariably any organ can be involved by extra nodal lymphoma in the body. The gastrointestinal tract most frequently involved (most common site is stomach among them). After this Waldeyer's ring followed by various other organs such as lung, liver, spleen, bone and skin.[1] Tumor origin from any non-lymph nodal tissue is termed as Primary Extranodal Lymphoma (P-ENL), whereas disease spread from lymph nodes to extranodal tissue through hematogenous route is Secondary Extranodal Lymphoma.[2] Comparatively extranodal involvement is seen more commonly in Non-Hodgkin's Lymphoma (NHL) than its counterpart Hodgkin's disease (HL).

In cases particularly with presence of both nodal and extranodal involvement, establishing the diagnosis of extranodal lymphoma is debatable**.** Dawson proposed first definition for gastrointestinal lymphomas, [3] which refined by Lewin [4] and Herrmann [5] later.

The criteria used for diagnosing a case as Extra Nodal Lymphoma (ENL) are: Rigid Criteria: Rigorously limited to extranodal disease at one or multiple anatomical sites in the absence of any nodal manifestation.[3] Liberal Criteria: Allows the existence of a 'minor' nodal component along with 'clinically dominant' extranodal involvement.[6] The majority of existing reports on PE-NHL are based on the latter definition.[6]

The second designation suggests use of Lymphatic and Extralymphatic Lymphoma which itself is questionable and may affect the classification of extranodal lymphomas, since lymphomas not considered as extranodal lesions if originating from tonsils, Waldeyer's ring, thymus, spleen, appendix, and Peyer's patches, as these were thought to be lymphatic tissues.

So, ENL designation is more preferred based on nodal vs. extranodal in routine clinical practice

There is also an issue for debate to designate lymphomas with stage III and IV also as P-ENL, however by many authors only stage I and II lymphomas consider in this category. Although, this motion is not depicting a clear picture as many extranodal lymphomas potentiate to disseminate. Conversely, an extranodal involvement represents a secondary spread in a disseminated disease. [7]

There is inescapably a selection bias interpose, by any of the above definition chosen for designation of ENL; a population-based registry from Dutch study showed that there fluctuation in frequency of extranodal NHL from 20% to 34%, depending on the adopted designation criteria. [6]

Non-Hodgkin's Lymphoma (NHL) is the most common form of ENL and account for 83.17% of lymphoid tumors. The proportion varies from one third to more than half of new NHL cases reported as extranodal presentation e.g. USA and Canada 27%, UK 38%, Denmark 37%, Holland 41%, Italy 48%, HongKong 29%, Thailand 58%, China (44.9%-61.4%) (Yang et al. .[8]) Pakistan and Saudi Arabia (up to 50%) (Nagi et al. .[9]), Kuwait (45%) (Temmim et al., .[10]), Northern Iraq (48.3%) (Yaqo et al. .[11]), Taiwan (47.2%) (Chen et al. .[12]), Japan (46.6%) (Fujita et al. .[13]) and Korea (69.9%) (Kim et al. .[14]) depicted important geographic variations.

Hodgkins Lymphoma accounts for 16.83% of all lymphomas and majority have Nodal presentation. Extra nodal Hodgkin's lymphoma (HL) is rare and has a reported incidence of 2–5%.[15]

Various factors lead to development and pathogenesis of ENL, among them infectious agents (*helicobacter pylori, campylobacter jejuni*, epstein barr virus, human T-lymphotropic virus 1, and hepatitis C virus), genetic susceptibility, autoimmune causes (primary & secondary) play an important role. .[16] Other epidemiologic factors associated with ENL are male gender, increasing age, family history of non-Hodgkin lymphoma, prior cancer history.

A complete history and physical examination are important to provide evidence for nodal and extra-nodal disease or a functional disturbance of an organ system. The diagnosis of Extra Nodal Lymphoma is entirely dependent on biopsy of lesion from relevant sites and final diagnosis is achieved by a combined evaluation of biopsy by light microscopic findings for morphology followed by immunohistochemical evaluation. .[17]

However, there are various challenging issues in Diagnosis of Extra Nodal Lymphoma. A frequent challenge in lymphoma diagnosis is due to significant variation at various level including morphologies, molecular alterations and clinical presentations. [6] There is a wide spectrum of differential diagnosis ranging from reactive condition to inflammatory lesions to poorly differentiated malignancies. Of evaluation by light microscopy and IHC, a small percentage of cases of NHL would further require molecular analysis and flowcytometry, which apart from being expensive and not available in all institute, requires experts for interpretations. [18] Far most important until very recently the literature on many of the specific types and sites of ENL was scant, contradictory & lacking uniformity. [7]

In majority of patients curative treatment for primary extra nodal lymphomas can be provided by the clinicians. In the guidance of modern imaging techniques and careful dosimeter watch, radiation became the cornerstone of extra nodal lymphoma treatment so far. For more aggressive presentations, such as those associated with large B-cell lymphoma, the major modality of treatment is multivalent chemotherapy; however, radiation, and occasionally surgery, often proves very helpful in assuring local control.

As of now primary extra nodal lymphoma is a well-known phenomenon but still needed a lot of work done to define uniqueness at molecular level for this entity.

Due to lack of literature from India and paucity of published studies on lymphoma from northern India, and more so the pronounced heterogeneities in the pathogenesis, clinic-pathological features, and outcome of the various primary extra nodal presentations are important reasons for a detailed consideration of the different sites of origin of these lymphomas and acknowledging the immense role of IHC to achieve an accurate diagnosis.

