Dynamic Properties of Human Stapedial Annular Ligament Measured With Frequency–Temperature Superposition

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*Stapedial annular ligament (SAL) is located at the end of human ear ossicular chain and provides a sealed but mobile boundary between the stapes footplate and cochlear fluid. Mechanical properties of the SAL directly affect the acoustic-mechanical transmission of the middle ear and the changes of SAL mechanical properties in diseases (e.g., otosclerosis) may cause severe conductive hearing loss. However, the mechanical properties of SAL have only been reported once in the literature, which were obtained under quasi-static condition (Gan, R. Z., Yang, F., Zhang, X., and Nakmali, D., 2011, “Mechanical Properties of Stapedial Annular Ligament,” *Med. Eng. Phys.*, **33**, pp. 330–339). Recently, the dynamic properties of human SAL were measured in our lab using dynamic-mechanical analyzer (DMA). The test was conducted at the frequency range from 1 to 40 Hz at three different temperatures: 5 °C, 25 °C, and 37 °C. The frequency–temperature superposition (FTS) principle was applied to extend the testing frequency range to a much higher level. The generalized Maxwell model was employed to describe the constitutive relation of the SAL. The storage shear modulus G' and the loss shear modulus G'' were obtained from seven specimens. The mean storage shear modulus was 31.7 kPa at 1 Hz and 61.9 kPa at 3760 Hz. The mean loss shear modulus was 1.1 kPa at 1 Hz and 6.5 kPa at 3760 Hz. The dynamic properties of human SAL obtained in this study provide a better description of the damping behavior of soft tissues than the classic Rayleigh type damping, which was widely used in the published ear models. The data reported in this study contribute to ear biomechanics and will improve the accuracy of finite element (FE) model of the human ear. [DOI: 10.1115/1.4027668]*

Keywords: stapedial annular ligament, dynamic mechanical analyzer, dynamic properties, complex modulus, frequency–temperature superposition

1 Introduction

SAL is located at the end of human ear ossicular chain and provides a sealed but mobile boundary between the stapes footplate and cochlear fluid [1–4]. During the stapes footplate piston-like vibration, in which the footplate moves inward and outward the cochlear fluid [5,6], the SAL mainly sustains high-frequency shear deformation.

Mechanical properties of the SAL directly affect the acoustic-mechanical transmission function of the middle ear [7] and the abnormal changes of SAL induced by ear diseases may cause severe conductive hearing loss. For example, in otosclerosis, the abnormal ossification of the SAL reduces the movement of the stapes and causes the conductive hearing loss [8,9]. Moreover, since the transmission of sound energy from the middle ear to cochlear fluid is largely dependent on SAL mechanical behavior, the properties of SAL have affected the outcomes of stapedotomy or stapesplasty surgical approaches with different prostheses or SAL grafts [10–12]. Therefore, understanding the mechanical properties of the SAL is one of important research subjects in middle ear biomechanics.

Despite the importance, mechanical properties of the SAL have only been reported once in the literature by Gan et al. [13]. They measured the shear modulus of human SAL through the quasi-static pulling test and the results showed that the SAL had typical nonlinear mechanical behaviors and the shear modulus varied

from 3.6 to 220 kPa when the shear stress increased from 2 to 140 kPa. However, the ear tissues work under the auditory frequency range from 20 to 20,000 Hz. The lack of dynamic properties of SAL over the auditory frequency range affects fully understanding of sound transmission from the middle ear to cochlea and the development of an accurate FE model of the ear. In most FE models of the human ear, mechanical properties of the SAL were assumed through the cross-calibration process and a constant elastic modulus varying from 0.065 to 5.5 MPa was used for the FE models [14–16]. There is an urgent need to measure SAL dynamic properties in the auditory frequency range.

DMA is widely used to measure dynamic properties of materials in the frequency domain. However, the current commercial DMA has limited higher frequency access and cannot reach the human auditory frequency range. Recently, a study on measuring the dynamic properties of human tympanic membrane based on the FTS principle has been reported by our group [17]. The basic theory of FTS considers the dynamic behavior of some materials as a function of two principle variables: frequency and temperature. The effect of temperature changing on dynamic properties of the material is equivalent to that of frequency changing [18,19]. The FTS as an empirical method has been validated by our previous study on tympanic membrane and can be applied to measure the dynamic properties of ear tissues in auditory frequencies.

