A Study to Standardize Quantitative Evaluation of Parotid Gland Scintigraphy in Patients with Sjögren's Syndrome

SUSUMU NISHIYAMA, SHOJI MIYAWAKI, and YASUHIKO YOSHINAGA

ABSTRACT. ***Objective.*** To standardize quantitative parotid gland scintigraphy for diagnosing Sjögren's syndrome (SS).

Methods. Forty-five patients with SS and 23 controls were studied. Dynamic images were obtained up to 50 min after the injection of 185 MBq ^{99m}Tc-pertechnetate and salivary excretion was stimulated with lemon juice orally at 40 min after the injection. Peak count and uptake speed in the uptake phase, and excretion speed and excretion fraction in the excretion phase were calculated.

Results. The levels of peak count, uptake speed, and excretion speed in the patients with SS were significantly lower than the levels in the controls, whereas there was no significant difference of excretion fraction level between the patients with SS and the controls. The calculations of peak count and excretion speed levels, which were closely related with the focus scores of minor salivary glands and the amount of stimulated whole saliva, showed higher reproducibility, sensitivity, and specificity than those of uptake speed and excretion fraction levels.

Conclusion. The calculations of peak count and excretion speed were eligible to standardize quantitative parotid gland scintigraphy for diagnosing SS. (J Rheumatol 2006;33:2470–4)

Key Indexing Terms:
RADIONUCLIDE IMAGING   SALIVARY GLAND   SJÖGREN'S SYNDROME

Sjögren's syndrome (SS) is an autoimmune disease characterized by lymphocytic inflammation and destruction of exocrine glands, particularly the salivary and lacrimal glands. Salivary gland scintigraphy is widely used to evaluate the salivary gland function and also used for diagnosing SS[1]. The major advantage of salivary gland scintigraphy, as compared to sialography, is that both parenchymal and excretion function of all salivary glands can be quantified simultaneously with a single intravenous injection[2]. Moreover, salivary gland scintigraphy is not as invasive as salivary gland biopsy.

In 1971, Schall, *et al*[3] built up a qualitative 4-grade system for evaluating salivary gland function, which was adopted by European and EU-US diagnostic criteria for SS[1,4]. In 1988, Sugihara and Yoshimura[5] proposed another approach for evaluating parotid gland functions classifying time-activity curves into 4 stereotypes: normal, median, flat, and sloped. Although these qualitative methods are simple, borderline data may be misclassified by the subjective judgment.

On the other hand, objective data can be obtained by adopting computer-assisted quantitative methods. Quantitative measures can be divided into the uptake phase and the excretion phase measures. Some investigators tried to establish practical measures and they showed usefulness of peak count, uptake speed, excretion speed, and excretion fraction[6-10] (Figure 1). However, they did not compare peak count and
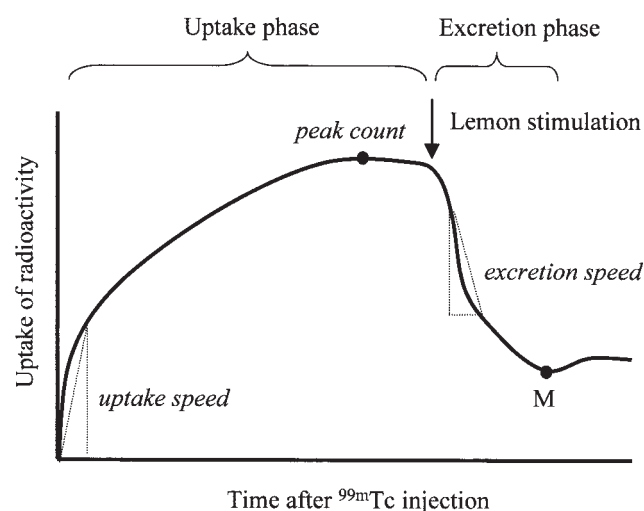
Figure 1. A typical time-activity curve and the quantitative evaluation measures. A time-activity curve of each parotid gland was drawn as described in Materials and Methods. X-axis represents time (s) after the ^{99m}Tc injection and y-axis represents uptake of radioactivity (count/s). Peak count (count/s) is the maximum count of the time-activity curve. Uptake speed (count/s^2) is expressed as the maximum incremental count per second in the uptake phase and excretion speed (count/s^2) is expressed as the maximum decrement per second in the excretion phase. Excretion fraction (%) is expressed excretion count (peak count – M) divided by peak count, where M indicates the minimum count after the lemon stimulation.

