The Laryngoscope

The Minor Salivary Gland Biopsy as a Diagnostic Tool for Sjogren Syndrome

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Objectives/Hypothesis: In suspected cases of Sjogren syndome (SS), patients are often referred for a labial minor salivary gland biopsy. However, studies have shown this test to be unreliable. Pathologic misinterpretation and immunosuppressive medications may affect the results of the biopsy. As a result, it is best to perform this procedure only when necessary. The purpose of the current study was to review clinical signs and symptoms of patients who underwent a lip biopsy to determine which patients benefited most from this procedure.

Study Design: Retrospective review.

Methods: A retrospective chart review of patients referred to otolaryngology for a lip biopsy for the diagnosis of SS.

Results: Joint pain, salivary gland swelling, and abnormal serology (anti-Sjogren syndrome A/anti-Sjogren syndrome B) were more prevalent in the positive lip biopsy group (grade = 3 or 4). Out of the 12 patients who had both sicca symptoms and positive serology, nine (75%) had a grade = 4. Presence of sicca symptoms and positive serology were predictive of a positive biopsy ($P = .017$). Excluding those patients who were on immunosuppression for more than 6 weeks prior to the biopsy, the correlation became stronger ($P = .011$).

Conclusions: In this study, clinical presentation of sicca symptoms and positive serology reliably predicted the results of a lip biopsy. The results of this study suggest that patients with clear criterion for SS may not require a lip biopsy, especially those patients on immunosuppression. When physicians suspect SS, a thorough clinical and laboratory examination is necessary to determine if a patient will benefit from a minor salivary gland biopsy.

Key Words: Sjogren syndrome, lip biopsy, minor salivary gland biopsy, xerostomia.

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INTRODUCTION

Sjogren syndrome (SS) is an autoimmune disorder usually characterized by xerostomia and xerophthalmia. However, the clinical presentation of SS can be highly variable with initial symptoms, ranging from the classic sicca presentation to atypical features such as peripheral neuropathy, cystitis, pulmonary insufficiency, and a host of other system manifestations. The pathological mechanisms for these protean symptoms are currently not well understood, and this disease remains a diagnostic and treatment challenge for physicians across many fields.

Although the labial minor salivary gland biopsy is a commonly used diagnostic tool for SS,[1–4] several studies have questioned its utility based on the invasiveness of the procedure and the high rate of pathologic misinterpretation.[5,6] Studies have stated that the lip biopsy is a simple and safe tool[7]; however, investigators have not determined when the procedure is necessary. Although it is true that some diagnoses of SS may rely on the results of a lip biopsy, the test is not required to make a diagnosis. Physicians are often faced with a decision to determine which of a heterogeneous patient group require biopsies. The study described herein was undertaken to examine the University of Chicago's experience with the lip biopsy as a diagnostic tool for SS. A blinded reevaluation of lip biopsies performed at this institution was completed in an earlier study, and this current review of the clinical presentation of those same patients is an extension of the original pathological study.[8] The purpose of the current study was to review clinical signs and symptoms of patients who underwent a lip biopsy to determine which patients would most benefit from this procedure.

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MATERIALS AND METHODS

Subjects

Patients who underwent a lip biopsy for a possible diagnosis of Sjogren syndrome (SS) at the University of Chicago hospitals between November 2002 and November 2004 were identified from hospital records using the American Medical Association Current Procedural Terminology (CPT) code for "biopsy of lip."[8] Those who obtained a lip biopsy for reasons other than SS were excluded. This study was approved by the institutional review board of the University of Chicago Medical Center and Biological Sciences Division.

Clinical Examination

Patients with symptoms suggestive of keratoconjunctivitis sicca (KCS) and xerostomia were assessed for extraglandular manifestations of SS, such as neuropathy, joint pain, and other connective tissue diseases at the Section of Rheumatology at the University of Chicago. Ophthalmology evaluation was obtained for patients with KCS.

Following a complete rheumatologic examination, patients were referred to otolaryngology for evaluation and a biopsy of the lower lip. The biopsy was performed under local anesthesia. A small ellipse of the lower lip mucosa was incised, and a small sample of submucosal tissue and several minor salivary glands were removed (Fig. 1). The biopsy was processed routinely in hematoxylin and eosin stain, examined by standard light microscopy, and assigned a positive, negative, or nondiagnostic result.