The present study is designed in the Histopathology Department of Histopathology, Santokba Durlabhji Memorial Hospital cum Medical Research Institute, Jaipur. The purpose is to study the morphologic spectrum of Extra-Nodal NHL and to study the clinic-pathological correlation regarding age, sex and Extra-Nodal site involvement with a brief review of literature.

Materials & Methods

This prospective study has been conducted on 300 patients out of which 70 were selected according to inclusion criteria by non- probability sampling, in the Department of Histopathology, Santokba Durlabhji Memorial Hospital cum Medical Research Institute, Jaipur.

A criterion used for designation of ENL in the present study was involvement of Extra Nodal site with 'minor' regional lymph node involvement. (Stage I and II disease). There is no liver, spleen and distant lymph nodes involvement. Cases with leukemic component or secondary bone marrow involvement, improperly fixed/over fixed tissues, biopsies

with inadequate material to perform immunohistochemical analysis were excluded. (Table-1)

After receiving the specimen in the histopathology department all relevant clinical information regarding age, sex, symptoms, systemic examination, CBC, radiological parameters available (CX-RAY, USG, CT, MRI) for staging and classifying the case as Primary Extra Nodal Lymphoma (P-ENL) of the patients under study had been recorded by interviewing the patient. Biopsy specimen was obtained by endoscopic procedures/excisional/incisional biopsy/ resected specimen. Processing, paraffin embedding, section cutting and staining was done by standardized methods routinely used in the department. Slides were stained by Hematoxylin (Meyer's) and Eosin and further evaluated by IHC markers. H&E-stained sections were examined for provisional morphologic diagnosis/ suspicion of lymphoma. IHC analysis [19] was done by antigen retrieval method BIO GENEX-EZ-Retriever V.3 (Temperature Controlled Microwave). The antibodies used were selected based on the morphologic diagnosis and applied step by step in panels including CD20, CD3, CD5,CD23, Cyclin D1, Bcl2, Bcl6, CD10, MIB-1, MUM-1, CD99, Tdt, LCA, CD15, MIB 1 and Alk protein antibodies. A panel of markers was decided based on morphologic diagnosis. An attempt was made to classify the lymphoma using WHO classification for Lymphomas.

Continuous variables were summarized as mean and standard deviation, while normal/categorical variable as proportion%. Unpaired t test was used for analysis of continuous variables, whereas Chi- square test and Fisher exact test was used for normal/categorical variables. P value < 0.05 was consider as significant. MedCalc Software 12.2.1.0 version was used for all statistical calculations.

Results

Primary extranodal lymphomas constituted 23.72% of all lymphomas, in the present study. 98.5% cases of P-ENL were NHL and 1.4% cases were extra nodal HL. The most common site involved by P-ENL was Gastrointestinal tract 48.57%, followed by nasopharynx 12.8%, testis 8.4%, salivary gland 7.1%, tonsil 5.7%, breast 4.2%, brain, bone, skin and spleen each 2.8%. 1 case (1.4%) of P-ENL was detected in each of sites lung, larynx, pancreas, eyeball and thyroid.

In current study maximum incidence of P-ENL including both sexes were seen in age group 41-60 years 30% followed by age group more than 60 years of age being 21.4%. (Table-2) In present study male predominance (65.72%) was seen with male/female ratio being 1.92:1.

Importance of morphology was in the ability for grouping the NHL according to "cell size" was done, so that panel of antibodies could be chosen to apply in a step wise manner. This optimized the no. of antibodies and was cost effective. (Table-3)

The antibodies were applied in a step wise manner for precise diagnosis and further subcategorized as 51 (74.2%) cases were of diffuse large B-cell lymphoma (DLBCL), 9 (14.2%) were of Marginal zone lymphoma, 2 (2.8%) were of Mantle cell lymphoma, 2 (2.8%) were of Burkitt's lymphoma, 1 (1.4%) case subtyped as Follicular lymphoma, 1 (1.4%) case subtyped as Lymphoblastic lymphoma of T-cell phenotype. 2 (2.8%) cases required a large panel of marker not available in our laboratory and were referred to specialized diagnostic centre for final diagnosis. 1(1.4%) case showed positivity for CD3, CD7, CD 103, CD 30, Granzyme, Perforin and TIA-1 antibodies and was diagnosed as Enteropathy Associated T-Cell Lymphoma (EATL). 1(1.4%) case showed positivity for CD2, CD 3, CD56, Granzyme, Perforin and TIA-1 antibodies. Tumour cells showed EBV RNAS by in situ hybridization was diagnosed as T/NK Cell lymphoma, nasal type. Extranodal Hodgkin's lymphoma constituted 1(1.4%) case of ENL and was subtyped as Classical HL, Mixed Cellularity type. The exact reason for lower occurrence of extra nodal Hodgkin's lymphoma appears to be unknown. (Figure 1-4) (Table-4)

An immunophenotypical subdivision of DLBCL into germinal centre-like(GCB) and non-germinal centre-like(non-GCB) subgroups has been done using a combination of antibodies CD 10, BCL-6 and MUM-1. Cases with CD10 +ve in 20(28.57%) cases and CD 10-ve, BCL-6 +ve and MUM 1 -ve in 8(11.43%) cases, totally 28 (40%) cases were diagnosed as Germinal centre-like(GCB) and all other cases ie. BCL-6 -ve 5(7.14%) cases and CD 10 -ve, BCL-6 +ve and MUM-I +ve 15(21.46%) cases, a total of 20 (28.6%) were diagnosed as Non-germinal centre like (non-GCB). (Figure 2)

B-cell phenotype predominated being 94.3% cases, followed by T-cell phenotype being 5.7% cases of P-ENL. (Table-4) There was no case of SLL/CLL in the present study.

