

IgG4-Related Systemic Sclerosing Disease of the Ocular Adnexa

A Potential Mimic of Ocular Lymphoma

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Upon completion of this activity you will be able to:

- describe the histologic appearance of IgG4-related systemic sclerosing disease.
- list the most common hematolymphoid malignancies arising in the orbit and ocular adnexa.
- examine biopsy specimens in which IgG4-related sclerosing disease is suspected and interpret the results of IgG and IgG4 immunohistochemical stains.

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Abstract

IgG4-related sclerosing disease has been described in the orbit and ocular adnexa. Of 164 biopsies of the ocular region for suspected lymphoma, we identified 6 cases of IgG4 disease, 4 of which were previously unrecognized. All 6 cases demonstrated increased plasma cells in a background of sclerosis and increased absolute numbers of IgG4-expressing cells. Our results confirm the difficulty in diagnosing IgG4-related sclerosing disease in the ocular region. Based on the findings, we suggest that specimens from biopsies of the eye and ocular adnexa for which a definitive diagnosis of lymphoma is not established undergo further workup for IgG and IgG4, particularly if increased plasma cells and sclerosis are present. When IgG4-expressing plasma cells account for greater than 50% of IgG-expressing plasma cells, a diagnosis of IgG4 disease should be considered. Timely recognition would benefit patients by allowing appropriate management with corticosteroid therapy and avoiding more aggressive or unnecessary therapeutic options.

The diagnosis and treatment of ocular disorders poses significant challenges. Aggressive ocular malignancies such as uveal melanomas, metastatic carcinomas, and intraocular large cell lymphomas can be life threatening and require the coordination of multidisciplinary teams for optimal management. Many conditions that involve the eye and ocular adnexa can also result in diplopia or vision loss from the disease process itself or from its treatment and may result in significant cosmetic disfigurement. The recognition and refinement of histopathologic criteria are of benefit for accurate diagnoses of disorders affecting the ocular adnexa.

Lymphomas comprise a heterogeneous group of malignancies in the orbital region and account for 7% to 8% of extranodal lymphomas.¹ Of these, extranodal marginal zone lymphoma (EMZL) is the most frequent.²⁻⁵ Although the criteria for the diagnosis of ocular lymphomas follow the basic tenets set forth in the 2008 World Health Organization lymphoma classification,⁶ their location necessitates a more thorough understanding of the overlap of these lymphomas with reactive entities and other morphologic mimics in their differential diagnosis.

One such recently characterized morphologic mimic is IgG4-related systemic sclerosing disease (or IgG4 disease). This immune-mediated systemic syndrome is characterized by multiorgan involvement and mass-forming lesions.⁷ It is typified by the infiltration of IgG4-expressing plasma cells associated with inflammatory T lymphocytes in various organs with an accompanying elevation in the levels of serum IgG4 and an acute phase response.⁸ Though first described in the pancreas in autoimmune pancreatitis,⁹

IgG4-related disease has now been identified in multiple other organ systems,⁸ including the eye and ocular adnexa.¹⁰⁻¹² In the orbit, IgG4 disease has been found to be associated with inflammatory pseudotumors,^{7,11,13} dacryoadenitis,^{14,15} xanthogranuloma,¹² and lymphadenopathy.^{16,17} A case of localized orbital amyloidosis composed of IgG4 heavy chain has also been described.¹⁸ IgG4 is also reportedly associated with EMZL.¹⁰ IgG4 disease shows an excellent response to glucocorticoid therapy, and it is therefore important to recognize and separate IgG4 disease from lymphomas and other entities involving the ocular adnexa.

We undertook this study to investigate the clinical manifestations, frequency, and histologic and immunophenotypic findings in cases of IgG4 disease diagnosed in specimens obtained from the ocular region for suspected lymphoma. There were 164 cases studied by immunohistochemical analysis for IgG and IgG4. Among these, we diagnosed 6 cases of IgG4 disease whose clinical manifestations and treatment course are described in detail. Of the 6 patients, 3 had systemic manifestation of disease resulting in biopsy or surgery outside the eye and orbit. We also reviewed this material. In 2 of these 3 cases, material was available for immunohistochemical studies, and we performed IgG and IgG4 stains. The diagnostic difficulty in these cases and possible criteria for the diagnosis of IgG4 disease in the ocular adnexal region are discussed.

Materials and Methods

Case Selection

A total of 164 biopsy specimens from the Department of Pathology tissue archive at Stanford University Medical Center, Stanford, CA, between 1991 and 2011, comprise this study. Institutional review board approval was obtained, and procedures were followed in accord with the ethical standards established by the Helsinki Declaration of 1975. Inclusion criteria for the study consisted of availability of tissue on which the initial diagnosis was made and availability of paraffin blocks or sufficient numbers of unstained slides to perform additional immunohistologic stains. Of 164 cases, 63 were from biopsies performed at Stanford, whereas 101 were consultation cases referred from outside institutions. In all cases, the biopsies were obtained because of clinical suspicion of lymphoma.

Morphologic and immunohistologic findings were reviewed by 2 pathologists (S.F.Y. and Y.N.) and selectively reviewed by 2 others (J.R.K. and R.A.W.). Immunostains performed at the time of initial diagnoses included the following stains, although not all stains were performed on every case: CD20, CD3, CD5, CD10, CD23, CD21,

BCL1, BCL2, BCL6, κ , λ , Ki-67, CD79a, CD4, CD8, EBV (Epstein-Barr virus), ALK1 (anaplastic lymphoma kinase), and CD163. Immunohistochemical staining for IgG and IgG4 was performed in all cases. In addition, the results of flow cytometry and molecular and cytogenetic studies were also reviewed whenever they had been performed in the diagnostic workup. The diagnoses were confirmed according to the 2008 World Health Organization criteria. Clinical parameters, such as systemic disease and the presence of circulating paraproteins and organomegaly, were also taken into consideration in arriving at the final pathology diagnosis. When there was insufficient support for lymphoma, a designation of “atypical lymphoid infiltrate” was made if morphologic or ancillary tests showed abnormal findings and the suspicion of lymphoma could not be ruled out. A designation of “reactive” was made if morphologic findings and the findings of ancillary studies did not support involvement by lymphoma.

Immunohistochemical Studies

Immunohistologic staining was performed on 0.4-mm thick formalin-fixed, paraffin-embedded whole tissue sections. Primary antibodies directed against IgG4 (clone HP6025, dilution 1:800; Invitrogen, Carlsbad, CA) and IgG (clone RWP49, dilution 1:400; Leica, Newcastle upon Tyne, England) were used. An automated stainer (BOND-MAX, Leica Microsystems, Buffalo Grove, IL) was used for immunohistochemical studies. For IgG4, DAKO citrate retrieval with EnVision+ (DAKO, Carpinteria, CA) was used, and for IgG, EDTA ER2 (Leica Microsystems) for 20 minutes was used.

In the cases of IgG4 disease, stained cells were counted in 3 high-power fields from areas with the highest density of positive cells using a 40 $\times$ objective and 10 $\times$ eyepiece of an Olympus BX41 microscope (Olympus, Center Valley, PA).^{10,12,15,16} The proportion of IgG4+ to IgG+ cells was considered significantly elevated if more than 30% of IgG+ also showed IgG4 immunoreactivity.