In a previously completed study, these biopsy specimens were reviewed and regraded under the currently accepted grading system.[2,3] The blinded grades were then compared to the results of the initial pathology report to determine accuracy of the original grades. The biopsy specimens were also assessed for levels of atrophy. The patient groups in this current study are based on the grades assigned to the biopsies during the blinded grade.[8]

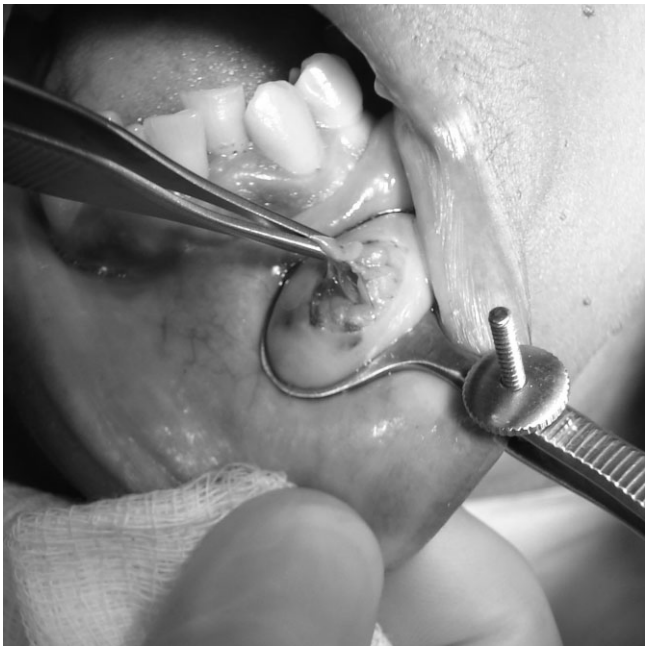

Fig. 1. Minor salivary gland biopsy.

Data Collection and Statistical Analysis

Medical history, rheumatology clinic notes, serologic tests (anti-SSA, anti-SSB, and antinuclear antibody [ANA]), minor salivary gland biopsy results, and presence of sicca symptoms, salivary gland enlargement, joint pain, peripheral neuropathy, and any other organ system involvement were extracted from medical charts. All of the data were entered into a Microsoft Excel database (Microsoft Corporation, Redmond, WA). All identifying information was removed from the database to insure personal health information protection.

Patients were grouped by the results of their lip biopsies. The clinical presentation and laboratory findings between the groups were compared to better characterize those patients who were more likely to have a positive minor salivary gland biopsy. Patients were also grouped by presence of anti-SSA antibodies, and the percentage of atrophy within their respective lip biopsies was compared.

Groups of patients were compared with Fisher exact tests and two sample t tests with unequal variances on STATA (Statacorp, College Station, TX).

RESULTS

Forty-nine patients were identified by the CPT query of the University of Chicago Hospitals between November 2002 and November 2004. Out of these patients, 46 patients were referred from physicians in the section of rheumatology for the purpose of evaluating for SS. One patient who had been referred for excisional biopsy of a lower lip mucocele was excluded. Another patient was diagnosed with mucosa-associated lymphoid tissue lymphoma and was removed from further analysis. Of the remaining 47 patients, one included in the first part of this study was not included in the present study due to incomplete clinical data.

Of the 46 patients included in the study, 41 (89%) were female and 5 (11%) were male. The mean age of patients was 49 years old (range, 20–85). The most common clinical symptoms were joint pain (40 patients), sicca symptoms (39 patients), and peripheral neuropathy (25 patients).

We previously published that 18 (39%) of these patients had a negative biopsy (grade <3) and 28 (61%) had a positive biopsy (grade $= 3$ or 4).[8] The clinical features between the two groups are summarized in Table I, and there were no significant differences between age, sex, or race between them. There were no statistically significant differences in clinical presentation between the two groups, but some characteristics followed trends. Joint pain (93% vs. 77%; $P = .191$), salivary gland swelling (21% vs. 6%; $P = .219$), and bladder involvement (43% vs. 17%; $P = .107$) were present more often in the positive lip biopsy group (grade $= 3$ or 4). The incidence of sicca symptoms, peripheral neuropathy, and extraglandular organ involvement were similar between the two groups.

When using a higher grade (grade $= 4$) as positive, salivary gland swelling was more prevalent in the lip biopsy positive group (30% vs. 4%; $P = .033$), but there were no other clinical differences between the two groups.

Serologic abnormalities, including ANA $>$1:80, anti-SSA $>$19, and anti-SSB $>$19, are summarized in Table I. Prevalence of an elevated ANA was similar between the positive and negative groups. Presence of elevated anti-SSA (36% vs. 11%; $P = .089$) and anti-SSB (21% vs. 6%;

TABLE I.
Demographic, Clinical, and Serologic Features of 46 Patients Undergoing Minor Salivary Gland Biopsy, Grouped by Grade.