Discussion

Extra nodal sites involvements by malignant lymphomas are fascinating. Extra nodal non Hodgkin lymphoma (ENL) is a heterogeneous disease in regard to geographical, ethnic, anatomic, etiological, and morphological diversities.[6] The frequency of ENL varies in different parts of the world, and is rising throughout the world.

The percentage of P-ENL out of all lymphomas in the present study is 23.72%. The incidence of P-ENL varies widely and is lower in India than other parts of the world.

Table 1: Distribution of 300 cases of Lymphoma according to inclusion and exclusion criteria

P-ENL (satisfying inclusion criteria)	70	Included
Nodal lymphomas	210	Excluded
Extra nodal site involvement with extensive disease (Stage III and IV disease)-secondary ENL	5	Excluded
Cases of lymphoma with leukemic component	10	Excluded
Inadequacy of tissues for IHC/ Not suitable for IHC	5	Excluded
Total	**300**	

Table 2: Distribution of 70 cases of primary Extra Nodal Lymphoma according to age.

Age group	No	%
<10	4	5.7
10 to20	2	2.8
21 to 30	7	10
31 to40	9	12.8
41 to 50	10	14.3
51 to60	11	15.7
>60	15	21.4
Total	**70**	**100**

Table 3: Distribution of 70 cases of P-ENL on the basis of "cell size". (Morphologic groups based on cell size).

Morphological diagnosis		N	%
NHL with "small size cells"	Low Grade NHL	12	17.14
NHL with Medium to large "centroblastic and centrocytic cells"	Intermediate grade NHL	49	70
NHL with Medium to large "Blastoid cells"	High grade NHL (lymphoblastic lymphoma)	1	1.4
	High grade NHL (Burkitts)	1	1.4
NHL with large "immunoblastic cells"	High Grade NHL (Immunoblastic)	6	8.6
RS Cells in reactive background	Hodgkin's Lymphoma	1	1.4
Total		**70**	**100**

Table 4: Distribution of 70 cases of P-ENL (WHO classification).

WHO classification		No	%
B-cell lymphoma 65(92.8%)	Diffuse large B cell lymphoma (DLBCL)	51	74.2
	Marginal Zone lymphoma (MZL)	9	14.2
	Mantle Cell Lymphoma (MCL)	2	2.8
	Follicular lymphoma(FL)	1	1.4
	Burkitt's Lymphoma	2	2.8
T-cell Lymphoma 4(5.7%)	Peripheral T-Cell lymphoma (ALCL Type)	1	1.4
	T-Lymphoblastic Lymphoma	1	1.4
	T/NK Cell Lymphoma, Nasal Type	1	1.4
	Enteropathy associated T-Cell Lymphoma	1	1.4
Hodgkin's Disease 1 (1.4%)	Classical Hodgkin's Lymphoma (Mixed cellularity type)	1	1.4
Total		**70**	**100**

Table 5: Percentage of P-ENL according To Different Sites as per Various Studies (Comparative review of literature).

S. No	Site/ Study	Present study	Yang et al	Kim et al	Abeer et al	Aparna et al	Padhi et al	Mishra et al	Yaqo et al
1	GIT	48.57	22.3	42	35.7	12.9	25	29	41
2	Nose/NP	12.8	20.4	12	10.9	22.5	11.8	9	16
3	Testis	8.4	-	-	-	-	-	2	-
4	Salivary gland	7.1	3	0.3	4.3	-	-	-	1
5	Breast	4.2	1.5	0.9	-	3.2	-	2	2
6	Brain	2.8	2	5	2	9.6	29.5	4	2
7	Bone	2.8	2	2.3	8.7	6.4	-	2	3
8	Skin	2.8	11.3	6	19.6	12.9	-	-	13
9	Spleen	2.8	-	1	-	-	-	-	-
10	Lung	1.4	-	1.6	-	3.2	-	-	-
11	Larynx	1.4	-	-	-	-	-	-	-
12	Thyroid	1.4	1	-	2.2	3.2	-	2	2
13	Pancreas	1.4		-	-	-	-	-	-
14	Eyeball	1.4	-	9.3	1	3.2	-	-	-
15	Waldeyers ring	-	23.7	12.8	8.7	-	-	16	-
	Others	-	12.8	6.8	6.9	22.9	33.7	34	20

Table 6: Percentage of different subtypes of P-ENL as per various studies (Comparative review of literature).

S. No	Subtype	Present study	Yang et al	Padhi et al	Mishra et al	Abeer et al	Yaqo et al
1.	DLBCL	72.8	42.2	69	54	47.9	46.46
2.	Marginal	12.8	11.0	13.7	12	21.7	8
3.	Mantle	2.8	2	-	-	2.2	1
4.	Burkitt's	2.8	2	2.8	4	-	29.30
5.	Follicular	1.4	2	-	-	-	1
6.	T- Lymphoblastic lymphoma	1.4	2	-	6	-	1
7.	EATL	1.4	0.24	-	-	-	1
8.	T/NK Cell	1.4	30.7	-	3	-	-
9.	PTCL	1.4	6	1.4	13	8.7	1
10.	Hodgkin's	1.4	-	-	-	-	-
11.	Others	0.4	0.16	12.1	8	19.5	11.24

Table 7: Incidence rate of B-cell and T-cell lymphoma in various studies.

