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The Crux

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CONTENTS

Editorial

Sjögren's syndrome (SjS, SS) is a long-term autoimmune disease that affects the body's moisture-producing (lacrimal and salivary) glands, and often seriously affects other organ systems, such as the lungs, kidneys, and nervous system. Primary symptoms are dryness (dry mouth and dry eyes), pain and fatigue. Other symptoms can include dry skin, vaginal dryness, a chronic cough, numbness in the arms and legs, feeling tired, muscle and joint pains, and thyroid problems. Those affected are also at an increased risk (15%) of lymphoma.

While the exact cause is unclear, it is believed to involve a combination of genetics and an environmental trigger such as exposure to a virus or bacterium. It can occur independently of other health problems (primary Sjögren's syndrome) or as a result of another connective tissue disorder (secondary Sjögren's syndrome). Sjögren's syndrome may be associated with other autoimmune diseases, including rheumatoid arthritis (RA), systemic lupus erythematosus (SLE) or systemic sclerosis. The inflammation that results progressively damages the glands. Diagnosis is by biopsy of moisture-producing glands and blood tests for specific antibodies. On biopsy there are typically lymphocytes within the glands. The "**DISEASE DIAGNOSIS**" segment is totally devoted to **SJS**.

"**INTERPRETATION**" portion highlights the various extractable nuclear antigens or ENAs while "**TROUBLESHOOTING**" division enumerates various methodologies available to test or ANAs and pros and cons of each one of them.

"**BOUQUET**" of jokes, brain teasers and words of wisdom stays as always!

DISEASE DIAGNOSIS

SJOGREN SYNDROME

Practice Essentials

Sjögren syndrome is a systemic chronic inflammatory disorder characterized by lymphocytic infiltrates in exocrine organs. The disorder most often affects women, and the median age of onset is around 50 to 60 years. Most individuals with Sjögren syndrome present with sicca symptoms, such as xerophthalmia (dry eyes), xerostomia (dry mouth), and parotid gland enlargement.

In addition, numerous extraglandular features may develop, such as the following:

- Arthralgia
- Arthritis
- Raynaud phenomenon
- Myalgia
- Pulmonary disease
- Gastrointestinal disease
- Leukopenia
- Anemia
- Lymphadenopathy
- Neuropathy
- Vasculitis
- Renal tubular acidosis
- Lymphoma

About 50% of patients with Sjögren syndrome have cutaneous findings, such as dry skin (xeroderma), palpable and nonpalpable purpura, and/or urticaria. (See Etiology, Presentation, and Workup.) Primary Sjögren syndrome occurs in the absence of another underlying rheumatic disorder, whereas secondary Sjögren syndrome is associated with another underlying rheumatic disease, such as systemic lupus erythematosus (SLE), rheumatoid arthritis (RA), or scleroderma. Given the overlap of Sjögren syndrome with many other rheumatic disorders, it is sometimes difficult to determine whether a clinical manifestation is solely a consequence of Sjögren syndrome or is due to one of its overlapping disorders. Importantly, classic clinical features of Sjögren syndrome may also be seen in infections with certain viruses. These include hepatitis C virus, human immunodeficiency virus (HIV), and human T-cell lymphotrophic virus (HTLV). Most patients with primary Sjogren syndrome have two specific antibodies: against Ro (SS-A) and La (SSB) antigens. Treatment for Sjögren syndrome is largely based on symptoms (eg, lotion for dry skin, artificial tears for dry eyes). Rituximab has shown promise in the treatment of severe extraglandular manifestations (eg, vasculitis, cryoglobulinemia, peripheral neuropathy). Patients must be monitored carefully for the potential development of lymphoma, as the risk for this disease is significantly higher than in the general population.

Classification criteria

American-European Consensus Group classification

The American-European Consensus Group (AECG) criteria for the classification of Sjögren syndrome are outlined below. These criteria allow a diagnosis of Sjögren syndrome in patients without sicca symptoms or who have not undergone a biopsy. According to the American-European classification system (as modified by Tzioufas and Voulgarelis), diagnosis of primary Sjögren syndrome requires at least four of the criteria listed below; in addition, either criterion number 5 or criterion number 6 must be included. Sjögren syndrome can be diagnosed in patients who have no sicca symptoms if three of the four objective criteria are fulfilled. The criteria are as follows:

1. Ocular symptoms - Dry eyes for more than 3 months, foreign-body sensation, use of tear substitutes more than 3 times daily
2. Oral symptoms - Feeling of dry mouth, recurrently swollen salivary glands, frequent use of liquids to aid swallowing
3. Ocular signs - Schirmer test performed without anesthesia (< 5 mm in 5 min), positive vital dye staining results
4. Oral signs - Abnormal salivary scintigraphy findings, abnormal parotid sialography findings, abnormal sialometry findings (unstimulated salivary flow < 1.5 mL in 15 min)
5. Positive minor salivary gland biopsy findings
6. Positive anti–SSA or anti–SSB antibody results

Secondary Sjögren syndrome is diagnosed when, in the presence of a connective-tissue disease, symptoms of oral or ocular dryness exist in addition to criterion 3, 4, or 5, above. Application of these criteria has yielded a sensitivity of 97.2% and a specificity of 48.6% for the diagnosis of primary Sjögren syndrome. For secondary Sjögren syndrome, the specificity is 97.2% and the sensitivity, 64.7%.

Exclusion criteria include any of the following:

- Past head-and-neck irradiation
- Hepatitis C virus infection
- Acquired immunodeficiency syndrome (AIDS)
- Prior lymphoma
- Sarcoidosis
- Graft versus host disease
- Use of anticholinergic drugs

ACR/EULAR classification criteria for primary Sjogren syndrome

According to the ACR/EULAR classification criteria, individuals are classified as having primary Sjögren syndrome if they have a total score of 4 or higher, derived from the sum of the weights assigned to the following :

- Focal lymphocytic sialadenitis and focus score of ≥1 foci/4 mm² in labial salivary gland biopsy samples – weight/score 3
- Anti-SSA/Ro positive – weight/score 3
- Ocular Staining Score ≥5 (or van Bijsterveld score ≥4) in at least one eye – weight/score 1
- Schirmer's test ≤5 mm/5 min in at least one eye – weight/score 1
- Unstimulated whole saliva flow rate ≤0.1 mL/min – weight/score 1

For inclusion, patients must have at least one symptom of ocular or oral dryness, defined as a positive response to at least one of the following questions:

- Have you had daily, persistent, troublesome dry eyes for more than 3 months?
- Do you have a recurrent sensation of sand or gravel in the eyes?
- Do you use tear substitutes more than three times a day?
- Have you had a daily feeling or dry mouth for more than 3 months?
- Do you frequently drink liquids to aid in swallowing dry food?

Exclusion criteria include any of the following:

- History of head-and-neck radiation treatment
- Active hepatitis C infection (with confirmation by polymerase chain reaction [PCR]) testing
- AIDS
- Sarcoidosis
- Amyloidosis
- Graft versus host disease
- IgG4-related disease

Complications

Complications related to Sjögren syndrome include the following (see Prognosis, Treatment, and Medication):

- Emergence of disorders associated with Sjögren syndrome, such as SLE and RA
- Infection of the parotid gland, typically staphylococcal, streptococcal, or pneumococcal - clues include unilateral worsening of symptoms, along with tenderness, warmth, and erythema
- Emergence of parotid tumors - watch for unusually hard or unilateral parotid enlargement
- Pregnant patients with antiRo/SS-A antidodies are at risk for fetal loss, complete heart block in the fetus ,and neonatal lupus syndrome in the newborn
- Emergence of pseudolymphomas (pleomorphic cells that do not meet the criteria for malignancy) and non-Hodgkin B-cell lymphomas type was identified. Microscopic section of parotid biopsy, stained with immunoperoxidase for kappa light chains (brown-stained cells), showed monoclonal population of B cells

Etiology

Sjögren syndrome can occur as a primary disease of exocrine gland dysfunction or in association with several other autoimmune diseases (eg, systemic lupus erythematosus [SLE], rheumatoid arthritis, scleroderma, systemic sclerosis, cryoglobulinemia, polyarteritis nodosa). These primary and secondary types occur with similar frequency, but the sicca complex seems to cause more severe symptoms in the primary form. Virtually all organs may be involved. The disease commonly affects the eyes, mouth, parotid gland, lungs, kidneys, skin, and nervous system. The etiology of Sjögren syndrome is not well understood. The presence of activated salivary gland epithelial cells expressing major histocompatibility complex (MHC) class II molecules and the identification of inherited susceptibility markers suggest that environmental or endogenous antigens trigger a self-perpetuating inflammatory response in susceptible individuals. In addition, the continuing presence of active interferon pathways in Sjögren syndrome suggests ongoing activation of the innate immune system. Together, these findings suggest an ongoing interaction between the innate and acquired immune systems in Sjögren syndrome.

Association with the human leukocyte antigen

The frequency of HLA-DR52 in patients with primary Sjögren syndrome is estimated to be 87%, but it is also significantly increased in secondary Sjögren syndrome that occurs with rheumatoid arthritis or systemic lupus erythematosus. The genetic associations in Sjögren syndrome vary among ethnic groups. In white persons, for instance, the condition is linked to human leukocyte antigen (HLA)–DR3, HLA-DQ2, and HLA-B8, whereas the linkage is to HLA-DRB1*15 in Spanish persons and to HLA-DR5 in Greek and Israeli persons. Some evidence indicates that the true association of Sjögren syndrome may be with HLA-DQA1, which is in linkage disequilibrium with HLA-DR3 and HLA-DR5. These HLA associations appear restricted to individuals with Sjögren syndrome who have antibodies to the antigens SSA and SSB rather than to the disease manifestations themselves.