Results

Of 164 cases, 67 (40.9%) were in men and 97 (59.1%) in women. The following anatomic sites were represented: orbit, 60 (36.6%); conjunctiva, 51 (31.1%); eyelid, 22 (13.4%); lacrimal apparatus, 23 (14.0%); canthus, 3 (1.8%); eyebrow, 2 (1.2%); and fornix, limbus, and subretina, 1 (0.6%) each. The frequency of each lymphoma type is shown in **Table 1**. Of the cases, 141 were diagnosed as lymphoma. EMZL was the most frequently diagnosed lymphoma subtype and constituted 81 (57.4%) of the 141 lymphomas, followed by follicular lymphoma (FL; 22 [15.6%])

and mantle cell lymphoma (10 [7.1%]). In addition, 6 cases (4.3%) were diagnosed as diffuse large B-cell lymphoma. One case showed features intermediate between diffuse large B-cell lymphoma and Burkitt lymphoma. T-cell lymphomas were infrequent and constituted 3 (2.1%) of the 141 lymphomas in this study cohort. In 1 case (<0.7%), a composite lymphoma with FL and lymphoplasmacytic lymphoma was represented. Of 164 cases, 5 (3.0%) were diagnosed as atypical lymphoid infiltrate and 18 (11.0%) were diagnosed as reactive lesions.

Among the MZL cases, CD43 staining was performed at the time of diagnosis in 61 cases; it was positive in 20 and lacking in 41. Among the FL cases, a germinal center marker (CD10 with or without BCL6) was done at the time of diagnosis for 7 of the cases. In other cases, BCL2 staining was performed to help differentiate FL from reactive conditions.

We identified 6 cases that showed increased total numbers and proportions of IgG4-expressing plasma cells; clinical data, diagnoses, and immunohistologic findings are summarized in **Table 2**. Four of these cases were initially diagnosed as reactive lymphoid hyperplasia. In the remaining 2 cases, a diagnosis of EMZL was favored. In all cases, histologic sections showed inflammatory infiltrates composed of abundant plasma cells and small, reactive-appearing lymphoid cells occasionally arranged in follicles. These infiltrates were set in sclerotic, densely collagenized stroma. In all 6 cases, immunohistochemical stains for IgG and IgG4 demonstrated markedly increased numbers of IgG4-expressing plasma cells as a proportion of IgG-expressing cells and in absolute numbers. There was no evidence of light chain restriction by in situ hybridization and/or flow cytometry. The clinicopathologic features of these cases are presented in detail and illustrated in the following sections.

Case 1

A 74-year-old woman had a 3-year history of irritation and injection of her left eye. Clinically, the patient had an elevated, erythematous, gelatinous-appearing vascular limbal mass that was of concern for squamous cell carcinoma; the clinical differential diagnosis also included amelanotic melanoma and papilloma. The original biopsy findings were interpreted as reactive plasmacytoid hyperplasia based on the polyclonal infiltrate. A biopsy of the left limbal conjunctiva showed edematous, reactive-appearing conjunctival epithelium overlying a dense collection of plasma cells and rare reactive lymphoid follicles. The plasma cells were associated with a fine collagenous network. In situ hybridization for κ and λ showed no evidence of light chain restriction; however, IgG and IgG4 immunohistochemical stains showed that IgG4 stained greater than 50% of the IgG-expressing plasma cells **Image 1**.

Table 1
Frequency and Subtype of 141 Lymphomas of the Ocular Adnexa

Diagnosis	Frequency (%)
Extranodal marginal zone lymphoma	81 (57.4)
Follicular lymphoma	22 (15.6)
Small lymphocytic lymphoma	7 (5.0)
Mantle cell lymphoma	10 (7.1)
Diffuse large B-cell lymphoma	6 (4.3)*
Peripheral T-cell lymphoma	3 (2.1)
Lymphoplasmacytic lymphoma	2 (1.4)
Plasmacytoma	1 (0.7)
Low grade B-cell lymphoma, not further classified	8 (5.7)
Composite lymphoma	1 (0.7)

* Includes 1 case of large B-cell lymphoma with features intermediate between diffuse large B-cell lymphoma and Burkitt lymphoma.

Case 2

A 50-year-old woman was found to have a subretinal mass clinically thought to represent granulomatous inflammation (the patient had a positive tuberculin [purified protein derivative, or PPD] test result) with associated retinal detachment and subretinal fluid collection. A blind painful eye developed, and the patient elected to undergo enucleation with resection of the orbital contents. Histologic sections of the enucleation specimen, originally interpreted as reactive lymphoid hyperplasia, showed orbital fat involved by a dense, sclerotic, collagenous mass featuring abundant plasma cells. Rare lymphoid follicles were present. This process obliterated the globe and distorted the sclera, retina, and lens. A CD20 immunostain highlighted B cells, including those within follicles. A CD3 stain highlighted T cells arranged around the follicles, and CD21 highlighted follicular dendritic cells within follicles. CD138 showed abundant plasma cells throughout the sclerotic collagenous stroma and within the distorted globe. IgG stained the majority of these plasma cells, of which more than 75% were positive for IgG4 **Image 2**.

Case 3

A 37-year-old man with bilateral submandibular gland enlargement underwent resection of the submandibular glands, the histologic features of which were interpreted as a benign lymphoepithelial lesion. One year later, he had right eye redness and tearing, and these symptoms persisted intermittently for 4 years. The patient was diagnosed clinically as having conjunctivitis and blepharitis and was given topical tobramycin therapy. A trial of ciprofloxacin was discontinued, and a biopsy of the orbit and lacrimal gland was undertaken owing to the persistence of symptoms. The biopsy specimen, which was initially interpreted as reactive lymphoid hyperplasia, showed a collagenous mass containing poorly preserved lymphoid follicles and numerous plasma cells. CD20 highlighted the B-cell follicles, and

Table 2
Characteristic Features of Cases With IgG4 Disease Involving the Ocular Adnexa

Case No./ Sex/Age (y)	Clinical History	Morphologic Features	Original Diagnosis
1/F/74	Limbal mass	Sheets of plasma cells below conjunctiva with subadjacent areas of lymphocytes	Reactive plasmacytoid hyperplasia of the conjunctiva
2/F/50	Subretinal mass	Sheets of lymphocytes; plasma cells with fibrous, collagenous proliferation and bony metaplasia	Benign lymphocytic hyperplasia with extensive fibrosis
3/M/37	Lacrimal gland mass	Sheets of atypical lymphoid cells, plasma cells, and sclerosis	Low grade B-cell lymphoma, most likely MZL
4/F/72	History of Sjögren syndrome and lacrimal gland enlargement	Diffuse proliferation of small lymphoid cells with monocytoid morphologic features around hyalinized blood vessels	Atypical lymphoid infiltrate, favor MZL
5/F/43	9-y history of eyelid swelling and diffuse infiltration of preseptal tissues and lacrimal glands	Sheets of lymphocytes arranged in follicles with extrafollicular plasmacytosis in a sclerotic, collagenous background	Reactive lymphoid hyperplasia
6/M/60	Bilateral orbital swelling and periocular pain	Sheets of lymphocytes arranged in follicles infiltrating the rectus skeletal muscle with associated plasmacytosis set in a sclerotic collagenous background	Reactive lymphoid hyperplasia

AFB, acid-fast bacillus; GMS, Gomori methenamine silver; LCA, leukocyte common antigen; MZL, marginal zone lymphoma.
* Values are given as number/total (percentage).

