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CASE REPORT

Bilateral conjunctival pediatric follicular lymphoma

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Abstract Follicular lymphoma (FL) is the most common type of B cell non-Hodgkin lymphoma in Western counties. FL is usually seen in adults and is very uncommon in pediatric population. Pediatric follicular lymphoma (PFL) is a rare indolent lymphoma characteristically seen in pediatric population, usually involving cervical lymph nodes, Waldeyer ring, and testis. They pose significant diagnostic challenges due to morphologic overlap with much more common florid follicular hyperplasia and lack of typical *t*(14;18)*IgH*/*BCL2* fusion and BCL-2 protein expression. We present the case of a 10-year-old male with bilateral conjunctival nodules, showing characteristic morphologic and immunophenotypic features of PFL with demonstrated clonal B cell immunoglobulin heavy chain gene rearrangement. Of note, the neoplastic cells expressed MUM1, MYC, and IgM, suggestive of a different disease pathogenesis from adult FL.

Keywords Pediatric · Follicular · Lymphoma · Conjunctival · Bilateral

Background

Follicular lymphoma is the one of the most common non-Hodgkin lymphomas in adults; however, it is very rare in the pediatric population. A subtype of follicular lymphoma—pediatric follicular lymphoma (PFL), is described in the World Health Organization, 2008 edition and occurs almost exclusively in children and young adults [1]. It is a distinct entity, which lacks the typical *t*(14;18)*IgH*/*BCL2* fusion and BCL-2 expression seen in usual follicular lymphoma, and shows an indolent clinical behavior despite high-grade morphologic features. They usually occur in the head and neck region, involving lymph nodes and the Waldeyer ring or sometimes extranodal sites like testis [2]. Lack of definite diagnostic criteria and differentiation from those with much more common florid follicular hyperplasia in childhood population poses significant challenges in making the accurate diagnosis. Conjunctival involvement of PFL has rarely been reported [3–5], and bilateral conjunctival presentation is even rare [6], with this being only the second reported case in literature.

Clinical history

A 10-year-old African American boy presented to the pediatric ophthalmology office with a pink lesion near the left caruncle for a few months according to the patient's mother, who had become concerned due to more recent growth. The patient reported occasional foreign body sensation and itch but denied significant pain, tearing, or discharge. He had no previous eye trauma or surgery and no personal or noteworthy family medical history. On slit-lamp microscopic examination he was found to have a 6.2 mm×4.3 mm pink mobile nodular lesion near the left caruncle with a similar-appearing 4.8×2.5 mm nodular lesion near the right caruncle (Fig. 1(A1–A2)). His visual acuity was normal, as was the remainder of his anterior and posterior segment examinations. Both lesions were completely excised under general anesthesia with 1–2 mm margins on all sides and submitted for histopathologic examination.

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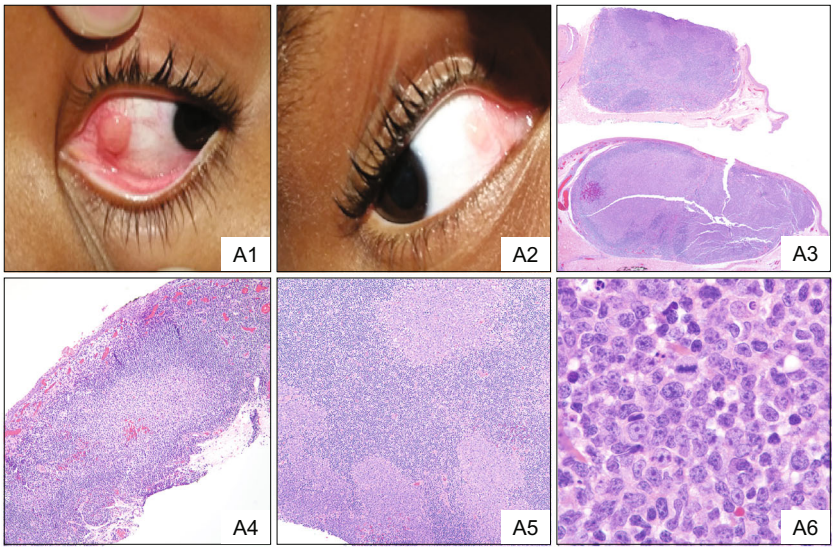