Possible disease triggers

Viruses are viable candidates as environmental triggers, although proof of causation has remained elusive, and certainly no single virus has been implicated. Epstein-Barr virus (EBV), HTLV-1, human herpesvirus 6 (HHV-6), HIV, hepatitis C virus (HCV), and cytomegalovirus (CMV) may have a role. Sjögrenlike syndromes are seen in patients infected with HIV, HTLV-1, and hepatitis C. Damage and/or cell death due to viral infection or other causes may provide triggering antigens to Toll-like receptors in or on dendritic or epithelial cells, which, by recognizing pathogen-associated patterns, are activated and begin producing cytokines, chemokines, and adhesion molecules. As T and B lymphocytes migrate into the gland, they themselves become activated by dendritic and epithelial cells, thereafter acting as antigen-presenting cells. Expressed antigens include SSA/Ro, SSB/La, alpha-fodrin and beta-fodrin, and cholinergic muscarinic receptors. Dendritic cell triggering by immune complexes formed from SSA-anti-SSA (or other immune complexes) may propagate the ongoing innate and acquired immune activation.

Glandular pathology

The pathology of a typical involved salivary or lacrimal gland in Sjögren syndrome reveals aggregations of lymphocytes—periductal at first, then panlobular. These cells are primarily CD4 T cells (75%) and memory cells, with 10% B cells and immunoglobulin-secreting plasma cells. Although individual lobules can be destroyed, salivary gland biopsy samples from patients with Sjögren syndrome typically retain 40%-50% of their viable glandular structure. Therefore, inflammatory destruction of salivary and lacrimal glands may not fully account for the symptoms of Sjögren syndrome. Studies suggest that the disease process of Sjögren syndrome has a neuroendocrine component. Proinflammatory cytokines released by epithelial cells and lymphocytes may impair neural release of acetylcholine. In addition, antibodies to acetylcholine (muscarinic) receptors may interfere with the neural stimulation of local glandular secretion, perhaps by interfering with aquaporin. Moreover, a study reports that M3 muscarinic receptor antibodies may cause autonomic dysfunction in patients with Sjögren syndrome. Current studies have also focused on the role of apoptotic mechanisms in the pathogenesis of primary Sjögren syndrome. A defect in Fas-mediated apoptosis, which is necessary for down-regulation of the immune response, can result in a chronic inflammatory destruction of the salivary gland, resembling Sjögren syndrome. Owing to these structural and functional changes in the lacrimal and salivary glands, their aqueous output is diminished. In the eye, tear hyperosmolarity results and is itself a proinflammatory stimulus, resulting in an inflammatory cascade on the ocular surface, with evidence of immune activation of the conjunctival epithelium and local cytokine and metalloproteinase production. Damage to the corneal epithelium, already vulnerable due to inadequate tear film protection, ensues, with resultant epithelial erosions and surface irregularity.

Extraglandular involvement

Extraglandular involvement in Sjögren syndrome manifests in part as hypergammaglobulinemia and the production of multiple autoantibodies, especially ANA and RF. This may be due to polyclonal B-cell activation, but the cause of this expanded activation is not known. Involvement of other organs and tissues may result from effects of these antibodies, immune complexes, or lymphocytic infiltration and occurs in one third of patients with Sjögren syndrome. Prolonged hyperstimulation of B cells may lead to disturbances in their differentiation and maturation and may account for the greatly increased incidence of lymphoma in these patients.

Sex hormones

Sex hormones may influence the immunologic manifestations of primary Sjögren syndrome, because the disease is much more common in women than in men. The prevalence of serologic markers tends to be lower in male patients than in female patients. Although the role of sex hormones (eg, estrogens, androgens) in the pathogenesis of primary Sjögren syndrome remains unknown, adrenal and gonadal steroid hormone deficiency probably affects immune function.

Prognosis

Sjögren syndrome carries a generally good prognosis. In patients who develop a disorder associated with Sjögren syndrome, the prognosis is more closely related to the associated disorder (eg, SLE, lymphoma). Interestingly, primary Sjögren syndrome is associated with lower cardiovascular risk factors and lower risk of serious cardiovascular events such as myocardial infarction and stroke, in comparison with SLE. Although salivary and lacrimal function generally stabilize, the presence of SSA and/or hypocomplementemia may predict a decline in function.

Morbidity and mortality

Morbidity associated with Sjögren syndrome is mainly associated with the gradually decreased function of exocrine organs, which become infiltrated with lymphocytes. The increased mortality rate associated with the condition is primarily related to disorders commonly associated with Sjögren syndrome, such as SLE, RA, and primary biliary cirrhosis. Patients with primary Sjögren syndrome who do not develop a lymphoproliferative disorder have a normal life expectancy.

Lymphoma

Among patients with Sjögren syndrome, the incidence of non-Hodgkin lymphoma is 4.3% (18.9 times higher than in the general population), with a median age at diagnosis of 58 years. The mean time to the development of non-Hodgkin lymphoma after the onset of Sjögren syndrome is 7.5 years. The most common histologic subtype of non-Hodgkin lymphoma in Sjögren syndrome is mucosa-associated lymphoid tissue (MALT) lymphoma, which can develop in any nonlymphoid tissue infiltrated by periepithelial lymphoid tissue—most commonly the salivary glands, but also the stomach, nasopharynx, skin, liver, kidneys, and lungs. The progression of these infiltrates to lymphoma occurs slowly and in a stepwise fashion. Lymphoma is present at more than 1 site in 20% of patients at initial diagnosis. The results of one study suggest that diagnostic labial salivary gland tissue biopsy can be used to detect germinal center-like lesions, which can be a highly predictive and easily obtained marker for non-Hodgkin lymphoma in primary Sjögren syndrome patients.

Risk factors for lymphoma include the following;

- Salivary gland enlargement
- Regional or generalized lymphadenopathy
- Hepatosplenomegaly
- Palpable purpura
- Leukopenia
- Renal insufficiency
- Loss of a previously positive polyclonal gammopathy
- Development of a monoclonal gammopathy or a monoclonal cryoglobulinemia
- RF positivity
- Anti-SSA/SSB positivity
- Hypocomplementemia

Pregnancy complications

Women with Sjögren syndrome are at higher risk for experiencing complications during pregnancy. Worsening of pulmonary hypertension and increased rates of spontaneous abortion and preterm deliveries have been reported. Children born to mothers with antibodies against SSA/Ro are at an increased risk of neonatal lupus and congenital heart block. If one such child develops congenital heart block, the risk for congenital heart block during a subsequent pregnancy is 15%.

Antiphospholipid syndrome

Patients with Sjögren syndrome who have antiphospholipid antibodies can develop the clinical features of antiphospholipid syndrome, which include increased fetal wastage and vascular thromboses.

Clinical Presentation

History

The clinical presentation of Sjögren syndrome may vary. Most patients are women, and onset is usually at age 40-60 years, but the syndrome also can affect men and children. The onset is insidious. The first symptoms in primary Sjögren syndrome can be easily overlooked or misinterpreted, and diagnosis can be delayed for as long as several years. Xerophthalmia (dry eyes) and xerostomia (dry mouth) are the main clinical presentations in adults. Bilateral parotid swelling is the most common sign of onset in children. Extraglandular involvement in Sjögren syndrome falls into two general categories: periepithelial infiltrative processes and extraepithelial extraglandular involvement. Periepithelial infiltrative processes include interstitial nephritis, liver involvement, and bronchiolitis and generally follow a benign course. Extraepithelial extraglandular involvement in Sjögren syndrome is related to B-cell hyperreactivity, hypergammaglobulinemia, and immune complex formation and includes palpable purpura, glomerulonephritis, and peripheral neuropathy. These latter manifestations occur later in the course of Sjögren syndrome and are associated with a higher risk of transformation to lymphoma. Symptoms of Sjögren syndrome can decrease the patient's quality of life in terms of its physical, psychological, and social aspects.

Sicca symptoms (dry eyes and dry mouth)

Although dry eyes and dry mouth are the most common symptoms in patients with Sjögren syndrome, most patients who report these symptoms have other underlying causes. The incidence of sicca symptoms increases with age. Indeed, more than one third of elderly persons have sicca symptoms. Whether this is part of the normal aging process (associated with fibrosis and atrophy observed on some lip

biopsy studies) or is due to the accumulation of associated illnesses and medications is unclear. Common medications that can cause sicca symptoms in any age group include antidepressants, anticholinergics, beta blockers, diuretics, and antihistamines. Anxiety can also lead to sicca symptoms. Women who use hormone replacement therapy may be at increased risk of dry eye syndrome.

Patients may describe the effects dry mouth in the following ways:

- Inability to eat dry food (eg, crackers) because it sticks to the roof the mouth
- Tongue sticking to the roof of the mouth
- Putting a glass of water on the bed stand to drink at night (and resulting nocturia)
- Difficulty speaking for long periods of time or the development of hoarseness
- Higher incidence of dental caries and periodontal disease
- Altered sense of taste
- Difficulty wearing dentures
- Development of oral candidiasis with angular cheilitis, which can cause mouth pain

Dry eyes may be described as red, itchy, and painful. However, the most common complaint is that of a gritty or sandy sensation in the eyes. Symptoms typically worsen throughout the day, probably due to evaporation of the already scanty aqueous layer. Some patients awaken with matting in their eyes and, when severe, have difficulty opening their eyes in the morning. Blepharitis can also cause similar morning symptoms.

Parotitis

Patients with Sjögren syndrome may have a history of recurrent parotitis, often bilateral. Although in some patients the parotid glands become so large that the patients report this as a problem, more often the examining physician discovers them.