Address reprint requests to Dr. S. Nishiyama, Rheumatic Disease Center, Kurashiki Medical Center, 250 Bakuro-cho, Kurashiki, Okayama, Japan. E-mail: susumu.nisiyama@ryumachi-jp.com

uptake speed in the uptake phase or excretion speed and excretion fraction in the excretion phase from the point of view of usefulness and reproducibility.

We investigated which measures were appropriate for quantitative evaluation of salivary gland scintigraphy.

MATERIALS AND METHODS

Sixty-eight patients (66 women and 2 men) suspected of having SS underwent salivary gland scintigraphy from January 1999 to December 2003. The subjects were classified into 45 (all women) with SS according to European Criteria, excluding salivary scintigraphy from the classification items. In those patients with SS, 34 had primary SS and 11 had secondary SS (5 with rheumatoid arthritis, 4 with mixed connective tissue disease, one with polymyositis, and one with scleroderma). The remaining 23 (21 were women) did not have SS (7 with rheumatoid arthritis, 2 with scleroderma, 2 with polymyalgia rheumatica, one with systemic lupus erythematosus, 2 with malignant lymphoma, and 9 with unknown cause of dry mouth).

After the intravenous injection of 185 MBq ^{99m}Tc-pertechnetate, sequential images of 30 s each were taken for 50 min (except from 35 min through 45 min, during which images of 10 s each were acquired) with a Millennium VG gamma camera (GE Yokogawa Medical System, Tokyo, Japan), using a parallel-hole collimator in an anterior position. Stimulation for saliva excretion was carried out with 1.5 ml of lemon juice (Pokka Corporation, Nagoya, Japan) orally at 40 min after the injection. Images were stored in a 128×128 matrix. A region of interest (ROI) over the right parotid gland and the same shaped background ROI over the nearby orbita were enclosed. ROI over the left parotid gland and the counterpart background were enclosed in the same way. A time-activity curve of each parotid gland was drawn using background subtraction and 3-point smoothing. In this study, submandibular glands were not evaluated because their images were too faint to detect in some patients.

Figure 1 shows a time-activity curve and the quantitative evaluation measures. Peak count (count/s) was defined as the maximum count of the time-activity curve. Uptake speed (count/s^2) was defined as the maximum incremental count per second in the uptake phase and excretion speed (count/s^2) was defined as the maximum decrement per second in the excretion phase. Excretion fraction (%) was calculated as follows: (peak count – M)/peak count, where M indicates the minimum count after the lemon stimulation. We programmed user-protocol to have the 4 quantitative measures calculated automatically by GENIE eNTEGRA (GE Yokogawa Medical System). Four pairs of measures were calculated on bilateral parotid glands in each individual and the larger value of each measure was used for the analysis.

Focus scores of labial salivary gland biopsy were determined according to the criteria of Greenspan, et al[11]. Stimulated whole saliva was collected and quantified by the Saxon test[12].

Mann-Whitney rank sum U test was used to test for the difference of the means in 2 groups and Kruskal-Wallis statistic was used to test for the difference in 3 groups.

RESULTS

Table 1 shows the demographic and serological data for the study population. The sample was predominantly female (97%). The mean age of the subjects was 56.7 years (SD 13.0). Percentages of ocular symptoms (feeling of dry eye or sand in the eye, or frequent use of tear substitutes) were about 40%. The mean amount of tear examined by Schirmer-I test was 5.4 mm/5 min (SD 7.9) and the mean rose bengal score was 1.0 (SD 1.4). Fifty-six percent of 64 patients had positive fluorescein dye test. Percentages of oral symptoms (feeling of dry mouth or frequently drinking liquids) were 67%, while 25% of patients had recurrent salivary gland swelling. The mean focus score of minor salivary glands was 1.6 (SD 2.0)

Table 1. Demographic and serological characteristics. Values are positive number (%) unless otherwise indicated.