In this paper, following the method developed in our previous paper [17], the dynamic properties of human SAL were measured using the DMA. The test was conducted at frequencies from 1 to 40 Hz at three different temperatures: 5 °C, 25 °C, and 37 °C. The FTS principle was applied to extend the testing frequency to a much higher level (at least 3760 Hz). The generalized Maxwell

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Fig. 1 (a) The schematic of the experiment setup for dynamic test of the SAL specimen in DMA. (b) The enlarged image for the fixation of stapes head to the mounting fixture. The SAL was hiding behind the bony structures.

model was employed to describe the constitutive relation of the SAL. The storage shear modulus G' and the loss shear modulus G'' were obtained from seven SAL specimens. The data reported in this study contribute to the biomechanics of the middle ear.

2 Methods

2.1 SAL Specimen Preparation and Experimental Setup.

Seven fresh human temporal bones (TBs) obtained through the willied body program at the University of Oklahoma Health Sciences Center were used for this study. The average age of donors was 68.7 (ranging from 64 to 75 years, three male and four female). The preparation of the SAL sample followed the method reported by Gan et al. [13] on measuring mechanical properties of human SAL under quasi-static conditions. Briefly, the TB was immersed in 0.9% saline solution mixed with 15% povidone at 5°C before the experiment to maintain SAL compliance and hydration. The TB was checked under the microscope for not having any abnormality before experiments. After opening the tegmen and removing the tympanic membrane together with the malleus and incus, the TB was cut into a nearly 2 cm × 2 cm × 1.5 cm cube to expose the stapes with attached SAL and surrounding bony wall. The scala vestibule wall near the oval window was open to release cochlear pressure and saline solution was added beneath the stapes footplate to maintain the physiological condition for the tissue.

Figure 1(a) is the schematic of experimental setup for dynamic test of SAL specimen in DMA. The bony part of the whole SAL

specimen was fixed on an X-Y translational stage and placed inside a temperature chamber, in which a thermocouple was placed at 2 cm away from the sample. There was a negative feedback circuit to control the selected temperature stably. The precision of temperature controlling was $\pm 1^\circ\text{C}$. The SAL sample was aligned in DMA (ElectroForce 3200, Bose, Eden Prairie, MN) by adjusting the X-Y translational stage and Z-translational column under microscope. A titanium partial ossicular-replacement prosthesis (PORP, Gyrus ENT, LLC, Memphis, TN) was used as a mounting fixture for stapes as shown in Fig. 1(b). The stapes head was mounted to the metal cap of the PORP with a tiny drop of cyanoacrylate gel glue. Care was taken not to allow the glue to reach the SAL.

2.2 Measurement of SAL Specimen Dimensions.

Dimensions of the SAL specimen were measured using the same method as reported by Gan et al. [13]. Since the SAL specimen was hiding between the stapes footplate and the cochlea bony wall and not visible during the test, the dimensions of SAL were measured after dynamic test, when the stapes footplate was pulled out from the oval window. The still images of the stapes and oval window were captured using a digital CCD camera. Figure 2(a) shows the measurements of stapes footplate length $L1$ and width $L2$, and Fig. 2(b) shows the oval window length $L3$ and width $L4$. Some histological studies on the human middle ear suggested that the SAL had larger thickness at the anterior side than posterior side [20,21]. However, the accurate measurement on the in vivo

Fig. 2 Images of the stapes footplate (a) and oval window (b) stapes, and (c) obtained after experiments on one temporal bone specimen for measuring the dimensions of SAL

Table 1 The dimensions of human SAL samples. Note: FP denotes stapes footplate and OW denotes oval window.

Specimens	SAL-1	SAL-2	SAL-3	SAL-4	SAL-5	SAL-6	SAL-7	Mean $\pm$ SD
FP length, L_1 (mm)	2.9	2.82	2.78	2.58	2.32	2.68	2.12	2.60 ± 0.28
FP width, L_2 (mm)	1.2	1.22	1.18	1.14	1.08	1.12	1.04	1.14 ± 0.07
OW length, L_3 (mm)	3.12	3	2.96	2.76	2.54	2.84	2.3	2.79 ± 0.29
OW width, L_4 (mm)	1.32	1.36	1.3	1.28	1.24	1.24	1.18	1.27 ± 0.06
SAL thickness, d (mm)	0.09	0.08	0.08	0.08	0.10	0.07	0.08	0.08 ± 0.01
SAL (FP) height, h (mm)	0.29	0.26	0.27	0.24	0.23	0.25	0.2	0.25 ± 0.03
SAL (FP) perimeter, C (mm)	8.56	7.86	7.74	8.32	7.26	8.04	7.06	7.83 ± 0.54

dimension of the SAL along the whole perimeter of the footplate is still not approachable due to the technique limitation. In this study, to simplify the problem and focus on the mechanical behavior of the SAL as a whole component, the thickness was assumed to be uniformly distributed along the perimeter of the footplate. Therefore, the length or width difference between the stapes footplate and oval window, L_3-L_1 and L_4-L_2 , can be calculated and the average value was considered as the SAL thickness, d . The possible errors caused by the inaccurate thickness measurement have been estimated in our previous study on the static properties of SAL [13]. When the thickness changes up to 0.02 mm, the relative error in this study is about 25% (based on the average thickness of 0.08 mm).