Cutaneous symptoms

Nonvasculitic cutaneous manifestations in Sjögren syndrome include the following :

- Dryness
- Eyelid dermatitis
- Pruritus
- Erythema annulare

Cutaneous vasculitis, such as palpable purpura, develops in some patients with Sjögren syndrome, especially those with hypergammaglobulinemia or cryoglobulinemia. Raynaud phenomenon is observed in approximately 20% of patients.

Pulmonary symptoms

Patients with Sjögren syndrome can develop dryness of the tracheobronchial mucosa (xerotrachea), which can manifest as a dry cough. Less often, patients develop dyspnea from an interstitial lung disease that is typically mild. Patients may develop recurrent bronchitis or even pneumonitis (infectious or noninfectious).

Gastrointestinal symptoms

Dryness of the pharynx and esophagus frequently leads to difficulty with swallowing (deglutition), in which case patients usually describe food becoming stuck in the upper throat. Lack of saliva may lead to impaired clearance of acid and may result in gastroesophageal reflux and esophagitis. Abdominal pain and diarrhea can occur. Rarely, patients develop acute or chronic pancreatitis, as well as malabsorption due to pancreatic insufficiency. However, caution is advised when interpreting laboratory results because an elevated amylase level may arise from the parotid gland. Patients with gastritis should be tested for *Helicobacter pylori* infection, because of its association with gastric mucosa–associated lymphoid tissue lymphomas. Patients with Sjögren syndrome are at increased risk for delayed gastric emptying, which can cause early satiety, upper abdominal discomfort, nausea, and vomiting.

Cardiac symptoms

Pericarditis and pulmonary hypertension, with their attendant symptomatology, can occur in Sjögren syndrome. Orthostatic symptoms related to dysfunction of autonomic control of blood pressure and heart rate is associated with increased severity of Sjögren syndrome.

Neurologic symptoms

The occurrence of central nervous system (CNS) and spinal cord involvement in Sjögren syndrome is estimated by various studies to be 8-40%, with manifestations including myelopathy, optic neuropathy, seizures, cognitive dysfunction, and encephalopathy. Attempts must be made to distinguish other causes of these symptoms, including concomitant SLE, multiple sclerosis, cerebrovascular disease, and Alzheimer disease. Sensory, motor, or sensorimotor peripheral neuropathy, often subclinical, can be detected in up to 55% of unselected patients with Sjögren syndrome. Symptoms of distal paresthesias may be present. Cranial neuropathies can develop, particularly trigeminal neuropathy or facial nerve palsy. Mononeuritis multiplex should prompt a search for a vasculitis. Progressive weakness and paralysis secondary to hypokalemia due to underlying renal tubular acidosis can occur and is potentially treatable.

Renal symptoms

Renal calculi, renal tubular acidosis, and osteomalacia, nephrogenic diabetes insipidus, and hypokalemia can occur secondary to tubular damage caused by interstitial nephritis, the most common form of renal involvement in Sjögren syndrome. Interstitial cystitis, with symptoms of dysuria, frequency, urgency, and nocturia, is strongly associated with Sjögren syndrome. Glomerulonephritis can be caused by Sjögren syndrome but is uncommon and is usually attributable to another disorder, such as SLE or mixed cryoglobulinemia.

Additional symptoms

Nasal dryness can result in discomfort and bleeding. Women may also have a dry vagina, which can lead to dyspareunia, vaginitis, and pruritus.

Patients with Sjögren syndrome may report fatigue, joint pain, and, sometimes, joint swelling. A careful review of systems must be performed to differentiate these from the manifestations of other disorders (see DDx). Fibromyalgia is common in patients with Sjögren syndrome, with a prevalence of about 31%. Women with Sjögren syndrome may have a history of recurrent miscarriages or stillbirths, and women and men may have a history of venous or arterial thrombosis. These are related to the presence of antiphospholipid antibodies (eg, lupus anticoagulant or anticardiolipin antibodies).

Secondary Sjogren syndrome

Secondary Sjögren syndrome appears late in the course of the primary disease. However, in some patients, primary Sjögren syndrome may precede SLE by many years. Secondary Sjögren syndrome is usually mild, and sicca symptoms are the main feature. Unlike patients with primary Sjögren syndrome, persons with the secondary type have significantly fewer systemic manifestations. These manifestations include the following:

- Salivary gland swelling
- Lung involvement

- Nervous system involvement
- Renal involvement
- Raynaud phenomenon
- Lymphoproliferative disorders

In secondary Sjögren syndrome, symptoms of the primary disease predominate. Secondary Sjögren syndrome does not modify the prognosis or outcome of the basic disease. Polyarteritis nodosa and Sjögren syndrome may also coexist, perhaps best viewed as an overlap syndrome.

Clinical Presentation

Physical Examination

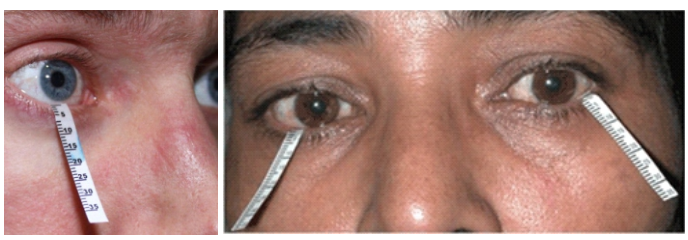
The physical signs of primary Sjögren syndrome can be divided into glandular and extraglandular signs.

Glandular signs

Ocular

While it is important to look for corneal lesions and a decreased tear pool in the lower conjunctiva during physical examination, patients with Sjögren syndrome should be referred to an ophthalmologist for more formal testing of keratoconjunctivitis sicca (KCS). This testing applies grading criteria of inflammatory changes that can direct therapy aimed at preventing corneal damage. In addition, conditions that mimic KCS, such as blepharitis, conjunctivitis, and uveitis can be ruled out or treated.

Patients with Sjögren syndrome may have dilated conjunctival vessels, as well as pericorneal injection and dullness or irregularity of the corneal image. Blepharitis may be present as an alternate or additional problem, particularly if the lower eyelid is inflamed. Mucinous threads and filamentary keratosis can be detected during a slit-lamp examination. The relative lack of the aqueous layer also leads to rapid tear breakup. In the Schirmer test, a bent piece of Whatman number 41 filter paper is placed in the lower conjunctiva, and the amount of tearing on the filter paper is recorded. Normal wetting is greater than 15 mm after 5 minutes, whereas a definitive positive result is less than 5 mm after 5 minutes. This test can help to exclude or confirm significant dryness of the eyes, but it is not disease-specific. Furthermore, false-positive results occur. An evaluation of the diagnostic performance of the Schirmer test yielded a sensitivity of 42% and a specificity of 76% for Sjögren syndrome. (See the image below.)

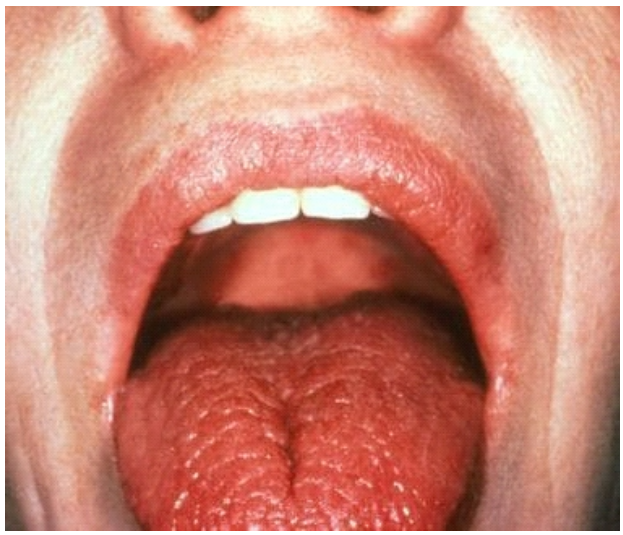


Photographs that demonstrates the Schirmer test, which is used to detect deficient tear production in patients with Sjögren syndrome. The filter paper strip is placed at the junction of the eyelid margins. After 5 minutes, 15 mm of paper should be moistened if tear production is normal, as shown here. Persons older than 40 years may moisten between 10 mm and 15 mm. Patients with Sjögren syndrome have less moistening. Sjögren syndrome is most common in patients with rheumatoid arthritis but may also occur without associated disease and in systemic lupus erythematosus, polyarteritis, systemic sclerosis, lymphoma, and sarcoidosis.

Oral

Oral signs include the following:

- Dryness
- Tongue - Red, smooth, and dry (see the image below)
- Dental caries - Severe and progressive
- Parotid duct narrowing
- Lips - Red, dry, and scaly
- Cracks at the corners of the mouth
- Chronic oral candidiasis



Dryness of the mouth and tongue due to lack of salivary secretion is characteristic of xerostomia associated with Sjögren syndrome. Mouth dryness may produce a deep red tongue, as shown here, and dental caries are common. Look for a decreased sublingual salivary pool. The tongue may stick to the tongue depressor. Patients with Sjögren syndrome may develop frequent caries, sometimes in unusual locations such as the incisor surface and the gum line. Patients with Sjögren syndrome are prone to develop oral candidiasis. In addition to white patches, watch for petechial lesions, loss of tongue papilla, erythema and fissuring of the tongue, erythema on other mucosal surfaces, and angular cheilosis. Oral candidiasis can be seen under dentures. Gingival inflammation has been found to be more evident in the individuals with Sjögren syndrome, particularly those with secondary Sjögren syndrome. Periodontal disease can lead to loss of teeth.