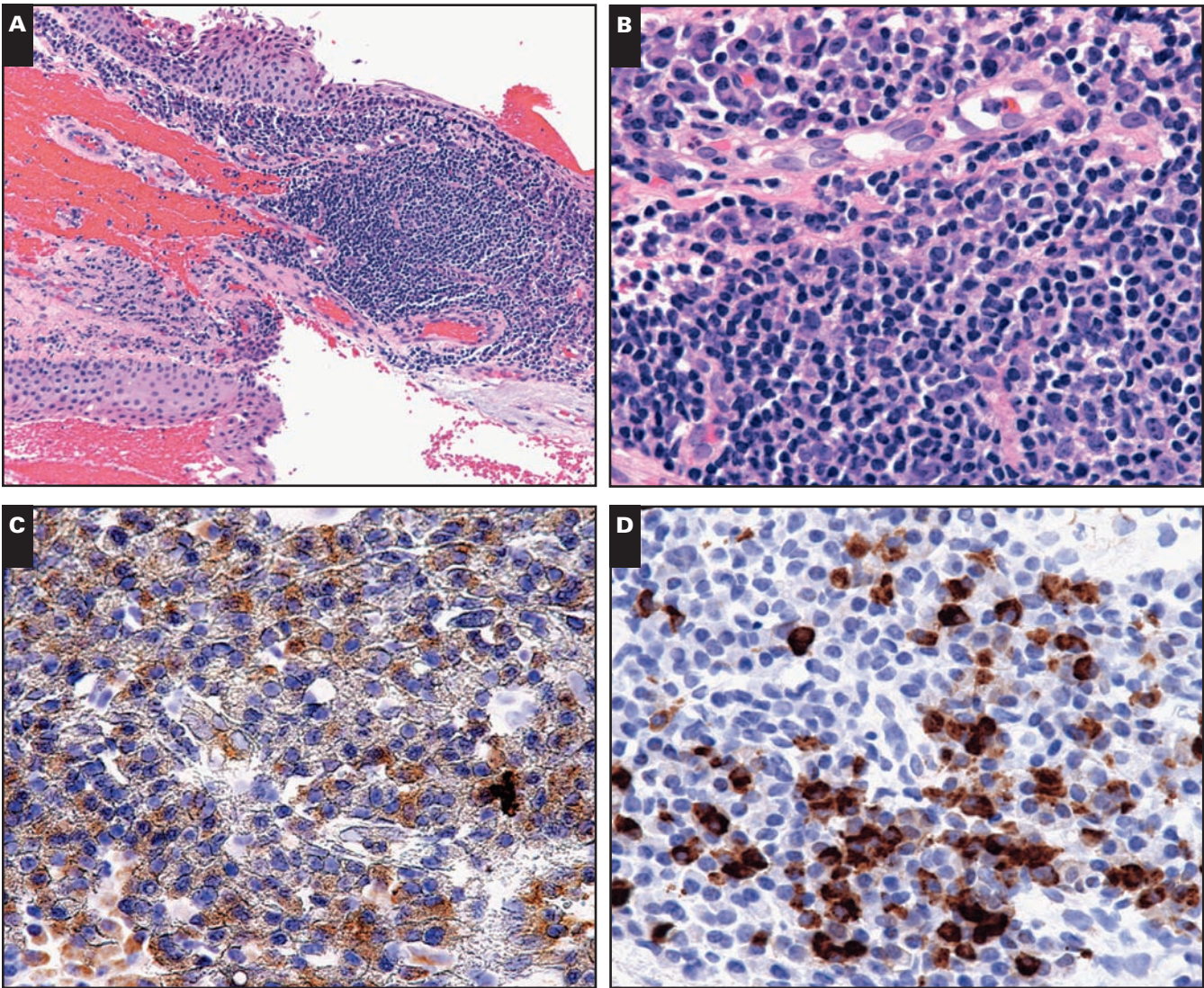


Image 1 **A**, Conjunctival mucosa with a lymphoid infiltrate with follicle formation (H&E, ×40). **B**, A lymphoplasmacytic infiltrate set in a background of wispy collagenous fibrosis (H&E, ×100). **C** and **D**, Immunohistochemical stains show increased absolute and relative numbers of IgG4-expressing plasma cells (>50%) (**C**, IgG, ×100; **D**, IgG4, ×100).

Ancillary Studies	IgG4/IgG*
κ and λ polyclonal	532/650 (81.8)
CD20, CD3, CD138 not consistent with lymphoma; AFB, GMS stains negative	542/685 (79.1)
CD20+, CD3-, CD5-, CD10-, CD21-, CD43-, κ + and λ + subset; Ki-67, 10%-20%	750/750 (100.0)
LCA+, CD20+, CD3-, CD43-, CD138-, polytypic κ and λ , negative immunoglobulin gene rearrangement	630/830 (75.9)
CD79a+, CD20+, CD3-, CD21-, no κ and λ monotypia by in situ hybridization, CD138+ (in plasma cells), CD30+(scattered)	690/717 (96.2)
No light chain-restricted population by flow cytometry	1,052/1,074 (98.0)

CD5, CD3, and CD43 stained numerous admixed T cells. In situ hybridization for κ and λ showed numerous plasma cells without evidence of light chain restriction.

Because the overall clinical impression was that of EMZL, the patient was initially treated with chlorambucil and prednisone, followed by 4 treatments of weekly rituximab (anti-CD20 monoclonal antibody). The patient's symptoms subsided for approximately 1 year. In early 2007, the patient was clinically diagnosed with recurrent disease on the basis of pruritus and periorbital and perimandibular swelling and treated with 6 cycles of R-CHOP (rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisone), which resulted in complete clinical remission. Mediastinal and

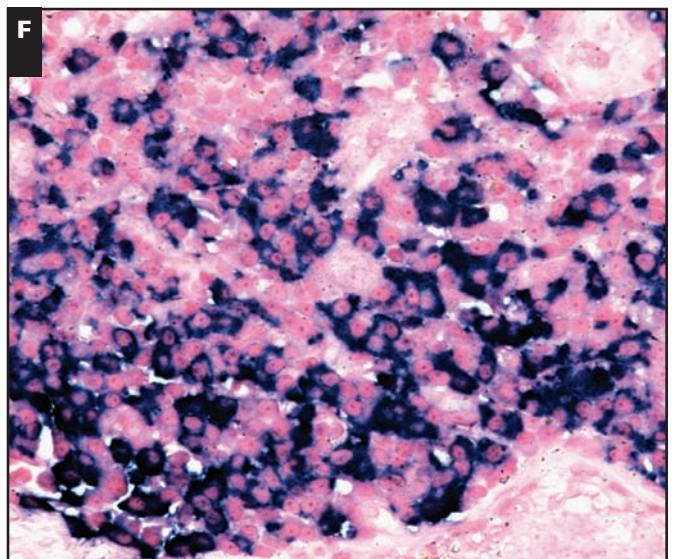
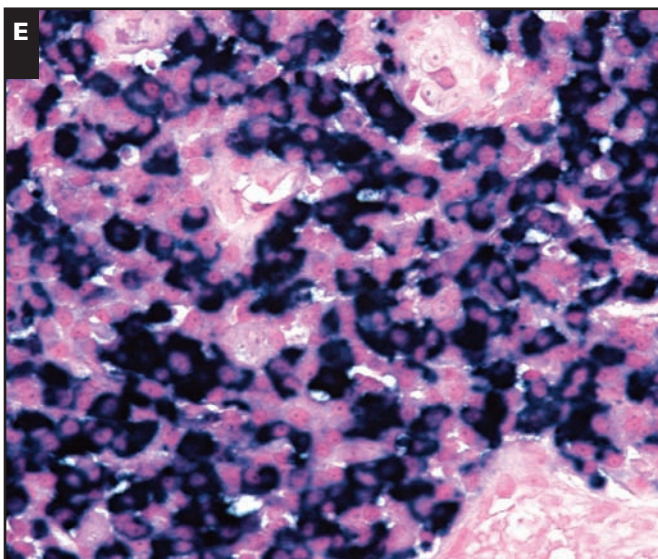
abdominal lymphadenopathy developed 1.5 years later, and the patient again underwent 6 cycles of R-CHOP.