The SAL height, which was considered to be the same as the height of stapes footplate edge h , was measured from the captured image of the specimen as shown in Fig. 2(c). The height was measured at three different locations (the posterior, anterior, and middle regions) and the average value was used in this study. The perimeter of the SAL inner rim or footplate perimeter C was calculated from the transferred binary image of the stapes footplate. Table 1 lists the dimensions of SAL specimens ($N=7$) with the mean and standard deviation (SD).

2.3 Viscoelastic Model of SAL Specimen. The SAL specimens underwent the shear stress and shear strain during the tests, as analyzed in Fig. 4 of Gan et al. [13]. The shear stress τ and shear strain γ can be calculated by

$$\tau(t) = F/Ch = F_0 e^{i(2\pi ft + \delta)} / Ch \quad (1)$$

$$\gamma(t) = u/d = u_0 e^{i2\pi ft} / d \quad (2)$$

where t is time, f is the frequency, δ is the phase difference between the force and displacement, F and u are the force and displacement, respectively, while F_0 and u_0 are their amplitude. The complex modulus G^* with the storage shear modulus G' and loss shear modulus G'' of the SAL are then calculated by

$$|G^*| = \frac{F_0/Ch}{u_0/d} \quad (3)$$

$$G' = |G^*| \cos \delta \quad (4)$$

$$G'' = |G^*| \sin \delta \quad (5)$$

In this study, SAL was considered as an isotropic and homogeneous material and the generalized Maxwell model was used. The complex shear modulus can be expressed as follows:

$$G'(\omega) = G_0 + \sum_{i=1}^n G_i \tau_i^2 \omega^2 / (1 + \tau_i^2 \omega^2) \quad (6)$$

$$G''(\omega) = \sum_{i=1}^n G_i \tau_i \omega / (1 + \tau_i^2 \omega^2) \quad (7)$$

$$\eta(\omega) = \tan \delta = G''(\omega) / G'(\omega) \quad (8)$$

where ω is the circular frequency and equal to $2\pi f$. G_0 , G_i , and τ_i are the material parameters to be determined through experiments.

Fig. 3 The complex shear modulus–frequency curves obtained at 5 °C, 25 °C, and 37 °C from two SAL specimens: (a) sample SAL-1 and (b) sample SAL-6.

Our previous study on TM dynamic properties shows that two-term linear solid model is proper to represent the viscoelastic behavior of middle ear tissues [17]. Two spring-dashpot elements ($n=2$) gave a total of five parameters: G_0 , G_1 , G_2 , τ_1 , and τ_2 , which were determined through the theoretical fitting with the experimental data for each SAL specimen.

As reported in our previous study on the quasi-static properties of human SAL [13], the SAL samples exhibited the nonlinear stress–strain relationship as a typical soft tissue. However, when the displacement was within 0.1 mm, it was in an approximately linear range. As the first attempt to measure the dynamic properties of human SAL, we employed the linear viscoelastic model for SAL to simplify the approach in this study.

2.4 Dynamic Test on SAL Specimen

2.4.1 Preconditioning Test. To stabilize the mechanical properties of SAL specimens, the preconditioning tests were conducted in DMA first. Five cycles of uniaxial vibration were performed at a frequency of 0.1 Hz with the displacement amplitude of 0.1 mm for each specimen before the dynamic test. At each frequency of the dynamic test, at least ten cycles of sinusoidal vibration were conducted on the TM sample. The first five cycles could also serve as the preconditioning which had the exactly same loading conditions as the test. Thus, the dynamic test itself already included

preconditioning. It is also noticed that at high frequencies, the tissue might not have sufficient time for relaxation following preconditioning and the dynamic properties obtained from this study would be reasonably higher than the static or quasi-static properties measured with a longer relaxation time.