Parotid glands

Sjögren syndrome appears to negatively affect the periodontal condition. Recurrent swelling of the parotid glands (22-66% of patients) occurs; submaxillary and sublingual gland swelling also sometimes takes place. Bilateral parotid gland enlargement is common in persons with Sjögren syndrome (see the image below). Some waxing and waning of size may occur. Exudates from the parotid gland are largely lymphocytes. Rock-hard or unilateral parotid gland enlargement should prompt referral to an otolaryngologist for biopsy to exclude a tumor. Other causes of parotid enlargement include diabetes, sarcoidosis, amyloidosis, diffuse infiltrative lymphocytic syndrome (DILS) of HIV disease, hepatitis C, and alcoholism. Acute, unilateral parotitis may be caused by Sjögren syndrome, infection, or obstruction, although the latter 2 conditions are more often associated with a very tender parotid gland and accompanying fever.

Other mucous membranes

Other mucous membrane signs include the following:

- Atrophic changes in the mucous membranes of the upper respiratory tract, leading to nasal dryness, recurrent infections, hoarseness, and aphonia
- Atrophic rhinitis
- Atrophic changes in the vulva and vagina resulting in pruritus and vaginitis
- Dryness of the anal and rectal mucous membranes (eg, pruritus, inflammation)

Cutaneous

Dryness of the skin occurs in 50% of patients with Sjögren syndrome; scaling occurs in about 25% of patients. The skin may be irritable, with secondary lichenification. Partial or complete loss of sweating may be present.

Hair may be dry, sparse, and brittle; diffuse alopecia may involve the scalp, limbs, axillae, or pubis. Nail folds may show capillaroscopic abnormalities, which are associated with the presence of antiendothelial cell antibodies. Erythema of the nose and cheeks may be present. Patients with Sjögren syndrome can develop a nonpalpable or palpable, vasculitic purpura, with lesions that are typically 2-3 mm in diameter and located on the lower extremities. The lesions, which can ulcerate, occur most often in patients with hypergammaglobulinemia or cryoglobulinemia. Annular erythema with scales, localized especially on the face and neck, is recognized as a cutaneous manifestation of Sjögren syndrome. The patches are recurrent and resolve without hyperpigmentation; no photosensitivity is observed. Annular erythema is a common cutaneous manifestation in Japanese and other Asian patients; however, it is rarely seen in white patents.

In Japanese patients with Sjögren syndrome, annular erythema is divided into the following 3 types:

- Sweet disease–like annular erythema with an elevated border
- Subacute cutaneous lupus erythematosus–like, marginally scaled erythema
- Papular erythema

Those lesions bear some clinical similarities to the annular lesions of subacute cutaneous lupus erythematosus, but their histopathologic features are distinct from those of subacute cutaneous lupus erythematosus. Significant mucin depositions are observed.

Extraglandular signs

Gastrointestinal

Gastrointestinal tract signs include the following:

- Esophageal motility abnormalities
- Pancreatic involvement
- Splenomegaly
- Digestive symptoms (due to atrophy of the gastric mucous membrane with achlorhydria)
- Hepatitis (13%)

Pulmonary

Pulmonary abnormalities occur in 9-29% of cases; they are similar in primary and secondary Sjögren syndrome. Lung signs include the following :

- Pulmonary fibrosis
- Pulmonary hypertension
- Recurrent chest infections
- Granulomatous infiltration and fibrosing alveolitis
- Restrictive ventilatory defect
- Impaired gas transfer
- Bibasilar rales – Can be heard in patients with interstitial lung disease

Articular

Articular changes (eg, arthritis) occur in 42% of patients with Sjögren syndrome; arthritis can be a component of either the primary or secondary form of the disease. One third of patients with RA have Sjögren syndrome. Symmetrical, polyarticular, inflammatory arthritis suggests underlying RA or a connective-tissue disease such as SLE or scleroderma. The arthritis in patients with primary Sjögren syndrome is typically nonerosive and mild.

Urinary tract

Patients with Sjögren syndrome have significantly more urinary problems than do those without Sjögren syndrome. Patients may have the following:

- Irritated bladder
- Suprapubic pain
- Renal tubular dysfunction - Patients with primary Sjögren syndrome commonly are first seen because of renal impairment, usually from renal tubular dysfunction
- Renal tubular acidosis - This affects one third of patients with Sjögren syndrome; a correlation apparently exists between hypergammaglobulinemia and distal renal tubular acidosis
- Interstitial nephritis - This is rare, occurring in 4% of cases; it is often accompanied by cryoglobulinemia, a decreased level of complement, and the presence of circulating immune complexes
- Impaired renal concentrating ability, generalized aminoaciduria

Neurologic

A combination of lesions and relapses can suggest multiple sclerosis. Myelopathy rarely occurs in the course of primary Sjögren syndrome. It appears as Brown-Séquard syndrome, acute transverse myelitis, or progressive myelopathy. Clinically, cases with nervous system involvement present with paraparesis or paraplegia resulting from lesions at the thoracic or cervicothoracic levels. Peripheral neuropathy occurs in 10-35% patients with primary Sjögren syndrome. Peripheral nerve dysfunction—such as trigeminal sensory neuropathy, mononeuropathy multiplex, distal sensorimotor polyneuropathy, or pure sensory neuropathy—may occur. This tends to be a small-fiber peripheral neuropathy. Painful, distal paresthesias in the feet may be evident, as may abnormal sweating. Examination may reveal findings that include decreased pinprick sensation. Isolated cranial nerve involvement rarely occurs in primary Sjögren syndrome. CNS involvement also is less common (10-25% of patients with Sjögren syndrome) than are other types of involvement; CNS pathology ranges from neuropathy, hemiparesis, transverse myelitis, and dystonia to encephalopathy and dementia. In Sjögren syndrome, focal brain lesions can be present in the cerebral white matter. In addition, dysregulation of hypothalamic-pituitary-adrenal and thyroid axes can cause some neurologic disturbances.

Differential Diagnoses

Diagnostic Considerations

Sjögren syndrome is associated with a wide variety of other disorders, which may contribute to underdiagnosis or misdiagnosis. A careful review of systems is needed to detect problems such as rheumatoid arthritis (RA), systemic lupus erythematosus (SLE), scleroderma,

polymyositis, chronic active hepatitis, idiopathic pulmonary fibrosis, primary biliary cirrhosis, and autoimmune thyroid disease. Xerophthalmia, xerostomia, and enlargement of the parotid glands can result from adverse effects of drugs and other diseases. HIV infection can result in diffuse infiltrative lymphocytosis syndrome (DILS), which is characterized by parotid enlargement; involvement of the renal, lung, and gastrointestinal systems; and a low frequency of autoantibody presence. Chronic graft versus host disease may mimic symptoms associated with idiopathic Sjögren syndrome. SLE might be considered, especially at onset of the disease.

Histologic findings of the following disorders can be consistent with Sjögren syndrome:

- Sarcoidosis
- Graft versus host disease
- HIV infection
- HTLV-1 infection
- Hepatitis C virus (HCV) infection
- Keratoconjunctivitis sicca

Sicca

Differential diagnoses to consider in patients with sicca include the following:

- Medications (eg, antidepressants, anticholinergics, beta-blockers, diuretics, antihistamines, some antiarrhythmic and antiepileptic drugs)
- Anxiety and depression
- Viral infections (eg, mumps)
- Complications from contact lenses
- Dehydration
- Hypervitaminosis A
- Neurotropic keratitis
- Mucous membrane pemphigoid
- Environmental irritants
- Mouth breathing
- Chronic blepharitis
- Chronic conjunctivitis
- Rosacea
- Therapeutic radiation or surgery to the head and neck
- Age
- Alzheimer disease
- Parkinson disease
- Bell palsy
- Amyloidosis
- Sarcoidosis
- Lymphoma

Parotid enlargement

Differential diagnoses to consider in patients with parotid enlargement include the following:

- Viral infection (eg, mumps, Epstein-Barr virus, cytomegalovirus, coxsackievirus A, influenza)
- DILS associated with HIV disease
- Granulomatous diseases (sarcoidosis, tuberculosis, leprosy)
- Hyperlipoproteinemia
- Hepatic cirrhosis
- Hepatitis C
- Bulimia
- Recurrent parotiditis of childhood
- Chronic pancreatitis
- Acromegaly
- Amyloidosis
- Gonadal hypofunction
- Diabetes mellitus
- Salivary gland tumor (primarily unilateral)
- Bacterial infection (primarily unilateral)
- Chronic sialadenitis (primarily unilateral)
- Lymphoma

Associated disorders

Importantly, evaluate the patient for disorders associated with Sjögren syndrome, including the following:

- AIDS
- Rheumatoid arthritis
- Systemic lupus erythematosus (SLE)
- Scleroderma
- Polymyositis
- Primary biliary cirrhosis
- Thyroiditis
- Chronic active hepatitis
- Mixed cryoglobulinemia
- Celiac sprue

Differential Diagnoses

- Bulimia Nervosa
- Chronic Pancreatitis
- IgG4-related systemic disease
- Immunoglobulin-Related Amyloidosis
- Polymyositis
- Rheumatoid Arthritis (RA)
- Benign Tumors of Major Salivary Glands
- Benign Tumors of Minor Salivary Glands
- Sarcoidosis
- Scleroderma
- Tuberculosis (TB)

Workup

Approach Considerations

Some laboratory tests can be used to assess salivary and lacrimal involvement in Sjögren syndrome. However, no single test is sufficiently sensitive or specific in the diagnosis of Sjögren syndrome. The condition is properly diagnosed only when the results of various tests are simultaneously positive and when subjective symptoms and serologic abnormalities are present.