Review of the patient's original submandibular gland biopsy showed salivary gland tissue focally effaced by bands of dense collagenous sclerosis with prominent lymphoid follicles and abundant plasma cells. IgG and IgG4 immunostains showed that the vast majority of plasma cells expressed IgG, and greater than 90% of those cells expressed IgG4. At the time of submission of this manuscript, the patient had no clinical evidence of IgG4 disease or EMZL. Review of the patient's submandibular gland revealed atrophic submandibular gland tissue featuring prominent sclerosis with a brisk lymphoplasmacytic infiltrate with follicle formation. As in the lacrimal gland specimen, IgG and IgG4 immunostains showed that greater than 90% of plasma cells expressed IgG4

■Image 3■.

Case 4

We reviewed a case in consultation from a 72-year-old woman with a reported history of Sjögren syndrome (not confirmed at the time of consultation) with lacrimal gland enlargement who underwent subsequent magnetic resonance imaging of the brain and orbit, which showed an expansile soft tissue mass at the periphery of the orbits. A biopsy of the right orbital mass showed a proliferation of small, reactive-appearing lymphocytes and plasma cells set in a collagenous stroma. A CD79a immunostain showed an abundance of B cells with no definite follicular architecture. Abundant CD138-expressing plasma cells were also seen. IgG and IgG4 immunohistochemical stains showed that approximately 75% of these IgG-expressing plasma cells also expressed IgG4 ■Image 4■.



E and **F**, In situ hybridization shows no evidence of light chain restriction (**E**, κ , $\times 100$; **F**, λ , $\times 100$).

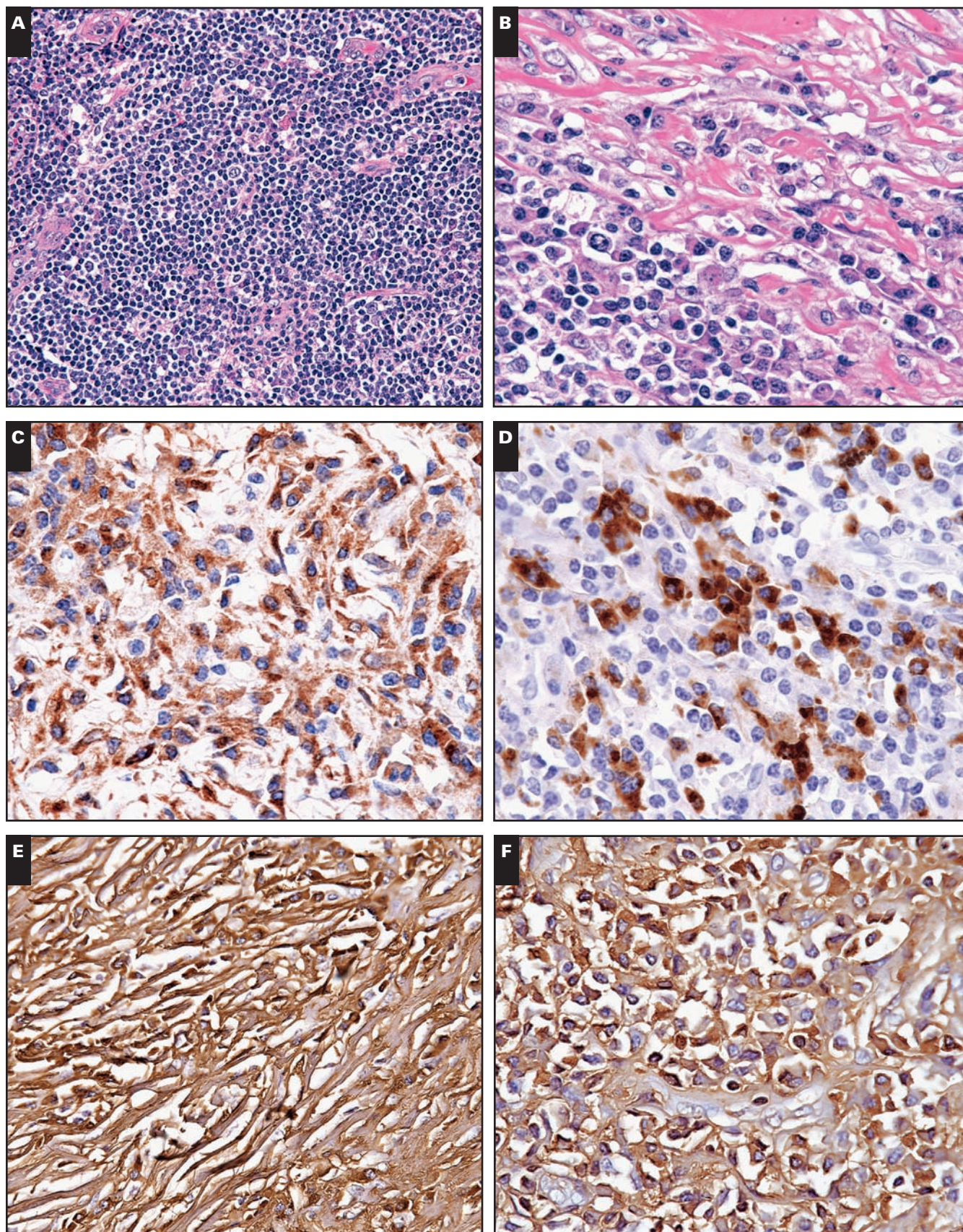


Image 2 **A**, Orbital soft tissue with a lymphoplasmacytic infiltrate (H&E, $\times 100$). **B**, Dense collagenous fibrosis (H&E, $\times 400$). **C** and **D**, Immunohistochemical stains show increased absolute and relative numbers of IgG4-expressing plasma cells ($>50\%$) (**C**, IgG, $\times 400$; **D**, IgG4, $\times 400$). **E** and **F**, Immunohistochemical staining shows no evidence of light chain restriction (**E**, κ , $\times 400$; **F**, λ , $\times 400$).

Case 5

A 43-year-old woman with a several-year history of eyelid swelling was initially diagnosed as having an allergic disease of unspecified etiology. The patient was initially treated with systemic corticosteroids, resulting in symptomatic improvement. After the corticosteroid course ended, the swelling worsened, eventually resulting in restrictive strabismus and bilateral proptosis. Imaging showed diffuse infiltration of the extraocular muscles, preseptal tissues, and lacrimal glands. A conjunctival biopsy was interpreted as lymphoid hyperplasia. A subsequent systemic workup revealed a positron emission tomography–positive mediastinal mass. This mass was biopsied and evaluated by

fine-needle aspiration and flow cytometry and interpreted as reactive follicular hyperplasia. IgG and Ig4 staining was not performed.

