Abstract

Background: Plasmablastic lymphoma (PBL) is an aggressive B-cell lymphoma that is characterized by the expression of plasma cell antigens and loss of pan B-cell antigens. The neoplasm is extensively reported in the oral cavity and anorectal region but rarely in the gastrointestinal tract where isolated case reports and small case series exist. In the current study, the morphologic, immunohistochemical and molecular features of 17 cases of gastrointestinal tract PBL were reviewed.

Materials and Methods: Ten-year retrospective study that reappraised the histomorphological and immunophenotypic features of HIV-associated PBL in the gastrointestinal tract those were diagnosed and coded as ‘plasmablastic lymphoma’.

Results: The average age of the study subjects was 41 years with a 3:1 ratio of males to females. The most frequent site of involvement was the small intestine (42%). Majority of the cases showed a predominant diffuse (82%) and single cell (29%) growth patterns. Immunoblastic and plasmablastic cytology was common in all cases. Sixty-five percent of the cases exhibited scattered centroblasts and one case demonstrated predominance of centroblasts. Other features observed include pseudoalveolar growth pattern, plasmacytic differentiation (25% cases), multinucleated giant cells, clear cell change, high mitotic activity with high proliferative indices (Ki-67 >90%), apoptosis (100% cases) and necrosis (71% cases). Immunohistochemistry revealed absence of pan B-cell antigens and expression of plasma cell antigens. Epstein-Barr virus-encoded RNA was expressed in 53% of the cases and one case showed c-MYC translocation.

Conclusion: This study highlights the spectrum of histopathological features of gastrointestinal tract PBLs. Additional observations not previously described or emphasized in literature includes pseudo alveolar growth pattern, Centro blast-predominance, multinucleated giant cells and clear cell change. Awareness of the histopathological spectrum and immunohistochemical profile of gastrointestinal tract PBLs may aid in accurate diagnosis and avert potential diagnostic errors.

Keywords: Plasmablastic lymphoma; HIV-related lymphoma; AIDS-related lymphoma; Gastrointestinal tract; Stomach; Small intestine; Colon

Introduction

Plasmablastic Lymphoma (PBL) is an aggressive, high-grade B-cell non-Hodgkin lymphoma characterized by the proliferation of large B-immunoblast like neoplastic cells that display plasma cell differentiation associated with loss of conventional B-cell antigens [1,2]. Delecluse et al. originally described PBL in HIV positive patients as a subtype of diffuse large B-cell lymphoma confined to the oral cavity [3]. The neoplastic cells displayed frequent association with Epstein-Barr Virus (EBV) infection. PBLs have been reported in the anorectal region, gastrointestinal tract, skin, lung, nasopharynx, paranasal sinuses, lymph nodes, central nervous system and soft tissues [4,5]. Several studies have also reported on PBLs in HIV negative patients with and without immunosuppression [6-9]. PBLs arising in HIV-negative and HIV-positive patients have different clinicopathologic characteristics, including younger age, male predominance and better response to chemotherapy in HIV-associated PBLs [6].

Extra-oral PBLs are associated with non-HIV related immunosuppression and commonly display plasmacytic differentiation [5]. Epstein-Barr Virus Encoded RNA (EBER) in situ hybridization positivity has been described in 60-75% of PBLs [10]. HIV positive patients more frequently have EBV-positive PBLs compared with HIV-negative patients [11]. PBLs can be subdivided into two morphologic subgroups including lymphomas comprised of a monomorphic population of immunoblasts and lymphomas with plasmacytic differentiation composed of immunoblasts, plasmablasts and cells showing plasma cell differentiation [1,5]. Cytogenetic studies have found c-MYC translocation in 50% of PBLs cases and this is associated with MYC-protein overexpression [12]. EBV positive neoplasms are more likely to have c-MYC rearrangements when compared to EBV negative neoplasms [12].

A considerable body of literature on oral plasmablastic lymphoma is available [13-16]. However, the histopathological features and descriptions of extra-oral PBLs are infrequently reported particularly in the stomach, small intestine and colon, hence the need for a
comprehensive examination of PBLs in these segments of the gastrointestinal tract to improve on timeous diagnosis and early intervention.