	Grade = 3 or 4, n = 28	Grade < 3, n = 18
Age	50	48
Caucasian, %	61	72
Women, %	89	89
Sicca, %	82	89
Joint pain, %*	93	77
PNS, %	57	50
CNS, %	7	6
Salivary gland swelling, %†	21	6
Pulmonary involvement, %	7	6
Cardiac involvement, %	7	0
GI involvement, %	14	0
Kidney involvement, %	11	0
Bladder involvement, %‡	43	17
Thyroid disease, %	0	6
Anti-SSA antibodies, %§	36	11
Anti-SSB antibodies, %‖	21	6
ANA>1:80, %	79	72

PNS = peripheral nervous system; CNS = central nervous system; SSA = Sjogren syndrome A; SSB = Sjogren syndrome B; ANA = antinuclear antibody.
*$P = .191$.
†$P = .219$.
‡$P = .107$.
§$P = .089$.
‖$P = .219$.

$P = .219$) antibodies were more prevalent in patients with positive lip biopsies (grade = 3 or 4). When using a greater grade (grade = 4) as positive, the correlation of anti-SSA and anti-SSB antibodies with a positive lip biopsy became stronger (SSA: 45% vs. 12%; $P = .017$; SSB: 30% vs. 4% $P = .033$).

The 12 patients with an abnormal anti-SSA level had on average 16.3% of the biopsy lobules atrophic compared to the 34 patients with a normal anti-SSA level who had on average 6.08% of their lobules atrophic (Table II). However, this was not a statistically significant difference between these two groups ($P = .16$).

Those patients that had both sicca symptoms and positive serology (elevated anti-SSA or anti-SSB) were more likely to have a strong positive lip biopsy (grade = 4) (Fig. 2) ($P = .017$). Out of the 12 patients who had both sicca symptoms and positive serology, nine (75%) had a grade of 4. Of the 34 patients who did not have both sicca symptoms and positive serology, 21 (67%) had a grade ≤ 3. Excluding those patients who were on corticosteroids (>10mg) for more than 6 weeks prior to the biopsy, the correlation became even stronger. Out of the 11 patients who had both sicca symptoms and positive serology, nine (82%) had a grade of 4. Of the 27 patients who did not have both sicca symptoms and positive serology, 18 (67%) had a grade ≤ 3 (Fig. 2) ($P = .011$).

TABLE II.
Association of Positive Anti-SSA With Percentage of Lobules Atrophied.

	Patients With Positive SSA	Patients With Negative SSA
No.	12	34
Average lobules atrophied	16.3%	6.08%
Standard error	.066	.012
P	.16	

SSA = Sjogren syndrome A.

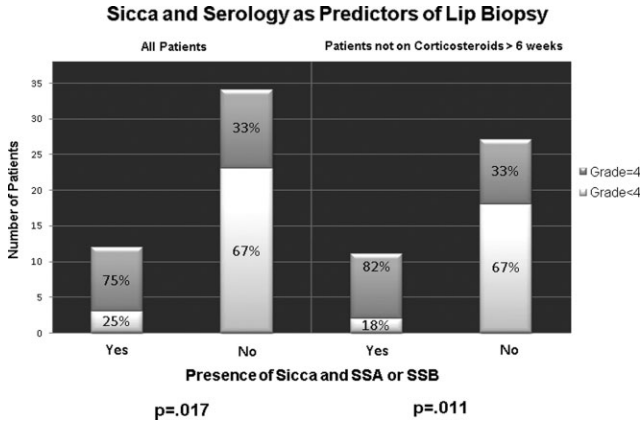

Fig. 2. Sicca and serology as a predictor of a lip biopsy when grade >3 is positive.

DISCUSSION

Determining which patients require a lip biopsy can be difficult for physicians. In this study, the best predictor of biopsy results was presence of sicca symptoms and a serological abnormality of either anti-SSA or anti-SSB antibodies. Patients with this typical presentation of SS do not derive additional benefit from a lip biopsy, and this procedure should be reserved for diagnosing patients with heterogeneous symptom complexes and equivocal or negative serological testing.

Those cases that are challenging to diagnose lack the classic SS presentation of sicca, but include other systemic symptoms, such as peripheral neuropathy and joint pain with positive SS serology (SSA or SSB). These patients with other manifestations of SS, such as neurological symptoms, may have an early stage of SS, with other more typical symptoms not yet present.[9] A lip biopsy in such cases would be appropriate and may help establish a diagnosis and treatment. If a lip biopsy were negative or inconclusive in such a case, a physician would have to use clinical judgment whether to pursue SS treatment or another diagnosis. The American-European classification criteria require either a positive lip biopsy or positive SS serology for an official diagnosis of SS. In the event of a negative biopsy, positive SS serology may direct a clinician toward SS treatment, whereas negative SS serology may suggest other diagnostic workup.