Authors Name (year)	B-cell lymphoma	T-cell lymphoma
Present study	94.4	5.6
Padhi et al	96	4
Yaqo et al	91	9
Aparna et al.	87	13
Abeer et al	85	15
Mishra et al	82	18

Table 8: Distribution of 70 cases of P-ENL according to age and subtype (Subtype and Age correlation).

Subtype of NHL	1-15		16-30		31-45		46-60		>60		Total	
	No	%	No	%	No	%	No	%	No	%	No	%
DLBCL	1	1.9	6	11.7	12	23.2	20	39.2	12	23.5	51	100
Marginal	-	-	-	-	6	66.7	3	33.3	-	-	9	100
Mantle	-	-	-	-	1	50	-	-	1	50	2	100
Burkitt's	2	100	-	-	-	-	-	-	-	-	2	100
FL	-	-	-	-	-	-	1	100	-	-	1	100
T-LL	-	-	1	100	-	-	-	-	-	-	1	100
T/NK Cell	-	-	1	100	-	-	-	-	-	-	1	100
EATL	-	-	1	100	-	-	-	-	-	-	1	100
PTCL	-	-	-	-	-	-	-	-	1	100	1	100
Hodgkin's	-	-	1	100	-	-	-	-	-	-	1	100
Total	**3**	**4.2**	**10**	**14.3**	**19**	**27.1**	**24**	**34.3**	**14**	**20**	**70**	**100**

Table 9: Distribution of 70 cases of ENL according to subtype and site. (Subtype and Site correlation).

SITE/ SUBTYPE	DLBCL		MZL		BL		FL		MCL		T-LL		PTCL		T/NK		EATL		HD		Total	
	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%
GIT	24	70.6	4	11.4	2	6	1	3	2	6	-	-	-	-	-	-	1	3	-	-	34	100
Nose/NP	8	88.9	-	-	-	-	-	-	-	-	-	-	-	-	1	11.1	-	-	-	-	9	100
Testis	5	83.3	-	-	-	-	-	-	-	-	1	16.7	-	-	-	-	-	-	-	-	6	100
Salivary Gl	3	60	2	40	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	5	100
Breast	2	66.7	-	-	--	-	-	-	-	-	-	-	-	-	-	-	-	-	1	33.3	3	100
CNS	2	100	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2	100
Skin	1	50	-	-	-	-	-	-	-	-	-	-	1	50	-	-	-	-	-	-	2	100
Spleen	1	50	1	50	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2	100
Bone	2	100	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2	100
Lung	-	-	1	100	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	100
Larynx	-	-	1	100	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	100
Pancreas	1	100	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	100
Eyeball	1	100	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	100
Thyroid	1	100	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	100
TOTAL	**51**	**72.9**	**9**	**12.9**	**2**	**2.8**	**1**	**1.4**	**2**	**2.8**	**1**	**1.4**	**1**	**1.4**	**1**	**1.4**	**1**	**1.4**	**1**	**1.4**	**70**	**100**