Characteristic	
Male/female	2/66
Age, yrs, mean ± SD	56.7 ± 13.0
Feeling of dry eye for more than 3 mo (N = 64)	23 (36)
Feeling of sand in the eye (N = 64)	31 (48)
Using tear substitutes more than 3 times a day (N = 64)	27 (42)
Schirmer-I test, mm/5 min, mean ± SD (N = 63)	5.4 ± 7.9
Rose bengal score, mean ± SD (N = 60)	1.0 ± 1.4
Fluorescein dye test (N = 64)	36 (56)
Feeling of dry mouth for more than 3 months (N = 64)	43 (67)
Frequent drinking of liquids when eating dry food (N = 64)	43 (67)
Recurrent salivary gland swelling (N = 64)	16 (25)
Focus score of minor salivary glands, mean ± SD (N = 65)	1.6 ± 2.0
Saxon test, g/2 min, mean ± SD (N = 66)	1.6 ± 1.3
Anticentromere antibodies	6 (9)
Anti-RNP antibodies	7 (10)
Anti-SSA/Ro antibodies	46 (68)
Anti-SSB/La antibodies	14 (21)

N: number of available data for analysis.

and the mean amount of saliva determined by the Saxon test was 1.6 g/2 min (SD 1.3). Forty-six patients (68%) had anti-SSA/Ro antibodies, while positive rates of anticentromere (9%), anti-RNP (10%), and anti-SSB/La (21%) were relatively low. Table 2 shows the levels of 4 quantitative measures in the patients with and without SS. The median levels of peak count, uptake speed, excretion speed, and excretion fraction in patients with SS were 28.5 count/s, 0.10 count/s^2, 0.25 count/s^2, and 85.0%, respectively, and the respective median levels in the patients without SS were 51.9 count/s, 0.13 count/s^2, 0.66 count/s^2, and 90.5%. The levels of peak count, uptake speed, and excretion speed in the patients with SS were significantly lower than those in the patients without SS (p = 0.0002, 0.0186, 0.0005, respectively), whereas there was no significant difference of excretion fraction level between the patients with SS and the controls (p = 0.0686). The p values of peak count and excretion speed were smaller than those of uptake speed and excretion fraction.

To determine the reproducibility of the measures, a doctor and a technician enclosed each ROI on the same gland independently and the 4 measures were computed automatically. The correlation coefficients of peak count (0.97) and excretion speed (0.98) were higher than those of uptake speed (0.90) and excretion fraction (0.89; Table 2).

Table 3 shows sensitivity, specificity, and accuracy for diagnosing SS at several cutoff levels of each measure. Cutoff levels of peak count, uptake speed, excretion speed, and excretion fraction that showed excellent balance between sensitivity and specificity were 43.0 count/s, 0.115 count/s^2, 0.450 count/s^2, and 88.9%, respectively. Peak count in the uptake phase and excretion speed in the excretion phase had higher sensitivity, specificity, and accuracy than those of uptake speed and excretion fraction. When peak count less

Table 2. Median level of the 4 quantitative measures in patients with and without Sjögren's syndrome (SS) and their reproducibility.

| | Median Level (25th, 75th percentile) | | | Reproducibility* |
	With SS, n = 45	Without SS, n = 23	p	
Peak count, count/s	28.5 (16.9, 53.6)	51.9 (40.9, 78.0)	0.0002	0.97
Uptake speed, count/s^2	0.10 (0.08, 0.13)	0.13 (0.10, 0.27)	0.0186	0.90
Excretion speed, count/s^2	0.25 (0.10, 0.65)	0.66 (0.42, 0.89)	0.0005	0.98
Excretion fraction, %	85.0 (71.9, 95.5)	90.5 (85.5, 94.7)	0.0686	0.89

* Two investigators determined the 4 parameters on the same gland independently, as described in Materials and Methods, and then each correlation coefficient of the levels of measures was calculated.

Table 3. Sensitivity, specificity, and accuracy for diagnosing SS at several cutoff levels of each quantitative measure.