The patient underwent an additional biopsy of the orbital mass, and the findings were again interpreted as reactive follicular hyperplasia. Sections of the latter biopsy showed orbital fat replaced by dense collagenous sclerosis containing abundant plasma cells and scattered reactive-appearing lymphoid follicles. Numerous eosinophils were also present in the sclerotic, collagen-rich areas. Flow cytometry was performed and again showed no evidence of a clonal B-cell or plasma cell population. An immunohistochemical workup showed no distinct immunoarchitectural abnormality or

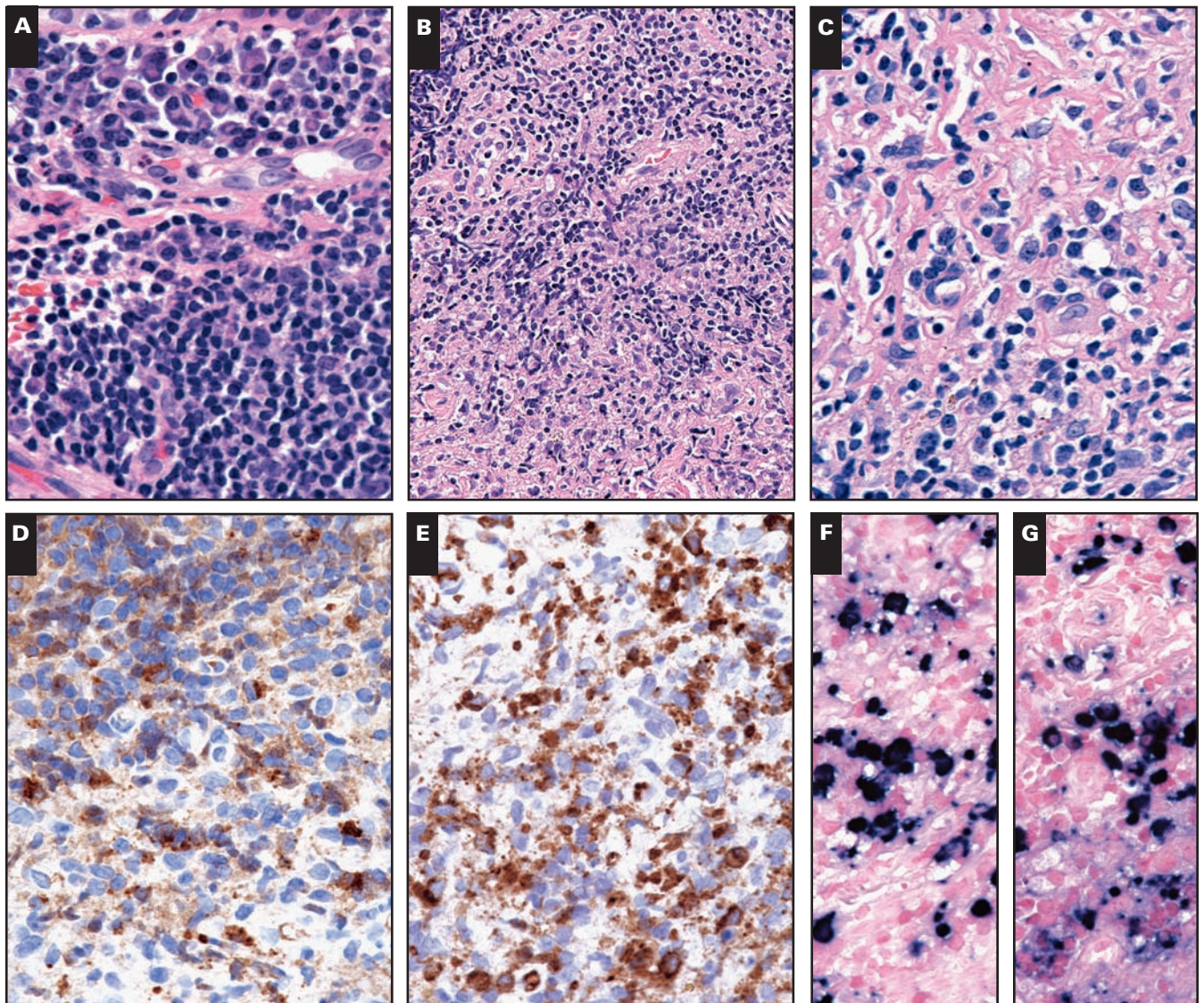


Image 3 **A**, Staining of orbital tissue shows a lymphoplasmacytic infiltrate (H&E, $\times 100$). **B** and **C**, Collagenous fibrosis with abundant plasma cells (**B**, H&E, $\times 40$; **C**, H&E, $\times 400$). **D** and **E**, Immunohistochemical stains show increased absolute and relative numbers of IgG4-expressing plasma cells ($>50\%$) (**D**, IgG, $\times 100$; **E**, IgG4, $\times 100$). **F** and **G**, In situ hybridization shows no evidence of light chain restriction (**F**, κ , $\times 400$; **G**, λ , $\times 400$).

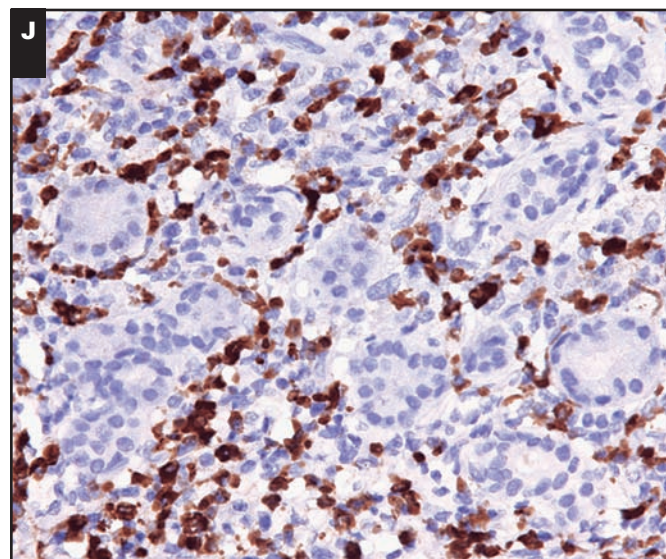
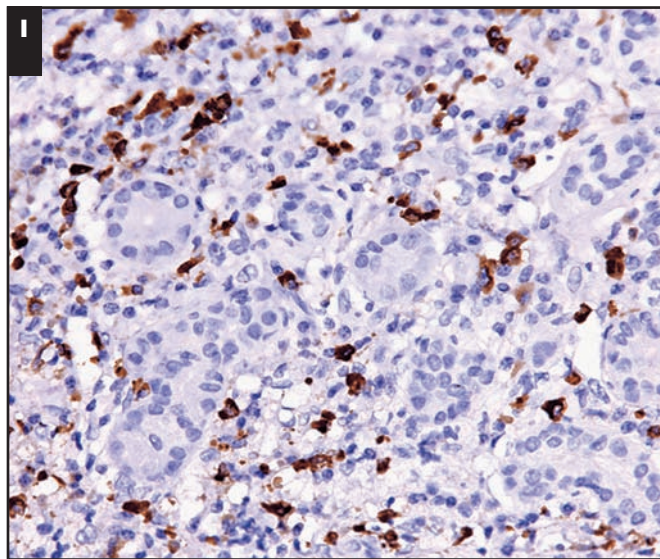
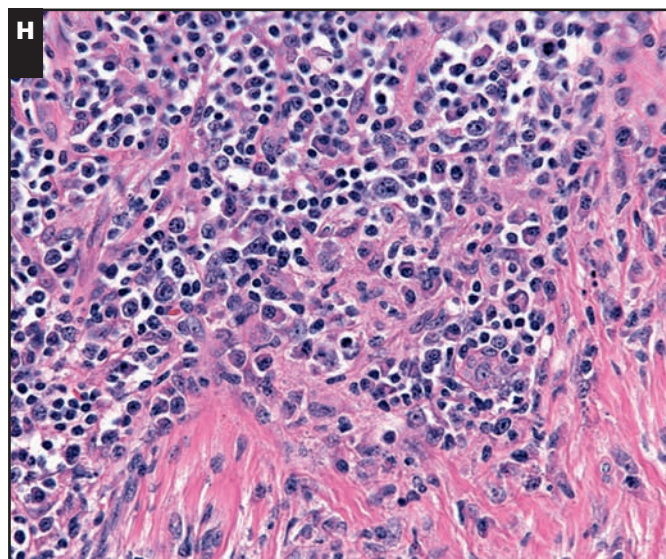
clonal B cells or plasma cells, although an abundance of CD138+ plasma cells was present in the areas of collagenous sclerosis. The patient was treated clinically as having a low-grade B-cell lymphoma and received 20 Gy to affected orbital structures, with partial response. A workup for IgG and IgG4 was undertaken and showed that nearly all plasma cells expressed both immunoglobulins ■Image 5■.