Laboratory test results may indicate the following:

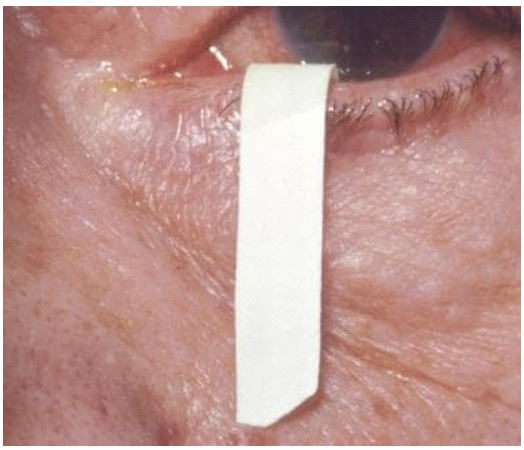
- Elevated erythrocyte sedimentation rate (ESR)
- Anemia
- Leukopenia
- Eosinophilia
- Hypergammaglobulinemia
- Presence of antinuclear antibodies (ANAs), especially anti-Ro and anti-La
- Presence of rheumatoid factor (RF)

- Presence of anti–alpha-fodrin antibody (reliable diagnostic marker of juvenile Sjögren syndrome)
- Creatinine clearance may be diminished in up to 50% of patients

Multiple autoantibodies are associated with Sjögren syndrome. In a study in which atypical autoantibodies were evaluated in 82 patients with primary Sjögren syndrome, an immunologic overlap (defined by the presence of autoantibodies typical of other systemic autoimmune diseases) was evident in 20% of the patients. The clinical significance of these atypical autoantibodies varied widely. Patients with primary Sjögren syndrome may have positive test results for lupus anticoagulant and/or anticardiolipin antibodies, and some patients develop clinical events (ie, fetal wastage, arterial and/or venous thrombosis) associated with antiphospholipid syndrome. Anti salivary duct antibodies are present in most cases of secondary Sjögren syndrome. Type II cryoglobulins are noted, particularly in patients with palpable and nonpalpable vasculitic purpura. Hepatitis C should be sought in these patients. In some studies, patients with Sjögren syndrome have an increased frequency of autoimmune thyroid disease with hypothyroidism (10-15%). Lymphocytic i)nfiltration can be observed in the thyroid gland. Elevations of serum immunoglobulin G4 (IgG4), found in IgG4-related plasmacytic disease (which can mimic the glandular infiltrations of Sjögren syndrome), are not seen in Sjögren syndrome. Antibodies to carbonic anhydrase 11 can be seen in patients with Sjögren syndrome who have primary billiary cirrhosis.

Schirmer test

The Schirmer test is probably the only test available in the emergency department (ED) that can be used to strongly support or refute suspicion of Sjögren syndrome. A test strip of number 41 Whatman filter paper is placed near the lower conjunctival sac to measure tear formation. Healthy persons wet 15 mm or more of the paper after 5 minutes. A positive test occurs when less than 5 mm of the strip is wet after 5 minutes. A Schirmer test is shown in the image below.



Photograph that demonstrates the Schirmer test, which is used to detect deficient tear production in patients with Sjögren syndrome. The filter paper strip is placed at the junction of the eyelid margins. After 5 minutes, 15 mm of paper should be moistened if tear production is normal, as shown here. Persons older than 40 years may moisten between 10 mm and 15 mm. Patients with Sjögren syndrome have less moistening. Sjögren syndrome is most common in patients with rheumatoid arthritis but may also occur without associated disease and in systemic lupus erythematosus, polyarteritis, systemic sclerosis, lymphoma, and sarcoidosis.

Rheumatoid factor

RF is present in 52% of patients with primary Sjögren syndrome and in 98% of patients with the secondary disease, occurring even when rheumatoid arthritis is not present. Consider a diagnosis of rheumatoid arthritis if the patient has symmetrical polyarticular synovitis. The presence of RF has been independently associated with elevated risk for lymphoma in patients with primary Sjögren syndrome. However, loss of a previously positive RF finding can be observed in some patients with Sjögren syndrome who develop lymphoma.

Antinuclear antibodies

ANAs are typically present in patients with Sjögren syndrome. Consider the diagnosis of systemic lupus erythematosus (SLE) in patients with ANAs only if symptoms and signs typical of SLE are present.

Serum protein electrophoresis

Patients with Sjögren syndrome often have a polyclonal gammopathy. Loss of a previously detected polyclonal gammopathy can be observed in some patients with Sjögren syndrome who develop lymphoma. Development of a monoclonal gammopathy can also signal the development of a lymphoma.

Staining

Rose bengal is an aniline dye that stains epithelial surfaces with diminished mucin protection or with exposed epithelial cell membranes. Conjunctival staining can be detected with the naked eye. Slit-lamp examination is performed after rose bengal staining to detect abnormal uptake in the cornea. Lissamine green staining works similarly but is less irritating to the eye. Fluorescein staining can be used to detect corneal damage.

Salivary testing

Sialometry is a good measure of the degree of decreased salivary flow and helps to establish xerostomia, but the findings do not narrow the differential diagnoses. Saliva from patients with Sjögren syndrome has elevated levels of sodium, chloride, lactoferrin, and IgA, but these findings are not specific.

Sedimentation rate

The ESR is elevated in 80% of patients with Sjögren syndrome, but the finding is nonspecific.

Protein profiling

Protein profiling (tear proteomics) has revealed reproducible patterns in patients with primary Sjögren syndrome and appears to hold promise as a diagnostic test for this disorder.

Additional test considerations

Other test results to consider are as follows:

- High total protein level or a low albumin level - Should prompt the clinician to perform serum protein electrophoresis
- High alkaline phosphatase level - Should prompt consideration of primary biliary cirrhosis
- Elevated transaminase levels - Consider the possibility of chronic active hepatitis, which can be associated with sicca symptoms, or hepatitis C, which can cause mild salivary gland enlargement; however, mild (< 2-fold) increases in transaminase levels have been observed in 22% of patients with Sjögren syndrome
- Low bicarbonate level - Consider evaluating patients with a low bicarbonate level for type I (distal) renal tubular acidosis; less commonly, patients can also develop proximal renal tubular acidosis with Fanconi syndrome

- Hypokalemia - This condition, which is occasionally severe enough to lead to periodic paralysis, can be observed in patients with type I renal tubular acidosis; however, it can also be observed in patients who have Sjögren syndrome without renal tubular acidosis.

SSA and SSB

Antibodies against SSA/Ro are found in approximately 50% of patients with the disease (75% of patients with primary Sjögren syndrome and 15% of patients with secondary Sjögren syndrome). Thus, the absence of anti-SSA/Ro antibodies does not eliminate the diagnosis of primary or secondary Sjögren syndrome. Anti-Ro is a polyclonal antibody directed against nuclear and nucleolar RNA binding protein of 60KD or cytoplasmic protein of 52KD (E3 ubiquitin ligase) . Patients can have a negative ANA and a positive Ro antibody test if they only have anti-Ro against cytoplasmic protein of 52KD. Anti-La is an oligoclonal antibody that is predominantly directed against nuclear 47KD RNA binding protein. Antibodies against SSA/Ro are present in 50% of patients with SLE and are sometimes found in healthy individuals. Thus, the presence of antibody against SSA/Ro cannot by itself be used to establish a diagnosis of Sjögren syndrome. Antibodies against SSB/La are present in 40-50% of patients with primary Sjögren syndrome and in 15% of patients with SLE. Finding antibodies against SSB/La in patients without antibodies against SSA/Ro is unusual, but this combination has occurred in patients with primary biliary cirrhosis and autoimmune hepatitis. Titers of anti-SSA/Ro and anti-SSB/La antibodies do not reflect disease activity. Current enzyme-linked immunosorbent assay (ELISA) tests for these antibodies are more sensitive than previous tests. Thus, the specificity is lower. Antibodies against SSA/Ro are also associated with the annular erythematous lesions of subacute cutaneous lupus. They are also found in the mothers of newborns with neonatal lupus syndromes and congenital heart block, and some of these mothers have or will develop Sjögren syndrome.

CBC

In patients with Sjögren syndrome, the complete blood count (CBC) is most often within the reference range, but anemia of chronic disease may be present. Pernicious anemia may be associated with the atrophic gastritis. An abnormal white blood cell (WBC) count, especially with an abnormal differential count, should prompt concerns for a lymphoreticular malignancy. In addition, although a low platelet or WBC count can occur in persons with primary Sjögren syndrome, the finding should also prompt consideration for coexisting SLE. A mild, normochromic, normocytic anemia is present in 50% of patients. Leukopenia occurs in up to 42% of patients.

Sialography and Scintigraphy

In sialography, radiopaque material is injected into the salivary glands. Sialography is useful to exclude the presence of obstructions or strictures, but the diffuse sialectasis of Sjögren syndrome is seen in various other diseases and is therefore not specific. Oil-based contrast medium may not be adequately cleared in patients with Sjögren syndrome and, consequently, may damage adjacent tissues and lead to a chronic granulomatous reaction. Performing this procedure with oil-based contrast should be avoided, especially during episodes of acute swelling. With salivary scintigraphy, the uptake and secretion of sodium pertechnetate technetium-99m (Tc) is a gauge of the salivary flow rates and can provide an objective measurement of salivary gland dysfunction. However, the finding of low flow rates is not specific to Sjögren syndrome. Positive findings on either sialography or scintigraphy fulfill a criterion for objective evidence of Sjögren syndrome by the American-European Consensus Group.