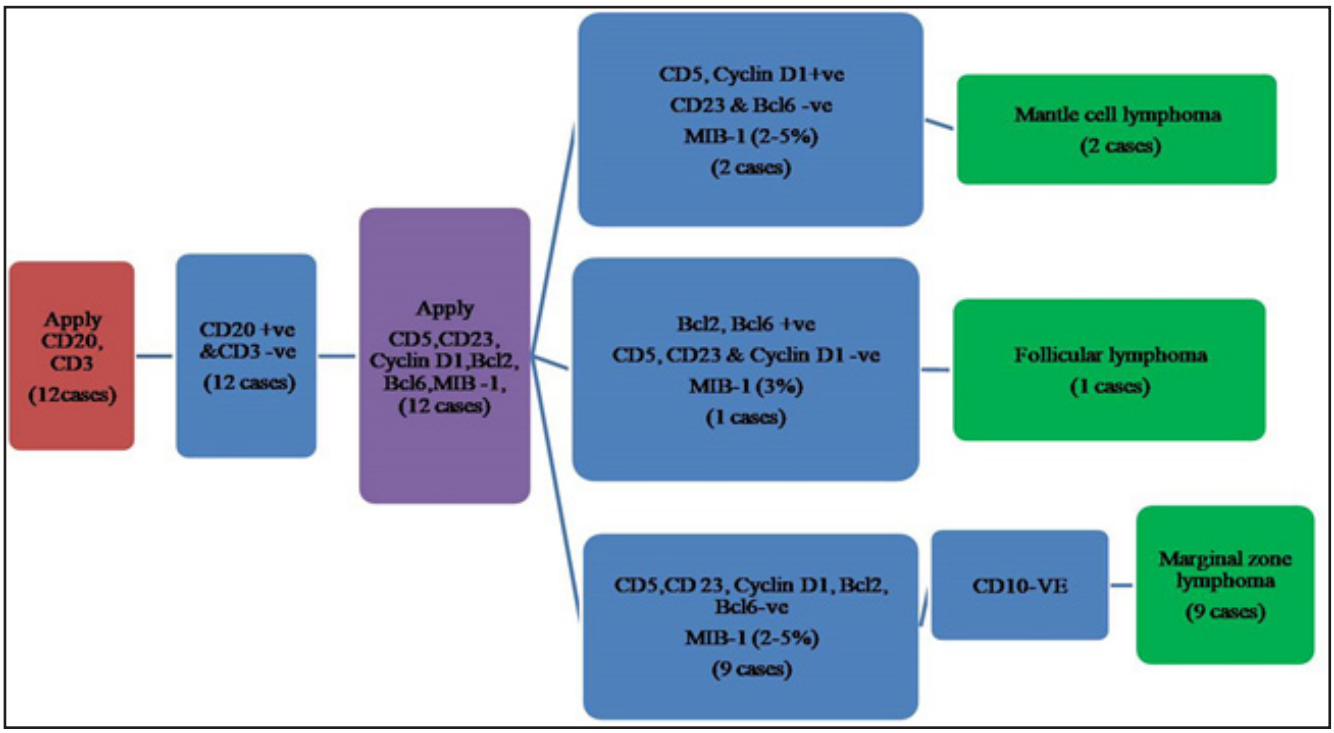

Fig. 1: Antibody panels used for diagnosis of NHL with "small & mature lymphoid cells" - 2 cases positive for CD20,CD5 & Cyclin D1 & negative for CD3, CD23, Bcl6, Mib index 2 and 4%, diagnosed as Mantle cell lymphoma. 1 case positive for CD20,Bcl2 & Bcl6, Mib index 4% and negative for CD3, CD 5, CD23 & Cyclin D1, was diagnosed as Follicular Lymphoma. 9 cases positive for CD20 & MIB1 index 2-5% & negative for CD3,CD5,CD23,Cyclin D1, Bcl6 & CD10, diagnosed as Marginal Zone Lymphoma.

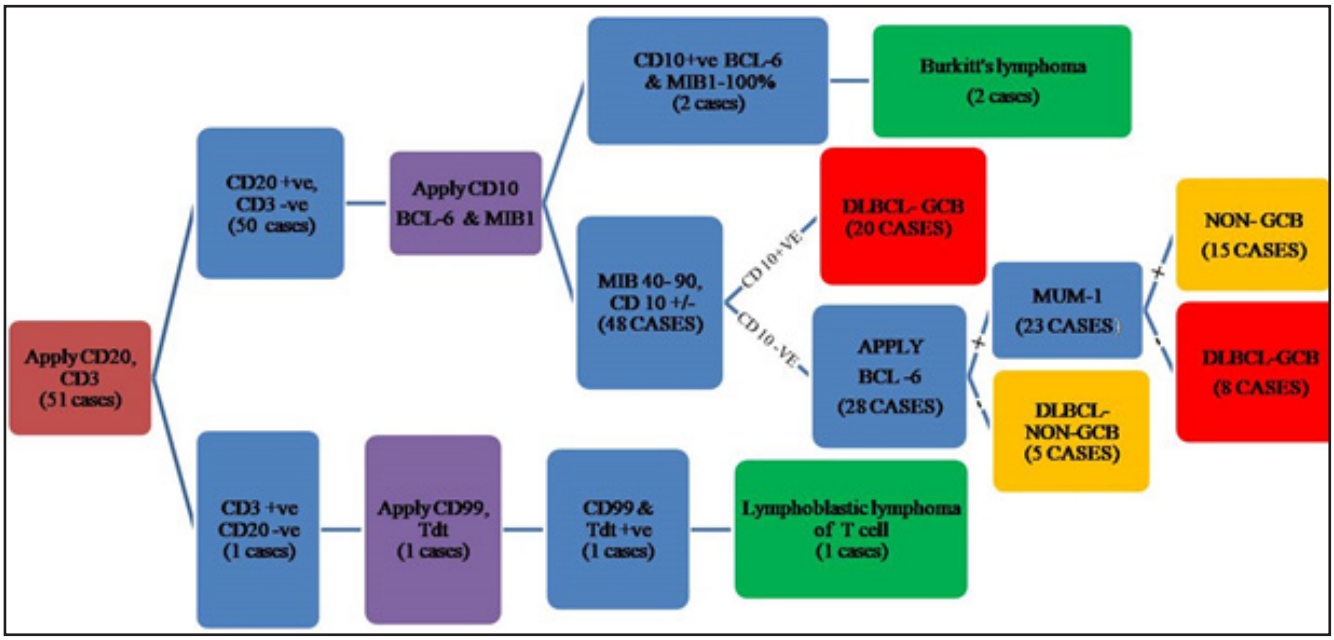

Fig. 2:Antibody panels used for medium to large "Blastoid cells" and "centroblastic & centrocytic cells"- 2 cases positive for CD20, CD10 & negative for CD3, Mib index 100%, diagnosed as Burkitt's lymphoma. 1 was positive for CD3, CD99, Tdt & negative for CD20, diagnosed as Lymphoblastic lymphoma of T cells. 48 were positive for CD 20, Mib index 40-90% & negative for CD3, diagnosed as DLBCL . DLBCL subdivided into GCB & non-GCB subgroups (using CD 10, BCL-6 & MUM-1).