Case 6

A 60-year-old man had a 1-year history of bilateral orbital swelling and associated periocular pain. Initial treatment with corticosteroids resulted in marked clinical improvement; however, a computed tomography scan showed multiple enhancing lesions of the bilateral optic nerve sheaths associated with proptosis. Magnetic resonance imaging also showed enlargement of the extraocular

muscles. The findings were clinically “suspicious” for lymphoma, and the patient underwent biopsy of his lateral rectus muscle. Sections showed skeletal muscle diffusely expanded and infiltrated by an inflammatory infiltrate composed of reactive-appearing lymphoid follicles and abundant plasma cells. The plasma cell-rich areas showed prominent collagenous fibrosis. Flow cytometry showed no clonal B-cell or plasma cell population. The findings were originally interpreted as reactive lymphoid hyperplasia. An immunohistochemical workup for IgG and IgG4 showed that almost all plasma cells expressed IgG, and greater than 90% of them expressed IgG4. The patient was subsequently treated with bilateral orbital radiation therapy to a dose of 25.2 Gy.

A follow-up computed tomography scan of the chest showed lymphadenopathy involving the mediastinum, hilum, and axilla. The patient underwent axillary node



■Image 3■ (cont) **H**, Submandibular gland with dense lymphoplasmacytic inflammation and collagenous fibrosis (H&E, ×40). **I** and **J**, Immunohistochemical studies of submandibular gland show increased absolute and relative numbers of IgG4-expressing plasma cells (>90%) (**I**, IgG, ×400; **J**, IgG4, ×400).

biopsy, which showed reactive-appearing lymphoid hyperplasia with follicles of varying size with preserved germinal center and mantle zone architecture with tingible body macrophages. Subsets of the follicles appeared atrophic with concentrically arranged mantle zone cells imparting an “onion-skin” pattern. In addition, interfollicular areas were expanded, and there was moderate hyalinization of vessels. The findings were originally interpreted as reactive lymphoid hyperplasia with Castleman-like features. IgG and IgG4 immunohistochemical stains were subsequently performed, which showed a marked increase in the absolute and relative number (>80%) of IgG4-expressing plasma cells in the lymph node **Image 6**.

Discussion

Non-Hodgkin lymphomas comprise a substantial proportion of neoplasms that involve the ocular region, and, therefore, surgical pathologists not infrequently encounter specimens from biopsies of the eye and ocular adnexa for suspected lymphoma. Atypical lymphoid infiltrates of the ocular region raise a broad differential diagnosis that includes acute and chronic inflammation and infectious causes. Inflammatory and infectious factors have indeed been associated with the most common subtype of low-grade B-cell lymphoma that affects the ocular adnexa, namely, EMZL.¹⁹⁻²¹

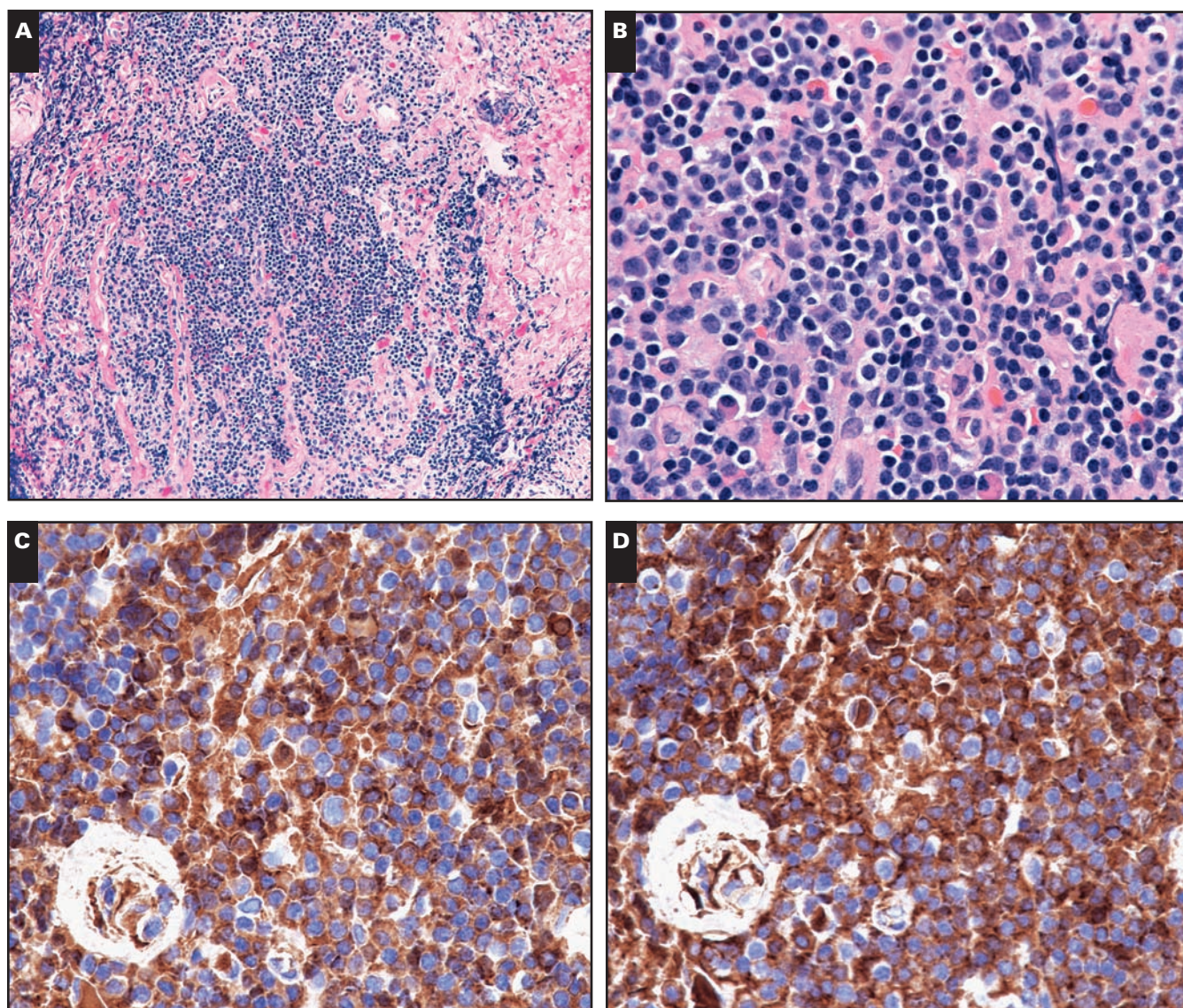


Image 4 **A** and **B**, Orbital tissue with plasma cell–rich infiltrate with admixed bands of collagenous sclerosis (**A**, H&E, $\times 40$; **B**, H&E, $\times 400$). **C** and **D**, Immunohistochemical staining shows that >90% of IgG-expressing plasma cells also show IgG4 expression (**C**, IgG, $\times 400$; **D**, IgG4, $\times 400$).