Biopsy

Minor salivary gland biopsy currently is the best single test to establish a diagnosis of Sjögren syndrome. In this procedure, an incision is made on the inner lip, and some minor salivary glands are removed for examination. In patients with a possible diagnosis of this disease but with severe extraglandular symptoms, a lip biopsy is often performed to firmly establish the diagnosis of Sjögren syndrome. Obtaining the biopsy sample from below normal-appearing mucosa is important in order to avoid false-positive results. At least 4 salivary gland lobules should be obtained for analysis. While this is the most definitive test, performing it is not absolutely necessary from a clinical standpoint. Patients with Sjögren syndrome are essentially treated symptomatically and are observed for the development of other rheumatic disorders or lymphoma. This can be initiated without performing a biopsy. If, however, the diagnosis is in doubt or if a definitive diagnosis is needed, then this is the best test. Salivary gland biopsy can also help to detect pseudolymphoma or lymphoma, as well as the noncaseating granulomas of sarcoidosis. One study showed that not all patients undergoing lip biopsy derived diagnostic benefit from this procedure and that clinical symptoms and serology did not predict a positive lip biopsy. In another study, however, a significant correlation was found between positive findings in minor salivary gland biopsy and the Schirmer test, the rose bengal test, xerostomia, and parotid swelling. The investigators utilized biopsy specimens from the lower lip of 360 patients.

Histologic Findings

Although pathologists use different classification systems, the characteristic findings of minor salivary gland biopsy in a person with Sjögren syndrome include the following (see the image below) :

- The biopsy shows focal aggregates of at least 50 lymphocytes, and, to a lesser extent, plasma cells and macrophages
- More than 1 focal aggregate is seen per 4 mm
- T cells, predominantly CD4 cells, are present, unlike the predominance of CD8 T cells seen in the salivary gland biopsy samples from patients with DILS associated with HIV disease
- Normal acini are replaced by lymphocytes
- Focal aggregates are seen in almost all glands
- Ten percent of the lymphocytes are CD5 B cells that produce IgM and IgG antibodies, often with a monoclonal or oligoclonal pattern
- Large foci are present, possibly showing germinal centers
- Epimyoepithelial islands are uncommon in the minor salivary gland but can be seen in the major salivary glands

Lymphocytic infiltrates are also seen in other organs. Findings from a gastric mucosal biopsy may show lymphocytic infiltrates with atrophic gastritis. A kidney biopsy may show interstitial lymphocytic infiltration. Lung biopsy can reveal infiltrating CD4 T cells of a lymphocytic interstitial pneumonitis. Salivary gland biopsy can help to detect pseudolymphoma or lymphoma, as well as the noncaseating granulomas of sarcoidosis.

Treatment & Management

Approach Considerations

No curative agents for Sjögren syndrome exist. The treatment of the disorder is essentially symptomatic.

Skin and vaginal dryness

Patients should use skin creams, such as Eucerin, or skin lotions, such as Lubriderm, to help with dry skin. Vaginal lubricants, such as Replens, can be used for vaginal dryness. Vaginal estrogen creams can be considered in postmenopausal women. Watch for and treat vaginal yeast infections.

Arthralgias and arthritis

Acetaminophen or nonsteroidal anti-inflammatory drugs (NSAIDs) can be taken for arthralgias. Consider hydroxychloroquine if NSAIDs are not sufficient for the synovitis occasionally associated with primary Sjögren syndrome. However, hydroxychloroquine does not relieve sicca symptoms. Patients with RA associated with Sjögren syndrome likely require other disease-modifying agents.

Additional treatment considerations

In patients with major organ involvement, such as lymphocytic interstitial lung disease, consider therapy with steroids and immunosuppressive agents, such as cyclophosphamide. While cyclophosphamide and similar agents may be helpful for treating serious manifestations of Sjögren syndrome or disorders associated with Sjögren syndrome, clinicians should understand that these agents are also associated with the development of lymphomas. Long-term anticoagulation may be needed in patients with vascular thrombosis related to antiphospholipid antibody syndrome. In a small group of patients with primary Sjögren syndrome, mycophenolate sodium reduced subjective, but not objective, ocular dryness and significantly reduced hypergammaglobulinemia and RF. Among the biologic therapies, the greatest experience in primary Sjögren syndrome is with rituximab, an anti-CD20 (which is expressed on B-cell precursors) monoclonal antibody. Anti-B–cell strategies, particularly rituximab, have a promising effect in the treatment of patients with severe extraglandular manifestations of Sjögren syndrome. Reports on the use of rituximab in patients with primary Sjögren syndrome have emerged in the literature. In a double-blind, randomized, placebo-controlled trial, Meijer et al found that rituximab significantly improved saliva flow rate, lacrimal gland function, and other variables in patients with primary Sjögren syndrome. In an open-label clinical trial, modest improvements were noted in patient-reported symptoms of fatigue and oral dryness. However, no significant improvement in the objective measures of lacrimal and salivary gland function was noted, despite effective depletion of blood B cells. In a randomized, placebo-controlled, parallel-group study of 120 patients with primary Sjögren syndrome, treatment with rituximab did not alleviate disease activity or symptoms at week 24, although it did alleviate some symptoms at weeks 6 and 16. Rituximab appears promising in the treatment of vasculitis and intravenous immunoglobulin (IVIG)–dependent ataxic neuropathy. Results from the AIR registry (French) indicated that rituximab appears to be effective in cryoglobulinemia or vasculitis-related peripheral nervous system involvement in primary Sjögren syndrome. In a prospective study of 78 patients with primary Sjögren syndrome treated with rituximab, significant improvement in extraglandular manifestations was reported, as measured by EULAR [European League Against Rheumatism] Sjögren Syndrome Disease Activity Index (ESSDAI) (disease activity score) and overall good tolerance reported. Several smaller studies of rituximab revealed improvement of arthralgias, regression of parotid gland swelling, and improvement of immune-related thrombocytopenia. Of the TNF inhibitors, both etanercept and infliximab have failed to demonstrate significant benefit in Sjögren syndrome. Combination therapy with leflunomide and hydroxychloroquine resulted in a significant decrease in ESSDAI scores and caused no serious adverse events, in a small phase 2a randomized clinical trial from the Netherlands. At 24 weeks, the mean difference in ESSDAI score in the leflunomide-hydroxychloroquine group (n=21), compared with the placebo group (n=7), was –4.35 points after adjustment for baseline values. Fewer data are available with regard to the role of anti-CD22, anti-BAFF, anti-IL-1, type 1 interferon, and anti-T–cell agents in treatment of primary Sjögren syndrome, with further investigations ongoing. The overall paucity of evidence in therapeutic studies in primary Sjögren syndrome suggests that much larger trials of the most promising therapies are necessary. The investigators concluded that further evaluation of leflunomide–hydroxychloroquine combination therapy in larger clinical trials is warranted.

Emergency department care

The diagnosis of Sjögren syndrome can be made from the ED if the index of suspicion is high. Patients may present with mild symptoms (eg, eye grittiness, eye dryness or discomfort, dry mouth, recurrent caries). Bilateral parotid gland swelling is also a common presentation. Patients with known Sjögren syndrome should not be taken lightly for their complaint of dry eyes or dry mouth, as these chronic problems can be very distressing and obtrusive.

Inpatient care

Give attention to artificial lubricants and humidified oxygen for intubated and/or sedated patients with Sjögren syndrome.

Outpatient care

Encourage patients with Sjögren syndrome to be active. In addition, patients should be encouraged to avoid exacerbation of dryness symptoms (eg, through smoking or exposure to low-humidity environments). All patients with Sjögren syndrome should be monitored by an ophthalmologist and dentist, in addition to their rheumatologist. Certain patients may be candidates for punctal occlusion, which is usually performed by an ophthalmologist.

Monitoring

Most patients with Sjögren syndrome can be monitored at follow-up visits every 3 months and, if the patient is stable, up to every 6 months. Patients with active problems or in whom an emerging associated illness is a concern can be seen as often as monthly.

INTERPRETATION

EXTRACTABLE NUCLEAR ANTIGEN

USAGE

Extractable Nuclear Antigens (ENAs) are over 100 different soluble cytoplasmic and nuclear antigens. They are known as "extractable" because they can be removed from cell nuclei using saline and represent six main proteins: Ro, La, Sm, RNP, Scl-70, Jo1. Most ENAs are part of spliceosomes or nucleosomes complexes and are a type of small nuclear ribonucleoprotein (snRNPS). The location in the nucleus and association with spliceosomes or nucleosomes results in these ENAs being associated with additional RNA and proteins such as polymerases. This quality of ENAs often makes it difficult to purify and quantify their presence for

Clinical Applications

An extractable nuclear antigen panel, or an ENA Panel, tests for presence of autoantibodies in the blood that react with proteins in the cell nucleus. Usually done as a follow up to a positive antinuclear antibody (ANA) test and one is showing symptoms of an autoimmune disorder. The ANA tests for the presence or absence of autoantibodies, while the ENA panel evaluates which proteins in the cell nucleus the autoantibodies recognize. The ENA panel helps diagnosis, distinguish between, and monitor the progression of autoimmune diseases and is performed with a simple blood draw. While the levels of autoantibodies may fluctuate through one's life, once you develop autoantibodies, you will always have them. Autoantibodies to these antigens are associated with particular connective tissue disorders. Indeed, in 84.3% of positive anti-ENA samples, ANA reagents were also found. The use of anti-ENA autoantibody tests can serve as additional verification of an autoimmune disorder, because a positive ANA test alone does not suffice for diagnosis. In fact, low levels of ANAs can be found in healthy patients. The applications of anti-ENA testing varies from excluding patient groups from specific groups, connective tissue diseases, and to monitor disease activity. In essence, it allows clinicians to exclude specific autoimmune disorders if a particular autoantibody is not present, and allows clinicians to track progression of a disease if the levels of these autoantibodies increase or decrease. To confirm the presence of anti-ENAs, it is currently recommended to use two or more methods to confirm anti-ENAs to avoid false positives. The diagnosis of autoimmune connective tissue diseases (CTDs) is done through analysis of clinical symptoms and signs, but also through the identification of the autoantibodies directed against nuclear antigens. A 2002 paper also seeks to compare the diagnostic tests used in immunology laboratories to measure anti-ENAs and ways to improve this testing and reporting. Double immunodiffusion (DID) and counterimmunoelectrophoresis (CIEP), two forms of gel-based techniques, are used to gain information on the clinical significance and the role of these antibodies in those with CTDs.