Fig. 1 Gross photographs and histologic features of conjunctival lesions: left eye (*A1*) and right eye (*A2*). Hematoxylin and eosin stained sections of the conjunctival lesions: large expanded follicles in left eye, ×2 (*A3*) and right eye, ×10 (*A4*). The left eye also shows multiple irregular smaller follicles, ×20 (*A5*). High-power view of the sheets of monomorphic large atypical cells within the follicles, ×100 (*A6*)

Materials and methods

The excisional biopsy specimens were formalin-fixed, paraffin-embedded, and stained with hematoxylin and eosin stain. Immunohistochemical stains were performed on a Bond III instrument (Leica Microsystems, Buffalo Grove, IL) and a Benchmark ULTRA system (Ventana, Tuczon, AZ), as per the manufacturer's instructions, using the following primary antibodies: CD3, CD20, CD10, BCL6, BCL2, Ki-67, CD21, MUM1, MYC, and P53. EBV encoding RNA (EBER) in situ hybridization was also performed. B cell immunoglobulin heavy chain *IgH* and kappa light chain *IgK* gene clonality assays were performed on paraffin-embedded tissue using the *IGH* + *IGK* B Cell Gene Clonality Assay (Invivoscribe, San Diego, CA) on the GeneAmp PCR System 9700 and 3500 Genetic Analyzer (Applied Biosystems/Life Technologies, Grand Island, NY). Interphase fluorescent in situ hybridization (FISH) analyses were performed on paraffin tissue using Abbott molecular probes specific for *t*(14;18)*IGH*/*BCL2* fusion, *BCL6* (3q27), and *cMYC* (8q24) genes.

Results

The conjunctival lesion in the left eye demonstrated a lymphoid proliferation with markedly expanded and distorted atypical follicles/nodules with attenuated to absent mantle zones. The neoplastic follicles contained a monomorphic population of large atypical lymphoid cells admixed with few tingible-body macrophages, numerous mitotic figures, and apoptotic debris (Fig. 1(A3–A6)). The lymphoid proliferation in the right eye was smaller and less dense than the left; however, it showed similar morphologic features, both of which very much resemble that of grade 3B type of follicular lymphoma in adult population (Fig. 1(A3–A6)). On immunohistochemical analysis of the left eye lesion, the atypical follicles express CD20, CD10, BCL6, MYC (subset, 60 %), IgM, MUM1 (subset, 50 %) and are completely negative for BCL2. Ki67 stain highlights a proliferation rate of over 90 % (Fig. 2(B1–B5)). CD21 immunohistochemical stain highlighted an expanded and distorted follicular dendritic meshwork (Fig. 2(B3)).

FISH studies for *t*(14;18)*IgH*/*BCL2* fusion, *BCL6*, and *cMYC* gene rearrangements showed no abnormalities. Molecular studies for B cell immunoglobulin heavy chain gene (*IgH*) rearrangement detected a clonal population (Fig. 2(B6)), while B cell immunoglobulin kappa light chain (*IgK*) gene clonality assay was negative.

The overall findings of the nodular growth pattern, expended and distorted follicles with CD10/BCL-6 positive large cell proliferation with high proliferative index, along with clonal *IgH* gene rearrangement were consistent with a diagnosis of PFL.

Discussion

Non-Hodgkin lymphomas in the pediatric population are usually aggressive diseases clinically like lymphoblastic lymphoma, anaplastic large cell lymphoma, Burkitt lymphoma, and diffuse large B cell lymphoma, all of which have been reported in the literature to involve the conjunctiva [7–10]. Lymphomas involving the conjunctiva and ocular adnexa in adults, however, are most commonly extranodal marginal zone lymphoma of mucosa-associated lymphoid tissue (MALT) type [10]. Conjunctival MALT lymphomas have also been rarely reported in children [11]. Follicular lymphomas are rarely reported in the conjunctiva in adults [10] and are extremely rare in the pediatric population [10]. PFL is a distinct subtype of follicular lymphoma described in the World Health Organization, 2008 edition [1], and is characteristically seen in children and young adults.