2.4.2 Dynamic Test. After preconditioning, the dynamic tests were conducted at 5 °C, 25 °C, and 37 °C in sequence. Three temperatures were selected based on our previous study on human tympanic membrane [17], which proved that these temperatures were proper for dynamic tests of ear tissues. Chan [22] discussed that there was no phase change (freezing) of fluid in tissue cells, and there was no denature of proteins between 5 °C and 37 °C. Thus, the structure of soft tissue in this temperature range should not change. At each temperature, the SAL sample was tested at 1, 2, 5, 10, 20, and 40 Hz with the displacement amplitude of 0.1 mm. Note that the SAL sample was kept in physiological moisture by adding saline solution through the opened hole on cochlea wall. To keep sample in the same moisture condition, the accurate amount of saline solution was controlled by a syringe.

2.5 FTS Principle. The FTS principle was first reported in the 1950s [23,24], and has been applied to polymers, plastics, and composites [25]. The later theories of FTS address a simple relationship between the temperature and frequency (or time) effects on the molecular behaviors of polymers and thus the dynamic properties of these materials [18]. Recently, researchers have applied the FTS principle to some biological tissues [17,22,26]. In practice, the curves of the complex shear modulus G^* obtained at a relatively low temperature T can be shifted along the frequency axis by a shift factor α_T to a higher temperature T_0 (served as reference temperature). This concept can be expressed by the equation

$$G^*(T_0, f) = G^*(T, f/\alpha_T) \quad (9)$$

where f is the frequency. The shift factor α_T quantifies the temperature's effect on the material's complex modulus. The shift factor α_T itself is temperature-dependent and need to obey WLF equation, which was first introduced by Williams, Landel and Ferry [24] in 1955 [22,24] and widely used for the FTS principle

$$\log \alpha_T = \frac{c_1(T - T_0)}{c_2 + T - T_0} \quad (10)$$

where T and T_0 are the absolute temperature in Kelvin, c_1 (dimensionless unit) and c_2 (unit in Kelvin) are empirical constants.

The detailed procedure about how to determine dynamic properties of the SAL samples at higher frequencies using FTS principle can be referred to our previous study [17]. Briefly, the complex shear moduli of SAL samples obtained at different temperatures (5 °C, 25 °C, and 37 °C) were plotted together as functions of frequency in logarithmic scale. Then, the complex modulus curves at the lower temperatures were shifted horizontally toward higher frequencies to connect with the curves obtained at higher temperatures and to create the final master curve at the reference temperature (37 °C). The shift factor α_T was then determined by Eq. (9). For each SAL sample, the constants, c_1 and c_2 , were further determined by Eq. (10) and the consistency between experimental data and WLF equation was verified.

3 Results

Dynamic experiments were conducted on seven SAL specimens. The storage shear modulus G' and the loss shear modulus G'' of these specimens were obtained over 1–40 Hz. Figure 3 shows the typical complex shear modulus–frequency curves at three different temperatures (5 °C, 25 °C, and 37 °C) obtained from two SAL specimens (SAL-1 and SAL-6). The storage shear modulus increased with the frequency increasing or temperature decreasing. However, the loss shear modulus decreased above

Fig. 4 The master curves of the complex shear modulus at 37 °C obtained from two SAL sample: (a) sample SAL-1 and (b) sample SAL-6

20 Hz at 5 °C. Larger slopes were found at the lower frequency range and lower temperatures for the loss modulus. Other SAL specimens had similar complex shear modulus–frequency curves to that shown in Fig. 3, with some individual variations.

Following the procedure described in Sec. 2.5, the complex shear modulus curves at the lower temperatures were shifted to higher frequency and adjacent with those curves obtained at higher temperatures. Thus, the master curves at reference temperature were created. Figure 4 shows the master curves of the complex shear modulus at 37 °C for specimens SAL-1 and SAL-6. The same horizontal shift factors were used for the storage modulus and loss modulus in each specimen. The complex shear modulus–frequency curves are generally well matched at the adjacent regions after the horizontal shifts. For specimen SAL-1, the storage shear modulus is 25.8 kPa at 1 Hz and increases to 62.1 kPa at 4160 Hz, while the loss shear modulus is 0.8 kPa at 1 Hz and 7.1 kPa at 4160 Hz. For specimen SAL-6, the storage shear modulus is 27.0 kPa at 1 Hz and increases to 65.5 kPa at 3760 Hz, while the loss shear modulus is 1.2 kPa at 1 Hz and 7.0 kPa at 3760 Hz. The shift factors and maximum frequency of the master curves for all seven SAL specimens are listed in Table 2. The mean value with SD of the shift factors from 25 °C to 37 °C (α_{25}) is 7.9 ± 1.4 . The mean value with SD of the shift factors from 5 °C to 37 °C (α_5) is 123.3 ± 24.4 . The maximum frequency ranged from 3760 to 6560 Hz with a mean value of 4931 ± 975 Hz.