Materials and Methods

The current retrospective study involved purposive sampling and evaluation of all biopsies diagnosed and coded as ‘plasmablastic lymphoma’ from 1st January 2009 to 31st December 2018. The cases were accessed from the electronic archive of the Department of Anatomical Pathology, Inkosi Albert Luthuli Central Hospital, National Health Laboratory Service, Durban, KwaZulu-Natal, South Africa, using the SNOMED word and code search engines.

The following limited clinical information was retrieved from departmental records: age, sex, HIV status and gross description of the tumor. All patients included in the study were HIV positive and only biopsies from the stomach, small intestine and colon were included. Biopsies from oral cavity and anorectal region were excluded from the study. Two pathologists (A.M., G.N) independently reappraised stored sections on Olympus BX43 (Olympus, Tokyo, Japan) microscopes. Whenever a discrepancy occurred, both investigators re-examined the slides to reach a consensus.

The original 4 μm thick hematoxylin and eosin stained slides were reviewed to evaluate the following histopathologic features: architectural growth patterns (diffuse, nodular, trabecular and/or single cells) and cytomorphology (plasmablastic, immunoblastic, centroblastic or plasmacytic differentiation). The mitotic activity (expressed as mitotic figures per 10 high power fields in the most mitotic area, using a 40x objective and a 10x ocular lens; field area: 0.237 mm²), apoptotic bodies and tumor necrosis were also evaluated. Immunohistochemical stainings used to establish the initial diagnosis by the primary pathologist were reviewed. If necessary and where enough material was available, staining’s were repeated on sections cut at 2 μm thickness, using manufacturer’s instructions on an automated immunostainer (Ventana Benchmark Ultra, Tucson, AZ). Appropriate positive and negative controls were used throughout.

Digital images for cytogenetic studies results were reviewed for two cases on which they had been conducted successfully. Fluorescence in situ hybridization (FISH) was performed with Vysis LSI MYC dual-color, break-apart rearrangement probe (Abbott Molecular, Des Plaines, IL, USA) and was carried out on 4 μm-thick formalin-fixed, paraffin-embedded tissue sections following manufacturer’s guidelines.

From the 18 archival cases, one case was excluded after reclassification as diffuse large B-cell lymphoma with CD20 immunopositivity. The Biomedical Research Ethics Committee of the University of KwaZulu-Natal approved the study (BREC Ref No: BE016/19).

Results

Seventeen cases of gastrointestinal tract PBLs were confirmed in the study period. Thirteen (76%) patients were male and four (24%) were female. Age was documented for 16 patients; it ranged from 29 to 68 years with an average of 41 years. Nineteen (19) percent were less than 30 years, 25% were from 30 to 39 years, 37% from 40 to 49 years, and 19%, 50 years or older. The biopsies were either incisional (53%, 9/17) or excisional (47%, 8/17). The region of involvement was in the small intestines (42%, 7/17), stomach 29% (5/17) and the colon 29% (5/17). Details on gross tumour size were available for six of the eight excisional biopsies and the tumour ranged in size from 3 to 8 cm (average 5.4 cm).

Histopathological features

On microscopic appraisal, the neoplasm showed one or more architectural patterns with diffuse being the most common (82%, 14/17), followed by scattered single cells (29%, 5/17), multinodular (24%, 4/17), trabecular (18%, 3/30) and pseudo alveolar (12%, 2/17) growth patterns (Figures 1A-F). A low power starry-sky pattern was seen in most excision biopsy specimens (75%, 9/17) (Figure 1A). The incision biopsy specimens showed starry sky appearance in 56% (5/9) of the cases.

All cases showed proliferation of large lymphoid cells with

![Figure 1](image.png)

**Figure 1**: Low power view showing a submucosal tumour with various growth patterns, (A-B) Diffuse architecture and starry-sky pattern; (C) Multinodular architecture; (D) Trabecular; (E) Pseudo alveolar; (F) Medium power- Single cells.
immunoblastic and plasmablastic morphology with occasional presence of paranuclear hof (Figures 2A-2C). Scattered centroblasts were identified in 11 (65%) cases. One case showed centroblasts as the predominant cell type (Figure 2D). Plasmocytic differentiation was identified in 29% (5/17) of the cases. Most cases (71%) showed amphophilic cytoplasm while the remaining 29% exhibited eosinophilic cytoplasm. Two cases showed focal clear cell change (Figure 2E). Scattered multinucleated tumour giant cells were observed in two cases (Figure 2F).