Wise et al.[10] and Shah et al.[11] found that serological markers are better predictors of lip biopsies than clinical symptoms. However, it is difficult for physicians to diagnose SS through serology alone. Of those in this study, 53% with normal anti-SSA and anti-SSB antibodies had

a positive lip biopsy, which demonstrates how this is not a perfect marker. Taking both symptoms and serology into consideration is more likely to yield an accurate clinical picture than either one alone.

The decision to biopsy a patient can often be blurred by a host of clinical situations. One example is the utility of a lip biopsy of a patient taking immunosuppressive medications. When excluding patients on long-term corticosteroids from the analysis of this study, there was a stronger correlation between the lip biopsy and clinical presentation of sicca with positive serology, suggesting that corticosteroids may have a tendency to confound biopsy results. Unfortunately, data on the effect of medications on lymphocytic infiltrate is limited. Zandbelt et al.[12] presented a unique case study in which the use of high dose corticosteroids not only relieved a patient's symptoms of SS, but also decreased the lymphocytic infiltrate of a second minor salivary gland biopsy. To avoid the confusion of false negative, clinicians should be wary of performing a lip biopsy of patients on immunosuppression with clear criterion for SS.

Chronic SS with subsequent development of atrophy and fibrosis of the salivary glands may also distort the results of the lip biopsy. Greenspan et al. noted that there was a correlation of salivary gland acinar depletion with increased grade, and that it is possible to see more healing and fibrosis in chronic cases.[3] Patients with longstanding sicca symptoms from SS may not benefit from a lip biopsy if their inflammatory response has already run its course. Although there is no marker for chronic SS, elevated values of anti-SSA antibodies have been implicated in pathogenic fibrosis in systemic lupus erythematous and congenital heart block.[13–15] Anti-SSA antibodies are a specific marker for SS, but its role in pathogenesis of SS is unclear. In this study, those patients with an elevated anti-SSA level had a larger degree of atrophy on their lip biopsies. It is possible that a similar relationship exists between anti-SSA and salivary gland fibrosis and atrophy often seen in lip biopsies of patients with SS. More investigation is required to validate the association of elevated anti-SSA antibodies with a higher degree of atrophy in lip biopsies and to determine if this is a possible marker for longstanding disease.

SS can often have systemic involvement, but the location and degree varies among patients. Bladder symptoms, such as urgency and frequency, can be symptoms of SS, but are less common than others such as joint pain and peripheral neuropathy. An interesting finding in this study was a prevalence of bladder symptoms in the positive lip biopsy group, but this association disappeared when a stronger criterion was used for a positive lip biopsy (grade = 4). The high incidence of bladder symptoms when using grades 3 and 4 as positive is most likely a result of the small patient population of the study. The systemic involvement of SS is unpredictable and can not be an official diagnostic criterion. However, clinicians should consider that a complex case that includes sicca symptoms with an extraglandular complaint may be unified by an underlying diagnosis of SS.

The greatest limitation of this study was the small patient population available. We believe that the major differences and trends found in this study will become more apparent and reliable in a larger scale study. However, most single institutions see a limited number of possible SS cases, and performing a large scale study on minor salivary gland biopsies is difficult.

Whereas the minor salivary gland biopsy has been supported in some literature as the best diagnostic test for SS, we have noted repeated inconsistency and unreliability with the lip biopsy. The high misinterpretation rate of the biopsies performed at this institution may be common due to the rarity of lip biopsies at most institutions.[8] In this study, we wanted to determine the clinical situations when this procedure is required to limit the possible error involved. Retrospective review of the patients who were evaluated at this institution revealed cases in which clinical presentation of sicca symptoms along with serologic values of elevated anti-SSA or anti-SSB antibodies reliably predicted the results of a lip biopsy and ultimately the diagnosis of SS. In these cases, the patients could have been spared this invasive procedure due to the fact that an official diagnosis of SS does not require both positive serology and a positive lip biopsy. When physicians suspect SS, a full and thorough clinical and laboratory examination is necessary to determine if a patient will benefit from a minor salivary gland biopsy.

CONCLUSION

The lip biopsy has been a widely used test to diagnose SS. However, physicians must take into consideration the limitations of the lip biopsy. Medications, such as corticosteroids, may have a tendency to confound biopsy results, and those with longstanding sicca symptoms may have lip biopsies with atrophic glands that are difficult for a pathologist to interpret. If a clinical picture is consistent with SS, performing a lip biopsy may be unnecessary and may only lead to confusion. In an era of medicine in which physicians are becoming more reliant on technology and laboratory tests, the clinical presentation of a patient should always remain the best tool a physician has. Before ordering a lip biopsy, physicians must consider the entire clinical picture to confirm that this invasive test is truly necessary.

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