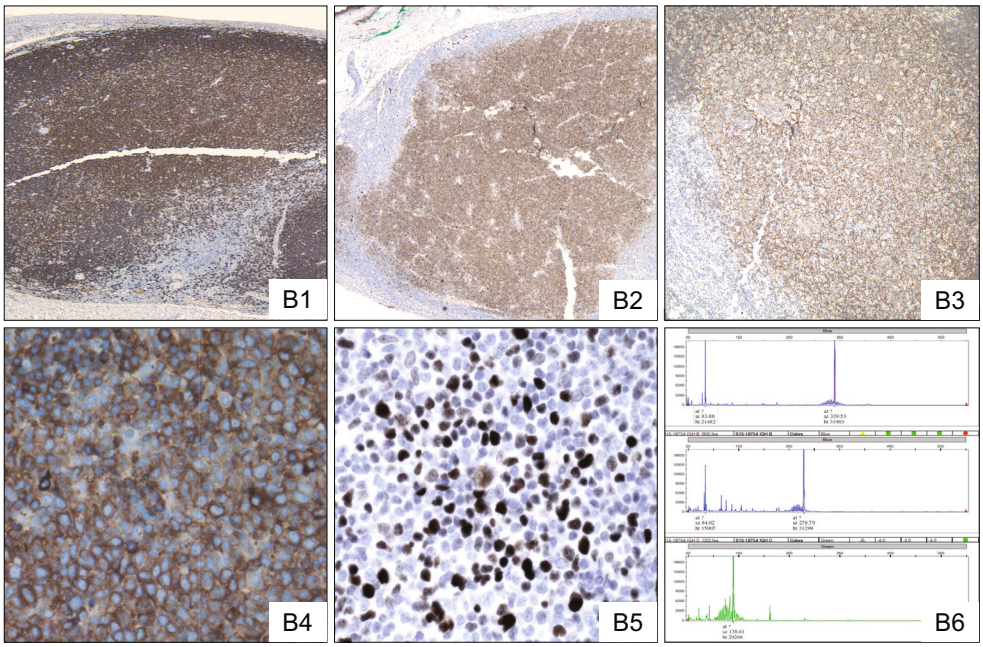

Fig. 2 Immunophenotypic characterization of the large expanded follicles: they are positive for CD20 (*B1*) and CD10 (*B2*) in an expanded and distorted follicular dendritic meshwork identified on CD21 stain (*B3*). The neoplastic cells are also positive for IgM (*B4*) and MYC protein (*B5*). Molecular studies detected a clonal IgH gene rearrangement (*B6*)

PFL are usually seen in male patients and most commonly involves the head and neck region—cervical lymph nodes, and Waldeyer ring, other peripheral lymph nodes and has also been well reported in the testis [2]. They typically lack *t*(14;18)*IgH*/*BCL2* fusion characteristically seen in majority of adult follicular lymphomas, and often do not express BCL2 protein. Morphologically, they show large, expanded follicles that are irregular or serpiginous in shape. Majority of them have high-grade cytologic features, equivalent of grade 3 in adult follicular lymphoma based on proportion of centroblast. Children with PFL usually have localized or early stage disease and have an indolent clinical behavior. It should be noted that adult type follicular lymphomas with *t*(14;18)*IgH*/*BCL2* fusion and BCL-2 protein expression may rarely occur in the pediatric population, which are usually low grade (grade 1–2) [2]. These should not be designated as PFL due to differing clinical behavior and disease pathogenesis.