All cases in the present study had identifiable mitotic figures, ranging from 16 to 62 (mean 32) per ten high power fields. Apoptotic bodies were observed in all biopsies and these were arranged as single cells or as confluent areas of apoptotic bodies. Necrosis was evident in 71% (12/17) of the biopsies.

**Immunohistochemistry**

Summarized in Table 1 are the immunohistochemical results. CD20 was negative in all cases. The neoplastic cells were positive for at least one plasma cell differentiation marker, with MUM1 being the most common (83%, 15/18), followed by VS38c (61%, 11/18) (Figures 3A-3D). One case showed T-cell infidelity with neoplastic cells showing immunopositivity for CD3 and CD4 whilst CD2, CD5 and CD8 were negative.

Thirty-three percent (5/15) of the cases showed patchy immunopositivity for CD30. All cases which were tested for broad-spectrum cytokeratin’s (7/17), CD56 (3/17), ALK-1 (10/17) and HHV8 (8/17) were negative. The Ki-67 proliferative index examined in nine cases was high (>90%) (Figure 3G). *In situ* hybridization, for EBER was positive in 53% (9/17) of the cases under review (Figure 3H).

![Figure 2: High power tumour morphology ranged from: (A-C) Proliferation of plasmablastic and immunoblastic cells with amphophilic and eosinophilic cytoplasm and occasional paranuclear hof; (D) Centroblasts with peripherally based nucleoli; (E) Two cases showed clear cell change; (F) Occasional multinucleated giant cells.](image)

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+/−: Weak positive; +: Positive; −: Negative; ND: Not done; CK: Cytokeratin (AE1/3, CAM5.2, MNF116)

**Table 1:** The immunohistochemical characteristics of the plasmablastic lymphoma cases.
Cytogenetic studies

Fluorescence In situ Hybridization (FISH) for c-MYC rearrangements was successfully performed on one case which showed c-MYC translocation. One case showed Immunoglobulin Heavy (IGH) chain clonality. The other cases in the study did not have cytogenetic analysis done or it was unsuccessfully attempted.

Discussion

Plasmablastic Lymphoma (PBL) is a highly aggressive CD20 negative B-cell lymphoma that is increasingly reported in HIV positive patients [16]. Although originally described in the oral cavity of HIV positive patients, occurrence in other sites and HIV negative patients has been documented [17]. Most of the gastrointestinal tract PBLs have been described in the stomach, small intestine and colon as case reports and small case series [18-25]. Herein, we report on 17 patients with gastrointestinal tract PBLs focusing on the morphological and immunohistochemical features.

The neoplasm showed a predilection for males and had age ranging between 29 to 68 years with an average of 41 years. The male predominance is like what has been reported in other smaller case series [25].

The major histopathological features of PBLs include a predominantly diffuse growth pattern, starry-sky appearance on low power, necrosis, apoptotic cells, and neoplastic cells showing predominantly immunoblastic and plasmablastic cytomorphology. The findings of the present study showed comparable features to those reported in the case studies and series on gastrointestinal tract PBLs [18-25]. The characteristic immunoblastic cells exhibit round to oval nuclei with vesicular chromatin and prominent central nucleoli while the plasmablastic cells show eccentrically placed nuclei with coarse chromatin and small conspicuous nucleoli [5]. In the current study, a single case of PBL was notable for a predominance of centroblasts with multiple peripheral nucleoli, which is a very rare presentation of the PBLs.

In small incisional biopsies, neoplastic cells may show trabecular and single cell growth patterns and less frequently show starry-sky appearance. The diagnosis of PBL in these circumstances becomes a challenge due to the unusual morphology compounded by the limited tissue available for examination and immunohistochemical workup.

An interesting growth pattern identified in two of the present cases was the pseudo alveolar growth pattern. In addition, two more cases showed multinucleated giant cells, tumour giant cells and clear cell change. Although the presentation of pseudo alveolar growth pattern and tumour giant cells has been reported before in a pediatric case study of PBL [26], the presence of multinucleated giant cells and clear cell change is a new finding. However, pseudo alveolar growth pattern and clear cell change might be due to tissue fixation or histopathological processing artefacts.