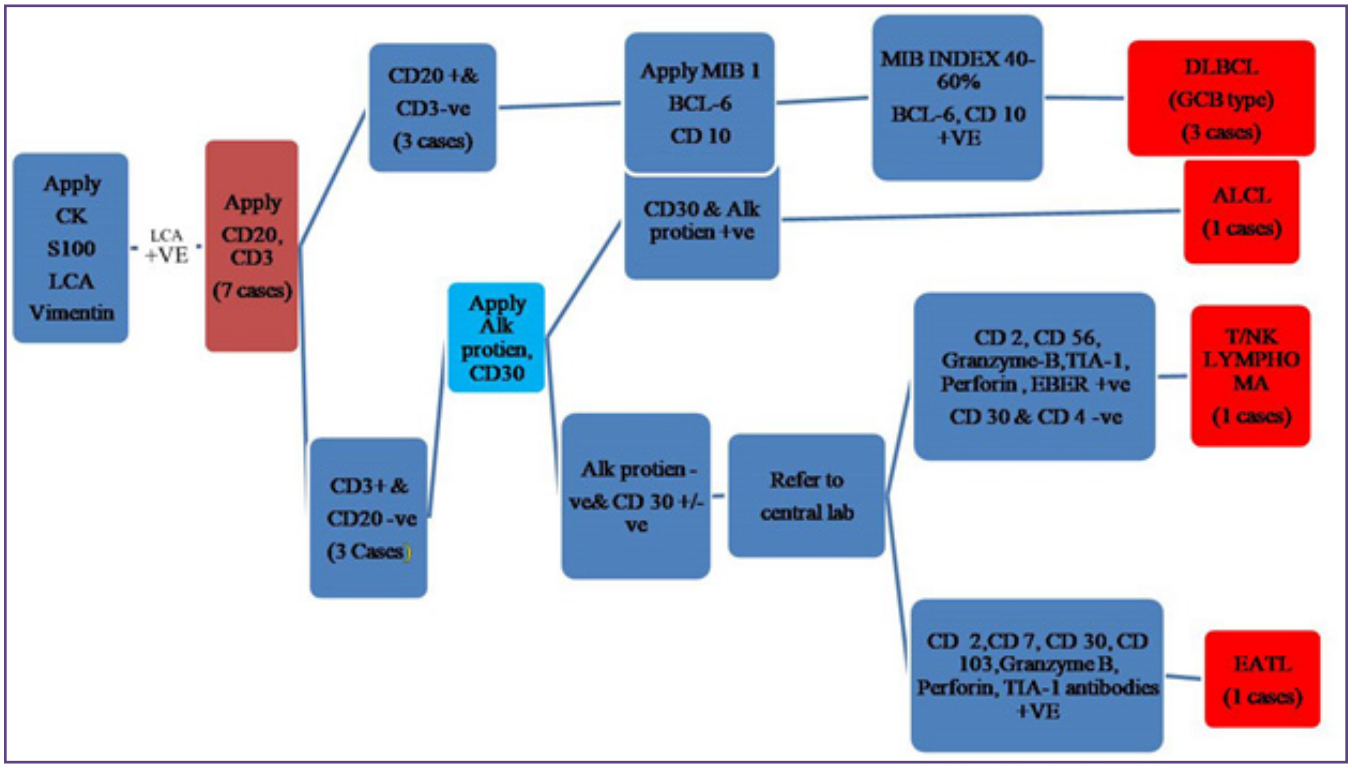

Fig. 3: Antibody panels used for diagnosis of NHL "large (Immunoblastic) cells" - 3 cases positive for CD20, BCL-6, CD 10 & negative for CD3, MIB1 index 40-60%, dignosed DLBCL- GC type. 1 showed positivity for CD3, CD30, Alk protien & negativity for CD20, diagnosed PTCL (ALCL type). 1 case positive for CD3, CD7, CD 103, CD 30, Granzyme, Perforin, TIA-1, diagnosed as EATL. 1 case showed positivity for CD2, CD 3, CD56, Granzyme, Perforin, TIA-1 & EBV RNAS, diagnosed T/NK Cell lymphoma.