Parameter	Cutoff	Sensitivity, %	Specificity, %	Accuracy, %
Uptake phase				
Peak count, count/s	31.0	64.4	91.3	73.5
	43.0	73.3	73.9	73.5
	58.7	82.2	47.8	70.6
Uptake speed, count/s^2	0.092	44.4	82.6	57.4
	0.115	66.7	65.2	66.2
	0.125	75.6	52.2	67.6
Excretion phase				
Excretion speed, count/s^2	0.382	64.4	82.6	70.6
	0.450	68.9	73.9	70.6
	0.763	86.7	43.5	72.1
Excretion fraction, %	79.0	37.8	95.7	57.4
	84.5	48.9	82.6	60.3
	88.9	66.7	65.2	66.2

than 43.0 count/s was taken to be indicative of diagnosis of SS, sensitivity, specificity, and accuracy were 73.3%, 73.9%, and 73.5%, respectively. When excretion speed less than 0.450 count/s^2 was considered to be the cutoff value, we obtained sensitivity of 68.9%, specificity of 73.9%, and accuracy of 70.6%.

Figure 2A shows the relationship between focus scores of minor salivary glands and the uptake phase measures. The numbers of patients with no focus, one focus, and 2 or more foci of the minor salivary glands were 23, 17, and 25, respectively. The levels of peak count declined significantly as the focus scores increased (p = 0.0039), while there was no significant relation between the grade of minor salivary gland and the level of uptake speed.

Figure 2B shows the relationship between the amount of stimulated whole saliva collection (the Saxon test) and the excretion phase measures. The numbers of patients with the Saxon test of 2 or more, 1 to 2, and less than 1 (g/2 min) were 20, 20, and 26, respectively. The levels of excretion speed decreased significantly among the 3 groups (p = 0.0023). In contrast, there was no significant difference between the levels of excretion fraction and the levels of the Saxon test.
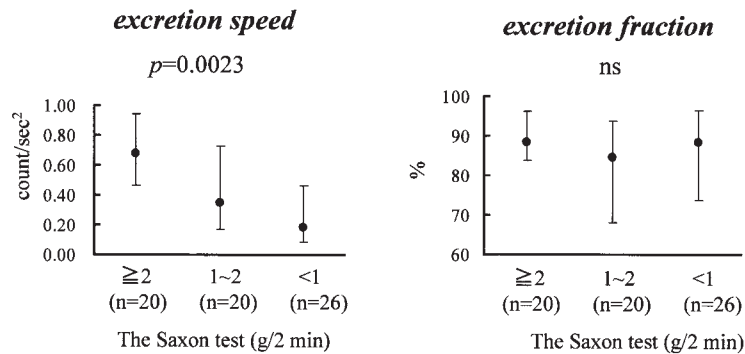
DISCUSSION
Salivary gland scintigraphy is widely used for evaluation of salivary gland function and also for diagnosing SS[1]. Schall's qualitative 4-grade system has generally been accepted for diagnosing SS[1-4]. However, there is fundamentally no arithmetical relationship among the different grades classified by qualitative methods. In addition, borderline data may be misclassified, since the classification depends on individual examiners. On the other hand, computer-assisted quantitative measures are continuous and objective, and reflect salivary gland function more precisely than qualitative measures. Among previous quantitative methods[6-10], Håkansson, *et al*[8] demonstrated that the levels of uptake speed, excretion speed, and excretion fraction in patients with SS were significantly lower than those in controls, but that the variance of peak count was too large to detect the difference between patients with SS and controls. The large variance of peak count in their study may be due to the small sample size of patients with SS (n = 17) rather than the aging and/or duration of sicca symptoms as they stated, since other studies[6,9] and ours, based on larger sample size, showed the usefulness of peak count. Moreover, there was no correlation between the level of peak