The immunohistochemical profile of the cases showed negative CD20, immunopositivity for at least one plasma cell differentiation marker (MUM1, VS38c, and CD138) and high proliferative indices (Ki-67). One case showed aberrant T-cell antigen (CD3 and CD4) expression with negative staining for CD2D, CD5 and CD8. The phenomenon of T-cell infidelity in which PBLs, which are B-cell lymphomas, express T-cell antigens is well described [27,28].

EBER expression in the current study, which was confined to PBLs in the gastrointestinal tract, showed positivity in 53% of the cases. This is at variance with rates seen in HIV-associated oral cavity and extracavitary PBLs (60-75%) [1,8]. The results in the current study further indicate that EBV infection is not the sole driver of this neoplasm. Although the sample size in the present study was small, the findings suggest that the EBV infection rates may be lower in the gastrointestinal tract PBLs when compared to other extra-oral and oral cavity PBLs.

Only two cases had documented molecular study results in the present study. One case showed c-MYC rearrangement and the other case showed heavy chain clonality. This finding of c-MYC rearrangement in one case should be taken with caution as it may not be representative.

Diagnostic challenges for PBL may arise in the gastrointestinal tract due to its morphology, lack of expression of CD45 and pan-B cell markers and occasional T-cell infidelity. This immunophenotype can inadvertently lead to the exclusion of lymphoma from the differential diagnosis. Morphologically, the neoplasm may mimic a diffuse large B-cell lymphoma (not otherwise specified), Anaplastic lymphoma kinase (ALK) positive large B-cell lymphoma, extracavitary primary effusion lymphoma (PEL), plasmablastic myeloma, poorly differentiated carcinoma and melanoma [29].

Diffuse large B-cell lymphoma not otherwise specified (DLBCL...
NOS) may have similar morphological features to PBL but is distinguished from PBL through its expression of pan-B-cell markers (CD20, CD19, CD79a and PAX5) [30,31]. The tumour cells in DLBCL NOS may not express one or more of these pan-B-cell markers [1].

ALK-positive large B-cell lymphoma is a rare aggressive neoplasm composed of ALK-positive large monomorphic immunoblast like B-cells. The cells usually lack pan B-cell markers and express plasma cell phenotype. All the cases are also EBV and HHV8 negative [32].

Extracavitary primary effusion lymphoma (PEL) and PBL have overlapping histopathological features and immunophenotypic profile; however, the presence of HHV8 infection supports the diagnosis of extracavitary PEL [33,34]. Extracavitary PEL displays a higher expression of CD20 and CD79a when compared to PBLs. Differentiating PBL from plasmablastic myeloma may be a diagnostic challenge. The two entities may have similar morphology and immunophenotype [35]. The presence of a high proliferative index (Ki-67) and EBV infection favors PBL whilst immunopositivity for Cyclin D1, CD117 and CD56 favors a myeloma [36]. Correlation with clinical and imaging findings is crucial [30].

The combination of CD45, pan-T-cell and pan-B-cell marker immunonegativity associated with EMA immunopositivity may lead to a diagnostic pitfall for an undifferentiated carcinoma. Carcinomas will however show positivity for broad-spectrum cytokeratin and not express MUM1 or show kappa light chain restriction. Aberrant cytokeratin expression has been reported in a small subset of PBLs [37]. Aberrant CD138 staining has also been described in undifferentiated carcinomas [38]. Based on immunohistochemical profile a, melanoma can be excluded as it stains with S100 and HMB45.

Conclusion

In summary, we have reported on a case series, largest to date, of patients diagnosed with PBL in the gastrointestinal tract broadening the reported spectrum of PBL. The neoplasm shows predilection for males. Our study highlights a constellation of findings observable on morphology and the immunohistochemical profile that can be a clue to the likely diagnosis. The study also highlights some unfamiliar features that may be encountered in PBLs. The stomach, small intestine and colon being uncommon locations, a high index of suspicion is required that may be encountered in PBLs. The stomach, small intestine and morphological and the immunohistochemical profile that can be a clue to recognition of such cases of gastrointestinal tract plasmablastic lymphoma in HIV infected adults in Malawi.

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References