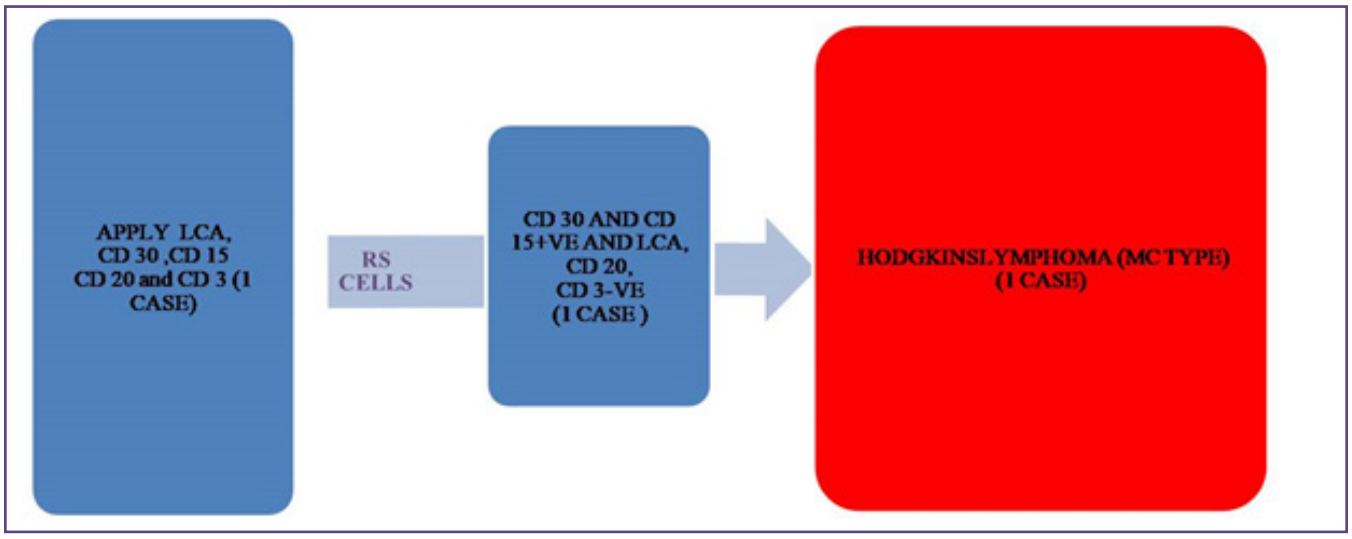

Fig. 4: Antibody panels used for diagnosis of P-ENL with "Reed sternberg Cells" - One case which showed presence of Reed sternberg cells on morphology. The RS Cells showed positivity for CD15 and CD 30 antibodies and showed negativity for CD 20, LCA and CD 3 antibodies and was diagnosed as Classical Hodgkin' s lymphomas, mixed cellularity type.

[21] Our study is in concordance with two studies from South India, Padhi et al[20] and Mishra et al[21] which have shown the incidence to be 22% each. In a study from North India by Singh et al[22] P-ENL constituted 44%. There is occurrence of 24-48% of extranodal NHL reported from different studies conducted in western countries.[7]

However, possible explanation of lower percentage of P-ENL in our study is due to small duration of study period with exclusion Lymphomas arising in tonsil and waldeyer's ring & stage III and IV lymphomas with involvement of extranodal site.

In the present study most common site involved by P-ENL is gastrointestinal tract 48.57%, followed by nasopharynx 12.8%, then testis 8.4%, salivary gland 7.1%, breast 4.2%, etc in decreasing order.

A comparative review of literature pertaining to site is given below. (Table-5)

Our study is in concordance with Kim et al[14], Abeer et al[23] and Yaqo et al[11] who reported GIT to be the most common site. This may be due to higher incidence of H.pylori and more Richter transformation of low grade MALT and follicular lymphoma. (Arora et al.) [24]

Our study reported nasopharynx to be the second most common site constituting 12.8% of cases. Epstein Barr Virus, agricultural pesticides, environmental pollutants, poor oro-dental hygiene/tobacco chewing more predilection of NK/T cell lymphoma phenotypes for head and neck region, were reported to be the possible explanation for this difference. Besides this, among Asians low DLBCL incidence and high NK/T ell lymphoma incidence as compared to the western and Middle East countries, possibly explains these diversities (Yaqo et al.). [11]

Padhi et al[20], reported Primary CNS lymphoma to be the commonest in their study 29.5%. The increased number of PCNL in their series was explained to be because of improved neuroimaging techniques and stereotactic brain biopsies studied rather than anything else.

Diversity in sites of P-ENL is difficult to explain. These variations could again be explained as genetic and geographical factors were different in various regions. Chiefly, all above mention studies are hospital based hence not presenting the true demographic profile of population.

A comparative review of literature pertaining different subtypes of P-ENL as per various studies (Table-6)

In the present study diffuse large B-cell lymphoma (DLBCL) is most common subtype comprising of 72.8%. Majority of the studies by Yaqo et al[11], Yang et al[8], Padhi et al[20], Mishra et al[21], Abeer et al[23] reported DLBCL to be the commonest P-ENL with a great variation in percentage. Marginal zone lymphoma is the second most common P-ENL comprising 12.8% cases. A similar percentage is reported in Indian studies by Padhi et al[20], Mishra et al[21] i.e. 13.7% and 12% respectively. Abeer et al[23] reported a higher percentage being 21.7%. Yang et al[8] reported a slightly lower percentage being 11%.

Burkitt's lymphoma was diagnosed in 2.8% cases. Padhi et al[20] reported Burkitt's lymphoma being 2.8% same as in present study. Naresh et al[25], Yang et al[8] and Mishra et al[21] reported a similar percentage of Burkitt's lymphoma being 1.8%, 2% and 4% respectively.

Yaqo et al[11] reported a much higher incidence of intestinal Burkitt's lymphoma in middle east countries being 29.30%.

In present study, mantle cell lymphoma, constituted 2.8% with a concordance with Abeer et al[23] and Yang et al[8] with the proportion being 2.2% and 2% respectively. Yaqo et al[11] reported a slightly lower percentage being 1%. In the present study follicular lymphoma, PTCL (ALCL Type) Lymphoblastic lymphoma, EATL, T/NK Cell Lymphoma, Nasal Type, Extranodal Hodgkin's Lymphoma each constituted 1.4% of all cases.

These regional variations could be attributed to differences in genetic and geographical factors. Poor Socio economic status in eastern countries compared to west accounting for delayed diagnosis of a pre-existent low grade MALT or follicular lymphoma which led to high grade transformation into DLBCL at extra nodal site, can explain the high percentage of DLBCL in these studies. [8] Geographic variation in molecular expression profiling in follicular lymphomas as well as between nodal and extra nodal sites may be one of the possible explanation of diversity, suggested by Biagi and Seymour[26] and Yang et al. [8]

A relatively higher percentage of Burkitt's lymphoma in populations of Eastern Mediterranean reported by Yaqo et al[11] requires further scrutiny, and is currently the subject of an ongoing study by their group.

Yang et al[8] reported a strikingly high percentage of Extra nodal T/NK Cell Lymphoma in their series. They proposed strong association of environmental factors such as pesticides and chemical agents as well as EBV infection with this disease. In addition, the high proportion of referral cases of head and neck in their study (Hospital Bias), may also contribute to the high percentage of Extra nodal T/NK Cell lymphoma.