The pathogenesis of PFL is currently unclear. Over 90 % of reported cases showed clonal *IgH* gene rearrangement and in cases lacking the former, showed clonal *IgK* gene rearrangement [2]. A subset of cases expressed MUM-1 protein, including few showing *IRF4* gene rearrangement and *IgH*/*IRF4* fusion [2]. Interestingly, majority of the MUM1 positive cases involved the Waldeyer ring [2]. MYC protein expression has not been previously studied in PFL, and expression of MYC protein in our case is a novel finding, suggesting the MYC pathway playing a possible role. Interestingly, although MYC protein expression in adult follicular lymphoma is thought to show an aggressive clinical course [12], PFL is likely an exception. We also identified strong expression of IgM in the neoplastic cells; a finding also seen in a prior case report [13] and reported previously in subset of adult follicular lymphomas [14], which suggests possible defective class switch recombination, characteristics similar to IgM+ memory B cells [15] or malignant transformation occurring in the naïve B cell stage. This hypothesis needs to be tested in a larger series of cases with additional molecular studies.

The most significant diagnostic challenge in PFL is differentiating it from florid follicular hyperplasia, especially in areas with prominent lymphoid tissue like Waldeyer ring, commonly occurring in childhood population. Rare cases of follicular hyperplasia have also been reported to show presence of clonal CD10-positive cells [16]. The presence of numerous tingible-body macrophages, lack of typical (14;18) *IgH*/*BCL2* fusion, and lack of BCL2 protein expression adds to the diagnostic dilemma. The presence of architectural atypia with expanded/serpiginous follicles and cytologic atypia with increased numbers of large atypical monomorphic cells should raise suspicion of PFL. High clinical and morphologic suspicion should trigger ancillary studies. Immunohistochemical findings like MUM1, MYC, and IgM positivity are supportive of the diagnosis. B cell gene rearrangement studies are extremely useful as over 90 % cases show a clonal gene rearrangement [2].

MALT lymphomas, being the most common lymphoma in the ocular adnexa, is a differential diagnosis to consider, especially in cases with a nodular architectural pattern due to marked follicular colonization and expression of IgM and MUM1. MALT lymphomas usually are composed of small-sized cells, or an admixture of small and larger cells, with a CD10 negative, BCL2 positive immunophenotype. CD10 expression has rarely been reported in low-grade MALT lymphomas [17]. On the other hand, PFLs are usually of high-grade

morphology with increased larger-sized cells (grade 3) and are usually CD10 positive, BCL2 negative in majority of cases. FISH studies for *MALT1* gene rearrangement may also be helpful and considered in difficult cases.

Translocations activating *IRF4* oncogene like *IGH*/*IRF4* fusion was recently identified as a distinct molecular subgroup of germinal center derived B cell lymphomas including diffuse large B cell lymphomas and grade 3 follicular lymphomas [18]. These are seen predominantly in the head and neck region of children and young adults, show *IRF4* and *BCL6* gene rearrangements, express MUM1 and BCL6 proteins, and lack *t*(14;18) *IGH*/*BCL2* fusion. Given that majority of reported extranodal PFLs in the head and neck region show *IRF4* aberrations and MUM1 expression [2], it is possible that this subset of PFL (including our case) and *IGH*/*IRF4* positive follicular and diffuse large B cell lymphomas represent lesions in the same disease spectrum. Nodal and testicular PFLs on the other hand are less often reported to show IRF4 and *BCL6* gene rearrangements, and may be biologically distinct [19].

PFL shows an indolent clinical behavior, in spite of the morphologic features equivalent to grade 3 adult follicular lymphoma. Even in the presence of concurrent diffuse large B cell lymphoma, they show very good clinical outcomes with therapy [20]. There is no current consensus on the optimal clinical management of PFL. Majority of localized lesions are treated by surgical excision with close follow up. However, Rituxan with or without adjuvant radiation is used for multifocal/disseminated disease. A multi-agent systemic chemotherapy like R-CHOP is considered for refractory or relapsed extensive disease or with the presence of concurrent diffuse large B cell lymphoma. The overall survival is excellent with very low risk of recurrence [20–22].

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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