Given the many complexities of evaluating atypical lymphoid infiltrates in the ocular region and the increasing awareness that IgG4 disease is likely underrecognized, we carried out a survey of 164 biopsies of the eye and ocular adnexa done for suspected lymphoma. We identified 6 cases of IgG4-related cases involving the ocular adnexa. Of the cases, 5 manifested as orbital masses. It is interesting that in our cohort of 164 cases, 4 (22%) of 18 initially diagnosed as reactive lymphoid hyperplasia were, in fact, found to be IgG4 disease by further review and immunohistologic workup for IgG and IgG4. Of the 63 cases from Stanford, 5 were received as surgical specimens, reflecting an in-house incidence of 8%. This finding further underscores our initial suspicion that IgG4 disease of the ocular adnexa is underrecognized.

Of our cases, 3 were thought to represent low-grade B-cell lymphoma. In 2 cases, a diagnosis of EMZL was favored, although there was no evidence of B-cell light chain restriction in any of our cases of IgG4-related disease. Of the cases for which we had clinical follow-up, 3 of the patients received chemotherapy and/or radiotherapy for presumed lymphoma. In addition to orbital pathology, all 3 of these patients also had systemic symptoms, including mediastinal or other lymphadenopathy. In all 3 of these cases, material from outside of the eye and orbit was also biopsied and interpreted as reactive lymphoid hyperplasia (salivary gland in case 3, mediastinal lymph node in case 5, and axillary lymph node in case 6). In case 6, the original interpretation noted Castleman-like features,

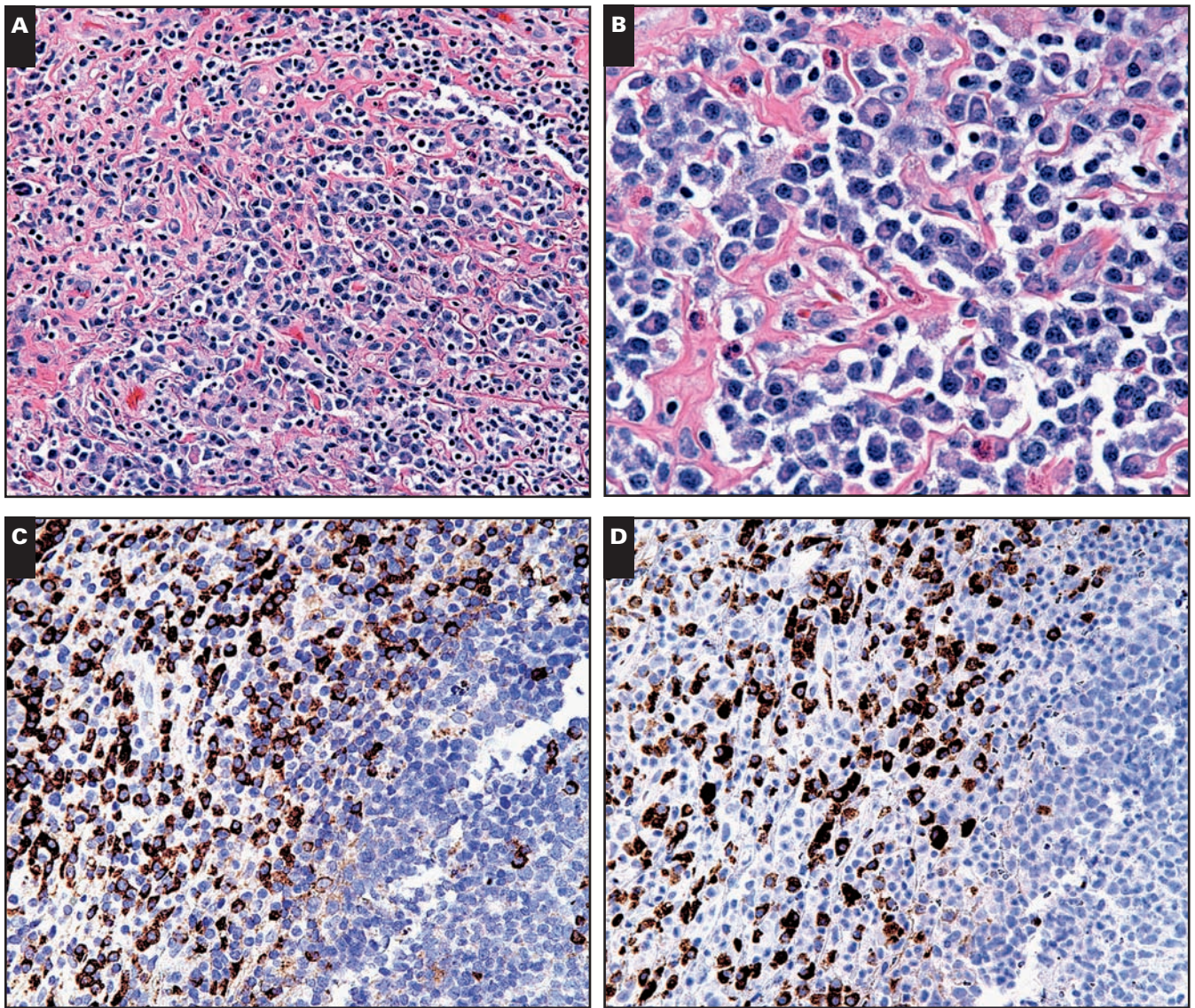


Image 5 **A** and **B**, Orbital tissue with clusters of plasma cells surrounded by thin bands of collagen (**A**, H&E, $\times 100$; **B**, H&E, $\times 400$). **C** and **D**, Immunohistochemical analysis shows that $>80\%$ of IgG-expressing plasma cells also show IgG4 expression (**C**, IgG, $\times 100$; **D**, IgG4, $\times 100$).