Despite a relative prominence of extra nodal presentations, the literature on their incidence and on most of the specific types and sites is scant and often contradictory.

This is primarily because these tumors, numerous when considered together, are distributed so widely throughout the body that it is difficult to assemble adequate series with enough numbers, of any given site. [7]

A comparative review of literature pertaining incidence rate of B-cell and T-cell lymphoma in various studies (Table-7)

In the present study, B-cell phenotype was predominant constituting 94.3% of P-ENL. Padhi et al[21], Yaqo et al[11], Aparna et al[27], Abeer et al[23] and Mishra et[21] al reported B cell lymphoma as predominant phenotype being 96%, 91%, 87%, 85% and 82% respectively.

In the present study only 5.7% of P-ENL showed T-cell phenotype. Padhi et al[21], Yaqo et al[11] reported similar percentage of T cell lymphoma being 4% and 9% respectively. Aparna et al[27], Mishra et al[21],and Abeer et al[23] reported a higher percentage of T-cell lymphoma 13%,18% and 15% respectively. Proposed reasons for a higher percentage of T-cell lymphomas in various studies of Aparna et al[27], Mishra et al[21] and Abeer et al[23] could likely be due to the strikingly high percentage of Extra nodal T/NK Cell Lymphoma, PTCL-NOS in these series. Research has suggested that strong association of environmental factors such as pesticides and chemical agents as well as EBV infection with T-cell lymphomas.

In current study maximum incidence of P-ENL including both sexes was seen in age group 41-60 years 30% followed by age group more than 60 years of age being 21.4%. Results of study conducted by Mishra et al[21] and Padhi et al[20] were in agreement with our study. They reported that 29.4 % and 36.8% of P-ENL between 4th and 5th decade respectively. Maximum no. cases of DLBCL (45.7%), follicular lymphoma (100%), PTCL - ALCL Type (100%) and mantle cell lymphoma (50%) were diagnosed in >46 years of age. Yang et al[8] reported incidence of DLBCL, follicular lymphoma, PTCL and mantle cell lymphoma in >50 years of age. In present study, all cases of Burkitt's lymphoma (100%) were diagnosed in <15 years of age. (Table-8)

P-ENL is more common in older adults than younger adults; hence age presents to be strong risk factor for this disease. Incidence data obtained from the United States National Cancer Institute's Surveillance, Epidemiology, and End Results (SEER) Program accounted that the incidence of total lymphoid neoplasm increased monotonically with age in all race and sex subgroups. [28]

In present study male predominance (65.72%) was seen with male/female ratio being 1.92:1. Most subtypes of P-ENL were prone to involve males. Male predominance was observed for diffuse large cell lymphoma, mantle cell lymphoma and Burkitt's lymphoma. Our study is in concordance with various studies by Padhi et al[20], Mishra et al[21] and Yaqo et al[11] with M:F ratio being 2:1, 1.3:1 and 2:1 respectively.

DLBCL was the most common subtype encountered at almost all the sites involved by P-ENL constituting 24 (70.6%) cases of all GIT lymphomas, 8(88.9%) cases of all nasopharyngeal lymphomas, 5(83.3%) cases of testicular lymphomas, 3(60%) cases of salivary gland lymphomas. Marginal zone lymphoma was the second most common P-ENL encountered constituting 4(11.8%) cases of all GIT lymphomas, 2(40%) cases of salivary gland lymphomas, 1(50%) case was of splenic marginal zone lymphoma. Lung and larynx showed 1(100%) case each of marginal zone lymphoma. (Table-9)

Concerning the distribution of histologic subtypes at various extranodal sites, Yang et al[8] reported DLBCL and EATL made up the majority of NHL in Waldeyer's ring; DLBCL and MALT composed over 80% of all lymphomas in the gastrointestinal tract; EATL primarily represented the sinonasal lymphomas. For skin lymphomas, except for MF, SPTCL and EATL were relatively common in their study.

To conclude this study reiterates the key role of IHC in diagnosis of P-ENL. IHC is required for the confirmation of diagnosis, phenotyping and classification of lymphoid malignancies, Using WHO classification for lymphomas all cases of P-ENL were diagnosed and subtyped into clinically and prognostically relevant entities.

This review has allowed us to identify following areas which need serious considerations. It should be mandatory to perform IHC for diagnosing all cases of P-ENL prior to chemotherapy. To know the frequency of subtypes of P-ENL in this region a similar study with larger no. of P-ENL cases, over a longer duration, should be attempted. A national lymphoma registry should be established.

Abbreviation

ALCL: Anaplastic large cell lymphoma

HL: Hodgkin Lymphoma

IHC: Immunohistochemical

MALT: Mucosa Associated lymphoid Tissue

Nodal-NHL: Nodal Non Hodgkin's Lymphoma

NHL: Non Hodgkin's Lymphoma

PBS: Phosphatase buffered saline

DLBCL: Diffuse large B cell lymphoma

PTCL: Peripheral T-cell lymphoma

SPTCL: Subcutaneous Panniculitis like T-cell Lymphoma.

EATL: Enteropathy Associated T-Cell Lymphoma.

MF: Mycosis Fungoides

MCL: Mantle cell lymphoma

MZL: Marginal zone lymphoma

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***Corresponding author:**
Dr Syeda Firdos Jamil, Plot No 17, Mukund Garh House, Sansar Chandra Road, Jaipur 302001
Phone: +91 9460496076
Email: Rajasthan,fidsjamil47@gmail.com

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