Techniques

Gel-Based

Since the discovery of ENAs, they have been used as a diagnostic tool in connective tissue disease. Two widely used gel-based techniques were used to identify anti-ENAs and their associations to disease in early work, double immunodiffusion (DID) [1] and counterimmunoelectrophoresis (CIEP). Both of these techniques require the precipitation of antigens for valid results. Depending on the anit-ENA being investigated, one technique may be used over the other. For example, Scl-70 antigen is less negatively charged, which can result in the antigen traveling in the same direction as the antibody. This would result in the antibody-antigen complex not precipitating; leading to invalid results. In addition, some anti-SS-B antibodies commonly identified in Sjögren syndrome may not be detected with this method. However, this method is economically feasible and specific to confirm a diagnosis. There are two sensitivities to note when viewing data from these gel-based techniques, assay sensitivity and disease sensitivity. Assay sensitivity is the ability to recognize when an antibody is present, while disease sensitivity is the ability to recognize the frequency in which the antibody occurs in a disease. Due to limitations of gel-based techniques in disease sensitivity, other techniques have been explored in order to increase assay sensitivity without decreasing disease sensitivity. For example, in patients with Systemic lupus erythematous (SLE), only 8-40% have detectable anti-SM when using gel-based assays. CIEP has been shown to be more sensitive than DID.

Hemagglutination, ELISA, Western Blot

Three additional techniques, passive hemagglutination, enzyme linked immunosorbent assay (ELISA), and western blotting (WB), can be used in order to identify ENAs and link them to specific diseases. Passive hemagglutination was popular in the late 1970s, but very few studies have been done using them and was restricted to anti-Sm and anti-ribonuclear protein (RNP) antibodies. Enzyme linked immunosorbent assay (ELISA) has become the most widely used technique for testing for anti-ENAs due to them being simple to perform, quantitative, and high volume output. While this method has increased assay sensitivity and is efficient for high volume labs, they have a much lower disease specificity than alternative techniques. This is due to the inability to properly isolate the ENAs without large costs due to their association with complexes in the nucleus of the cell. Another worry with the ELISA technique is that anti-Sm antibodies have been reported in patients without SLE which would lead to over-investigating, but could be due to the quality of the antigen source used. Western blotting has a major disadvantage in that antibodies targeted against conformational epitopes can not be detected. On top of that, false positive can occur and the disease specificity is lower than other techniques. While each technique has their advantages and disadvantages, ELISA has the least severe disadvantages of potential for false positives (which are less dangerous than false negatives) and expensive. Many labs use a combination of both of these techniques to improve efficiency without sacrificing specificity. The current recommendation by European Consensus workshops is to screen for positive anti-ENAs with the ELISA technique. A more specific test such as CIEP will follow with samples that are identified as positive. The six main antigens used in immunological laboratories for detection are Ro, La, Sm, RNP, Scl-70 and Jo1, which are screened for by Ouchterlony double immuno diffusion techniques and confirmed by immunoblotting. On anti-nuclear antibody tests, these antigens have a speckled pattern.

Terminology

ENAs originally referred to proteins found in a saline extract of cell nuclei. Components have since been more clearly identified and in fact include many cytoplasmic molecules. The misnomer, however, has remained. These proteins are intimately associated with various RNA molecules and are thus called Ribonucleoproteins, but the nomenclature used for them is often a source of confusion, Sm, Ro and La were named after the first 2 letters of the surnames of the patients in whom they were first found. Two proteins associated with Sjogren's Syndrome were independently described as antigens A and B, but are now known to be identical to Ro and La respectively. i.e. SS-A = Ro and SS-B = La. ENA (extractable nuclear antigen) panel tests, test for autoantibodies to proteins in the cell nucleus. The term "extractable" is derived from the ability to remove the autoantibodies from the nuclei with saline and common proteins. The method of identifying these specimens is why they are also referred to as Antibodies to Saline-extracted Antigens.

ENA

Anti-ENA is a grouping of antibodies often used to screen for mixed connective tissue disease (MCTD), Sjögren's syndrome and systemic lupus erythematosus and commonly is composed of six tests:

- anti-Sm (for SLE)
- anti-RNP (for MCTD)
- anti-La/anti-SS-B (for Sjögren's)
- anti-Ro/anti-SS-A(for Sjögren's)
- anti-Scl70 (for Scleroderma)
- anti-Jo (for Dermatomyositis)

Sensitivity and specificity of these tests depends on the type of assay employed, and will therefore vary by lab. The following table illustrates the sensitivity and specificity of ENA antibodies at detecting SLE with the ELISA technique.

Antibody (tested using ELISA)	Specificity (%) for SLE	Sensitivity (%) for SLE
Anti-Ro (SS-A)	61	80-93
Anti-La (SS-B)	27-35	88-97
Anti-Sm	34-45	88-100
Anti-RNP	39-64	84-97
Reference for all values:		

In addition, the use of ENA testing has also been used for the study of wheat related disorders such as celiac disease. A study conducted in 2018 screened patients with wheat related disorders for 10 anti-ENA antibodies.

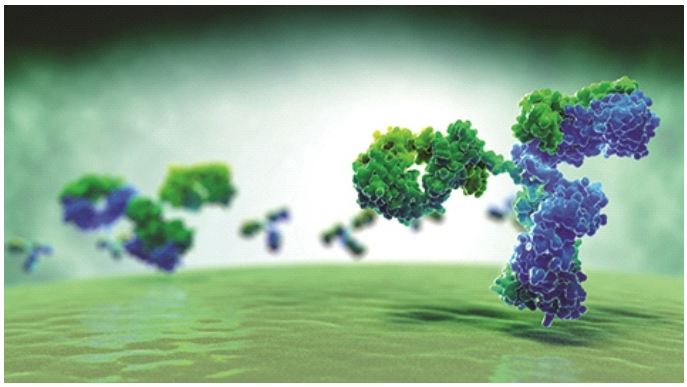
- SSA (Ro)
- SSB (La)
- RNP/Sm
- Jo-1
- Sm
- Scl-70
- Chromatin
- Centromere
- Histone
- RNA polymerase III

73% of celiac disease subjects tested positive for anti-histone and was the most prevalent, which is typically associated with Drug-induced lupus erythematosus. This implicates a high probability of an autoimmune disorder in patients with wheat-related disorders.

TROUBLESHOOTING

ANTINUCLEAR ANTIBODIES

Laboratories must consider several key factors before deciding which method is best for their patients and staff



Imagine your lab has decided to take the plunge and implement antinuclear antibody (ANA) testing in house, taking it off the send-out menu. You might first ask, What is the best method for ANA testing? Or, what if your lab already performs ANA testing, but the expert technologist who has been reading ANA indirect immunofluorescence (IIF) slides for 30 years has just announced that she is going to retire. This might prompt you to ask, Is it time for us to move from IIF ANA testing to a newer methodology? These are important and relevant questions, but without easy answers. This review aims to provide practical information on ANA testing methodologies, including their diagnostic utility and performance characteristics.

ANA TESTING HISTORY AND CONTEXT

ANAs refer to a collection of autoantibodies that target a variety of nuclear and cytoplasmic antigens. First described more than 50 years ago, ANAs remain the most sensitive serologic mar-er for evaluating patients with suspected connective tissue diseases (CTDs), also referred to as ANA-associated rheumatic diseases (AARDs) (1). The diagnostic potential of ANAs originated with the discovery of LE cells, described as mature polymorphonuclear leukocytes containing phagocytosed nuclear material. LE cells were so-named because they were found only in patients with systemic lupus erythematosus (SLE). LE cells could be produced in vitro by taking patient plasma and mixing it with peripheral blood from healthy controls that had been “damaged” by vortexing with glass beads. Ultimately, research demonstrated that immunoglobulin from patient plasma was binding to nuclei from the “damaged” peripheral blood, which neutrophils in turn phagocytosed. IIF was used to further characterize this immunoglobulin, demonstrating its specific binding to cellular nuclear material. This immunoglobulin is what we now know as the ANA. ANA testing generally involves two parts (2). First, for patients with a suspected AARD, a screening ANA is ordered to detect the ANA regardless of the antigen specificity. Second, for patients with positive screening assay results, additional tests characterize the antigen specificity of their ANA. Identifying the antigen specificity has important diagnostic and prognostic implications for patients. Although dozens of antigens have been associated with ANAs, only a small number are available for routine clinical testing. Depending on a patient's clinical scenario, a positive ANA may require testing for anti-double standard DNA antibodies, antibodies against one or more of the extractable nuclear antigens (SS-A, SS-B, Sm, Scl-70, Jo-1, and RNP), anti-ribosomal P antibodies, or anti-centromere antibodies.

METHODOLOGIES FOR ANA TESTING

Three primary methods are available to clinical laboratories as screening ANA tests: IIF, enzyme immunoassay (EIA), and multiplex immunoassay (MIA). IIF detects antibodies that bind to a tissue substrate which, for ANAs, is usually fixed HEp-2 cells. IIF accomplishes this detection with a fluorescently labeled anti-human immunoglobulin. With EIA, an antigen mixture adhered to a solid surface (usually a 96-well plate) takes the place of the HEp-2 cells, and detection occurs through an enzyme-labeled anti-human immunoglobulin. MIAs are based on polystyrene bead sets distinguished from one another based on their fluorescent signature. Each bead set is conjugated to a known ANA antigen, and the different sets are then combined into a bead cocktail. A patient sample is added to the bead cocktail, and binding of a patient antibody to any of the beads is accomplished with a fluorescently labeled anti-human immunoglobulin.