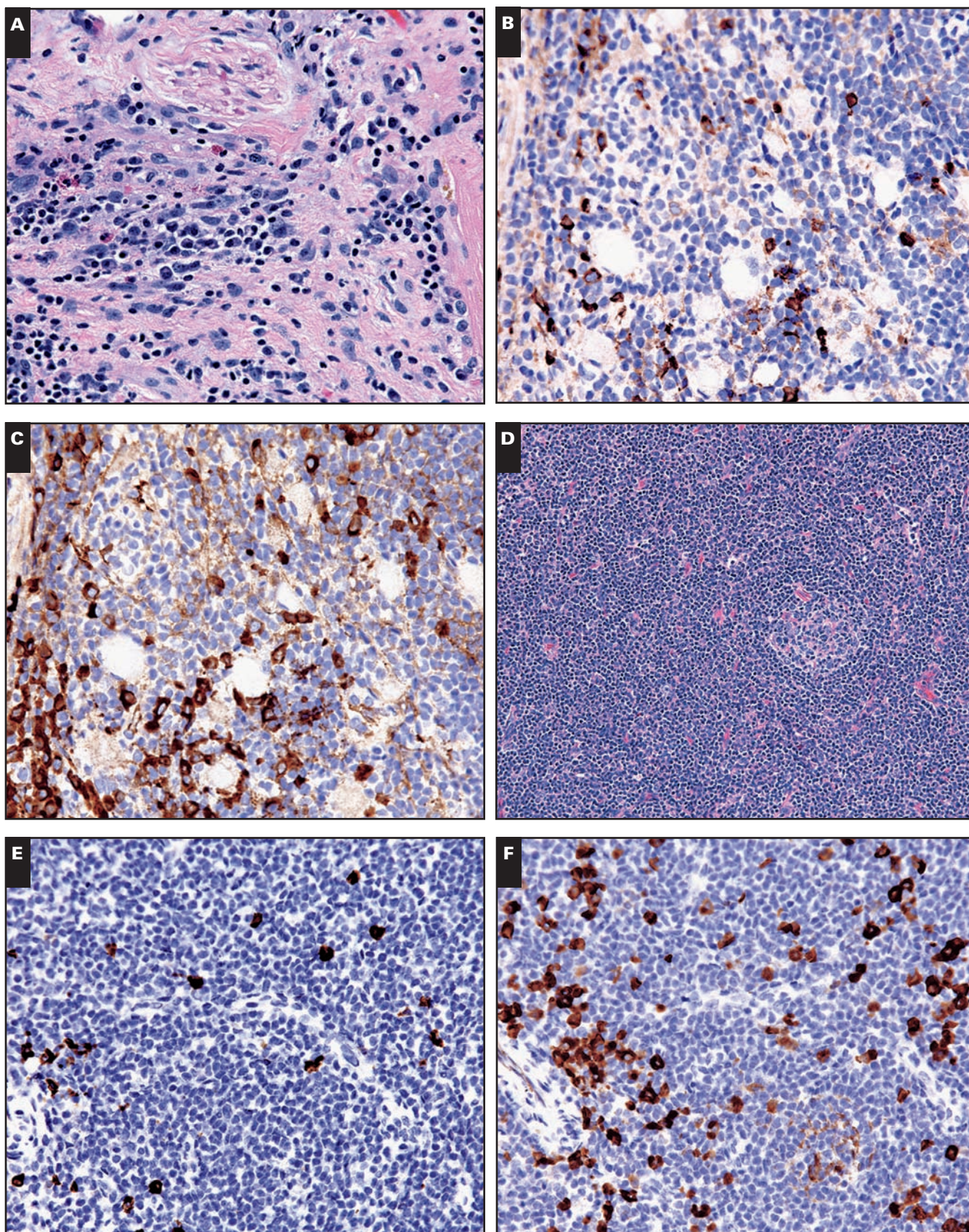


Image 6 **A**, Mixed inflammatory infiltrate with lymphocytes, eosinophils, and plasma cells set in collagenous fibrosis (H&E, ×200). **B** and **C**, Staining shows >95% of IgG-expressing plasma cells with IgG4 expression (**B**, IgG, ×100; **C**, IgG4, ×100). **D**, Axillary lymph node featuring atretic follicles with Castleman-like morphologic features (H&E, ×40). **E** and **F**, Immunohistochemical staining shows that >95% of IgG-expressing plasma cells also show IgG4 expression (**E**, IgG, ×100; **F**, IgG4, ×100).

which is a known pattern of systemic involvement in cases of IgG4 disease.¹⁶

These cases illustrate the difficulty in making a definite diagnosis of low-grade B-cell lymphoma in the context of IgG4 disease, particularly given reports of overlapping pathologies. Cheuk and colleagues¹⁰ reported 3 cases of ocular adnexal lymphoma (2 EMZL and 1 FL) arising from IgG4-related chronic sclerosing dacryoadenitis. They also reported 3 cases of EMZL arising in a background of sclerosing inflammation with increased numbers of IgG4-expressing plasma cells. Of their 6 cases, 2 also showed no evidence of clonal immunoglobulin gene rearrangement by polymerase chain reaction, though both cases demonstrated light chain restriction by immunostaining. These cases highlight the difficulty in diagnosing EMZL in this context. Patients with no detectable clonal population and IgG4-related pathology may benefit from a trial of corticosteroid therapy before receiving more aggressive chemotherapy and radiotherapy-based treatment regimens.

The biopsies reviewed in this series were performed owing to concern for lymphoma. Biopsy has advantages over fine-needle aspiration because it allows for evaluation of the background. As is evident from the cases in our series, IgG4 disease can exhibit various morphologic patterns. The most common morphologic pattern closely resembles reactive follicular hyperplasia with interfollicular plasmacytosis and mild to significant background sclerosis. It is also associated with subtle changes in follicular architecture, including Castleman-like atrophic follicles or progressive transformation of germinal centers, some of which have also been reported by other investigators.^{10,17} The dense lamellar fibrosis we observed in several of our cases is similar to that reported by Zen and Nakanuma.¹⁵

The criteria for a diagnosis of IgG4-related sclerosing disease of the orbit have not been well established: Increased numbers of IgG4-expressing plasma cells and an increased ratio (>30%) of IgG4/IgG-expressing plasma cells have been suggested as possible criteria.^{10,15} All of our 6 cases had these features. In addition, the ratio of IgG4-expressing plasma cells exceeded 50% of the IgG-expressing plasma cells in all 6 of our cases.

The ratio of IgG4- to IgG-expressing plasma cells may ultimately be the criterion that results in the diagnosis of IgG4 disease; however, it is important to be aware that other histologic changes in these biopsy specimens should also be recognized to lead diagnosticians to obtain additional immunohistologic stains such as IgG and IgG4 in the workup of an ocular lesion.

In our series, the most salient histologic features that raised IgG4 as a consideration in the differential diagnosis included the increased numbers of plasma cells without significantly atypical cytologic features and the presence

of even minimal amounts of sclerosis. In addition, architectural effacement, a prominent lymphoplasmacytic infiltrate, and lack of specific findings associated with infectious or lymphoid lesions, such as lymphoepithelial lesions, were also important considerations. Only 1 of 18 cases of reactive lymphoid hyperplasia featured a combination of a background of sclerosis and increased plasma cells. The absence of a clonal population of plasma cells given the increased numbers and often sheet-like proliferation of plasma cells is a good indication to consider additional immunohistologic investigation for IgG4 disease.

Given that about 22% of the cases in our series were initially diagnosed as reactive lymphoid hyperplasia, we suggest that specimens from all biopsies performed for suspected lymphoma in which a diagnosis of malignancy is not clearly established should undergo further workup for IgG4 disease. This practice would include cases with an atypical lymphoid or lymphoplasmacytic infiltrate in which light chain restriction or coexpression of aberrant markers on B cells (such as CD5, CD10, or CD43) is not demonstrable to render a definitive diagnosis of B-cell lymphoma. In tissue biopsy specimens, CD138 is a marker for quantifying plasma cells to which IgG and IgG4 should be added in an extended workup for the suspected diagnosis of IgG4 disease. Our results also support suggesting a diagnosis of IgG4-related systemic pathology for all cases in which the IgG4/IgG ratio exceeds 30%.

What is still unclear, however, is whether IgG4 disease is in fact a specific entity whose etiology remains unknown or whether it represents an epiphenomenon associated with several other immune-mediated disorders.

Our results bring attention to the frequency of IgG4-associated pathology in the ocular region and the extensive morphologic overlap with reactive lesions and low-grade B-cell lymphomas. In addition to an index of suspicion and awareness of this potential pitfall in diagnosis, the key morphologic features that raise concern for IgG4 disease are increased plasma cells and sclerosis. IgG4 disease is underrecognized, and, although its molecular pathogenesis is unknown, its accurate diagnosis is imperative because of its high response rate to corticosteroid therapy. Therefore, patients suspected of having IgG4 disease could be given a trial of corticosteroid therapy, which, if clinically efficacious, may avoid potential overtreatment with aggressive chemotherapy or radiotherapy.

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