A   Uptake phase parameters

peak count

uptake speed

Focus scores of minor salivary gland

Focus scores of minor salivary gland
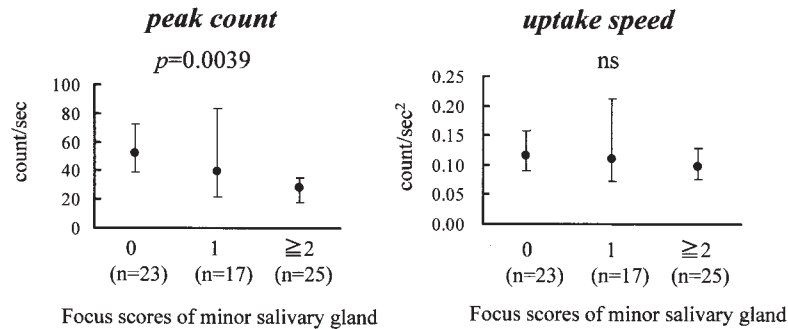
B   Excretion phase parameters

excretion speed

excretion fraction

The Saxon test (g/2 min)

The Saxon test (g/2 min)

Figure 2. The median levels of the uptake phase measures stratified by focus scores of the minor salivary glands (A) and the median levels of the excretion phase measures stratified by the amount of stimulatory whole saliva collection determined by the Saxon test (B). Error bars indicate 75th and 25th percentiles. There was significant difference among the 3 groups in the level of peak count but not in uptake speed, and the level of peak count declined as the focus scores increased (A). There was significant difference among the 3 groups in the level of excretion speed but not in excretion fraction, and the level of excretion speed diminished as the saliva excretion amount decreased (B).

count and the aging and/or duration of sicca symptoms in our study (data not shown).

The excretion fraction has been useful to discriminate SS from controls in previous reports[6-9]. However, we could find no significant difference of excretion fraction level between the patients with SS and the controls. Excretion fraction was defined as excretion count divided by peak count, as described in Materials and Methods. Therefore, when the level of peak count was near zero, even a small variation of excretion count caused large variation of excretion fraction. In order to avoid this large variation, Wang, *et al*[6] excluded data of low peak count and Aung, *et al*[9] substituted zero for excretion fraction when peak count was near zero. In contrast, we calculated excretion fraction without intentional modulation, which may cause different results on the usefulness of excretion fraction.

Because of the lack of reports that compared peak count with uptake speed in the uptake phase and excretion speed with excretion fraction in the excretion phase, we determined which measure was superior to the other. The lower p values

of peak count and excretion speed indicated that differences of peak count and excretion speed levels between the patients with SS and the controls were more significant than those of uptake speed and excretion fraction. Peak count and excretion speed were more reproducible than uptake speed and excretion fraction. In addition, sensitivity, specificity, and accuracy of peak count and excretion speed for diagnosing SS were higher than those of uptake speed and excretion fraction. All these findings suggested that peak count and excretion speed were superior to uptake speed and excretion fraction.

An animal experiment and clinical studies showed that the destruction of salivary acinar cells and the loss of acinar component caused the reduction of uptake levels of ^{99m}Tc-pertechnetate, which led peak count and excretion speed levels to decrease[4,5,8,9,13-15]. In our study, most patients with low peak count had low excretion speed; however, there was a discrepancy between peak count and excretion speed in some patients (data not shown). This discrepancy was probably due to unknown factors. We compared peak count and uptake speed

among 3 groups divided by focus scores of minor salivary glands. The level of peak count decreased significantly as the salivary gland damage progressed (p = 0.0039), whereas the level of uptake speed did not. Meanwhile, the level of excretion speed declined as the amount of whole saliva collection decreased and there was significant difference in the level of excretion speed (p = 0.0023), but not in the level of excretion fraction among the 3 groups divided by the Saxon test. These findings again indicated that peak count in the uptake phase and excretion speed in the excretion phase were eligible for standard quantitative measures.

What are the benefits of using peak count and excretion speed? First, since peak count and excretion speed are continuously measurable and highly reproducible, these quantitative measures would detect an individual tiny change in a followup study. Secondly, these quantitative data could be compared with others from different institutes more reliably than data obtained by uptake speed or excretion fraction.

To accept peak count in the uptake phase and excretion speed in the excretion phase as the standard measures for diagnosing SS, we need to unify the amount of ^{99m}Tc or timing of lemon stimulation. And finally, multicenter studies should be performed to determine the universal cutoff levels of peak count and excretion speed, and the sensitivity and specificity of parotid gland scintigraphy for diagnosing SS.

ACKNOWLEDGMENT

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