Reporting of ANA Test Results

From a physician's perspective, one of the most obvious differences between ANA screening methods is how results are reported. In most cases, MIAs are reported qualitatively as “ANA positive” or “ANA negative,” with screen results being based on the collective assessment of all the individual antigen specificities included in an assay. If all the included antigen specificities are negative, then the ANA screen is interpreted as negative. Conversely, if one or more of the beads show fluorescence exceeding a certain threshold, a sample would be identified as positive. Importantly, for ANA positive samples, the identities of the antigen specificities are not revealed to the laboratory and thus are not reported to patients' medical records. If a clinician wants to determine the antigen specificity of a patient's ANA, he or she would need to order the clinically relevant tests. In contrast, most EIAs are reported as a numeric value with an arbitrary unit of measurement. There is no traceable standard for these assays, so each manufacturer establishes the units and analytical measuring range for its tests. EIAs' quantitation is based on light absorbance. The enzyme linked to the detection antibody converts a colorless substrate to a colored product, the absorbance of which is compared to a standard curve. Manufacturers will provide a recommended cutoff, which is the unit value above which a sample would be considered “ANA positive”. As with MIAs, a positive EIA result does not reveal the antigen specificity of the ANA, and further testing would be necessary if a clinician wants to know those details. ANA by IIF is generally reported with both a titer and a pattern. Labs screen all samples initially at a single dilution, usually 1:40 or 1:80. Any sample identified as positive at the screening dilution is titered out either to endpoint or to a pre-defined dilution, depending on the laboratory's preference. The titer is determined by serial dilution, with the reported titer being the last dilution for which the IIF would be identified as positive. The pattern interpretation is based upon recognition of specific cellular features to which a patient's antibody has bound. Because IIF pattern interpretation is based on visual interpretation, standardization in reporting has been a challenge. The International Consensus on ANA Patterns (ICAP), a subcommittee of the Autoantibody Standardization Committee, promotes discussion and generates consensus regarding the morphologic features associated with specific ANA patterns (4). ICAP has also made recommendations

regarding how laboratories should report ANA patterns. The group has defined six nuclear patterns as "Competent-Level": homogeneous; speckled; dense fine speckled (DFS); centromere; discrete nuclear dots; and nucleolar. ICAP recommends that any laboratory performing ANA by IIF should be able to accurately and reproducibly identify these patterns. The remaining nuclear patterns are designated as "Expert-Level" and might be recognizable only by individuals with particular expertise in IIF analysis.

ANA CLINICAL SENSITIVITY AND SPECIFICITY

When considering which ANA test to implement, understanding each method's clinical sensitivity and specificity is critical. Many studies have compared the clinical sensitivity and specificity of the different methods. Because IIFs, EIAs, and MIAs report results so differently, these studies have focused primarily on qualitative agreement. Although seemingly very straight-forward, these types of comparisons are more difficult than they appear, largely because estimated sensitivities and specificities and the agreement between methods is heavily dependent on the cutoffs used to differentiate between positive and negative. Historically, IIF has been considered the most sensitive method for identifying patients with AARDs. In a 2009 position statement on ANA testing methods, the American College of Rheumatology identified IIF as the "gold standard for ANA testing" primarily based on its high sensitivity (>95%) for the diagnosis of SLE (5). However, the statement also acknowledges that the specificity of ANA by IIF is a limitation. In a cohort of patients for whom ANA testing was ordered as part of routine clinical care, we demonstrated that IIF at a titer cutoff of 1:40 had a sensitivity of 94% for the general diagnosis of AARDs (6). This was higher than the sensitivity of either EIA or MIA, at 74% and 67%, respectively. However, the IIF's higher sensitivity was at the expense of specificity, which, at the 1:40 cutoff, was only 43%. In comparison, the corresponding EIA and MIA specificities were 80% and 87%, respectively. When we increased the cutoff for IIF to 1:80, the specificity improved to 62% but the sensitivity decreased to 84%. Some data suggest that the titer of the ANA may help in distinguishing between patients with and without AARDs. In a study from 2011, Mariz et al. demonstrated that 45.8% of positive ANAs in healthy controls had a titer of 1:80, while 88.5% of ANA-positive AARD patients had an ANA titer ≥1:320 (7). Many laboratories that perform ANA by IIF are moving away from screening at the 1:40 dilution, opting for improved specificity even with some loss in sensitivity. When labs use higher screening dilutions, the sensitivities of IIFs are on par with those of EIAs and MIAs. Although IIFs have the capability of maximizing sensitivity, from a practical perspective, EIAs and MIAs provide a good balance of sensitivity and specificity. IIF's sensitivity is attributed to its broad antigen specificity. This method detects antibodies against any of the hundreds of nuclear and cytoplasmic antigens present in a cell. However, not all antigen specificities are relevant for the diagnosis of AARDs. For example, the DFS pattern appears almost exclusively in patients with no evidence of an AARD (7). It has been suggested that the presence of the DFS pattern could be used to rule out an AARD in an individual with a positive ANA. The antigen specificity associated with this pattern has been identified as lens epithelial-derived growth factor, also referred to as DFS70 (8). Further studies have confirmed that monospecificity for DFS70 in the context of a DFS pattern is not consistent with an AARD. This pattern, and perhaps others like it that have yet to be characterized, may help to address some of the specificity challenges associated with ANA testing by IIF.

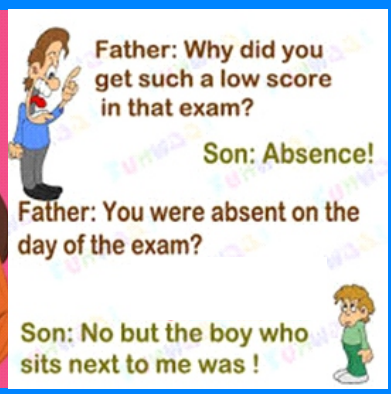
PERFORMANCE CONSIDERATIONS FOR ANA METHODOLOGIES

When labs are considering which ANA method to implement, availability of a qualified technologist to perform the testing is likely a significant concern. Other key considerations include throughput, workflow, and automation of a method. Although automation of immunological testing has not reached the level of chemistry platforms, significant strides have been made over the last decade, particularly with EIAs and MIAs. EIAs can be performed manually, although more often than not, labs perform this testing on semi-automated or automated platforms. The semi-automated platforms may dilute patient samples and add reagents to the plate, but a technologist's intervention might be required to wash and move the plate to an absorbance reader. A fully automated system processes an EIA in its entirety, only requiring technologists to load samples and reagents. Most MIA systems are also fully automated. In addition, MIAs have the advantage of being random access, which facilitates improved workflows. In contrast, EIAs are batched, which, for labs with lower volumes of ANA orders, could have a negative impact on workflow and on turnaround times. Another advantage of MIA systems is they offer labs the opportunity to expand their test menus. Most MIA systems are not limited to ANA testing, and have reagents available for other autoimmune conditions (celiac disease, antiphospholipid syndrome, and vasculitis) and for infectious diseases (Epstein-Barr virus, HIV, and herpes simplex virus). Being able to perform additional testing and maximize an instrument's utilization could make an MIA system an attractive option. Historically, IIF has been the ANA method requiring the most clinical technologist resources and expertise, with automation limited to dilution of patient samples and perhaps addition of sample and reagents to slides. In addition, slide reading was a manual process that relied on experienced technologists to interpret numerous complex patterns. Now, however, systems are available that automate almost the entire process, from slide processing to reading. Processing the slides includes not only sample and reagent pipetting but also slide incubation and washing. After processing, the slides can be moved to an enclosed microscope with a high-resolution digital camera, which obviates the need for a darkroom. This means such systems can be used on a bench in an open laboratory. Cameras in these newer IIF systems capture several digital images from different areas of slides. The fluorescence intensity of the stain is measured, and values above a certain cutoff are considered positive. For samples identified as positive, the computer algorithm reads the pattern of and interprets the fluorescence intensities in the context of known ANA patterns. Although this step automates the previously manual process of slide reading, final qualitative and pattern interpretation still requires a technologist's expertise. For each sample, a technologist must confirm the computer-generated result. If he or she disagrees, the result can be changed. Most automated readers recognize the common ANA patterns, and some identify certain mixed patterns. More complex patterns unidentifiable by the computer still require a technologist's interpretation. Some automated readers not only automate pattern interpretation at least partially but also estimate titers. These instruments use the fluorescence intensity of an image to estimate a sample's titer rather than relying on serial dilutions. This can be accomplished either from a single patient dilution or a limited number of dilutions. As with pattern interpretation, an estimated titer can be replaced with a titer from serial dilutions, depending on the pattern and the technologist's judgment. Overall, although not completely automated by chemistry standards, the availability of automation for IIF, EIA, and MIA gives labs several options for complex ANA testing in a time of shrinking resources.

BOUQUET

Wisdom Whispers

In Lighter Vein



Brain Teasers

1. **In SLE which of the following antibodies is usually decreased?**
   A. IgG
   B. IgM
   C. IgA
   D. All are usually increased

2. **In Hodgkin's disease which of the following antibodies is decreased?**
   A. IgG
   B. IgM
   C. IgA
   D. All are usually normal

3. **Which of the following is useful to stimulate antibody production?**
   A. An adjuvant
   B. A hapten
   C. Antiserum
   D. Purified antigen
   E. Crude antigen

4. **Complement component C3 is cleaved by:**
   A. C3b
   B. C3bBb
   C. Factor B
   D. Factor D
   E. Factor H.

ANSWER: 1: D, 2: D, 3: A, 4: B

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