The temperature dependence of the shift factor α_T was tested by fitting experimental data into the WLF equation (Eq. (10)) and two constants c_1 and c_2 were determined. The values of c_1 and c_2 for all SAL specimens were listed in Table 2. The value of c_1 ranges from 16.8 (sample SAL-6) to 34.0 (sample SAL-7) with the average and SD of 25.3 ± 5.5 , while the value of c_2 ranges from -73.5 (sample SAL-6) to -200.7 (sample SAL-7) with the average and SD of -138.1 ± 41.5 .

Table 2 The shift factors, c_1 and c_2 of WLF equation, and maximum frequency of human SAL samples

Specimen	SAL-1	SAL-2	SAL-3	SAL-4	SAL-5	SAL-6	SAL-7	Mean $\pm$ SD
α_{25}	7.0	6.7	8.5	8.1	7.9	10.6	6.8	7.9 ± 1.4
α_5	104	94.0	142	119	132	164	108	123.3 ± 24.4
$\ln \alpha_{25}$	1.94	1.90	2.14	2.09	2.07	2.36	1.92	2.06 ± 0.16
$\ln \alpha_5$	4.64	4.54	4.96	4.78	4.88	5.10	4.68	4.80 ± 0.20
c_1	28.1	27.3	23.7	21.0	26.3	16.8	34.0	25.3 ± 5.5
c_2 (K)	-162.0	-160.4	-120.9	-108.5	-140.5	-73.5	-200.7	-138.1 ± 41.5
Maximum frequency (Hz)	4160	3760	5680	4760	5280	6560	4320	4931 ± 975

Fig. 5 The master curves of the storage modulus and loss modulus at 37 °C from seven SAL samples and the mean master curves of the storage modulus and loss modulus

Figure 5 shows the master curves of the complex shear modulus for all seven SAL specimens. The complex shear modulus generally increases with the frequency increasing except the loss shear modulus decreases a little between 60 to 200 Hz and above 3000 Hz. The storage shear modulus ranges from 23.8 to 41 kPa at 1 Hz. The loss shear modulus ranges from 0.18 to 0.45 MPa at 1 Hz. The mean complex shear modulus was calculated over the common frequency range for the seven SAL samples (1–3760 Hz) and the mean master curves of the storage shear modulus G' and the loss shear modulus G'' with SD were plotted up to 3760 Hz in Fig. 5. The mean storage shear modulus was 31.7 kPa at 1 Hz and 61.9 kPa at 3760 Hz. The mean loss shear modulus was 1.1 kPa at 1 Hz and 6.5 kPa at 3760 Hz.

The generalized Maxwell model was used to describe the viscoelastic behavior of the SAL specimens. The experimental data of the storage shear modulus were used to fit the theoretical storage modulus–frequency relation (Eq. (6)) to determine the five parameters (G_0 , G_1 , G_2 , τ_1 , and τ_2). The theoretical loss shear modulus was then calculated by substituting the values of the parameters into Eq. (7), and the results were compared with the experimental data. As an example, the five parameters for SAL-1 were determined as $G_0 = 27.6$ kPa, $G_1 = 15.2$ kPa, $G_2 = 19.0$ kPa, $\tau_1 = 13.4$ ms, and $\tau_2 = 174$ μ s, while for SAL-6, the five parameters were determined as $G_0 = 28.1$ kPa, $G_1 = 21.8$ kPa, $G_2 = 18.4$ kPa, $\tau_1 = 6.0$ ms, and $\tau_2 = 69.7$ μ s. Table 3 lists the

viscoelastic parameters determined from all seven SAL specimens with the mean values and SD. The relatively large difference between the relaxation time constants in Table 3 may attribute to the variations of sample conditions. The square of correlation coefficient, r^2 , was calculated to estimate the goodness of fitting process for experimental data. The values of r^2 for seven samples are also listed in Table 3 (bottom row). The lowest value of r^2 is 0.9741 for sample SAL-3, which demonstrated a satisfied match between the theoretical model and experimental data. Figure 6(a) shows the complex shear modulus of specimen SAL-1 derived from the generalized Maxwell model in comparison with the experimental results and Fig. 6(b) shows that complex shear modulus curves for specimen SAL-6. The mean experimental complex modulus curves shown in Fig. 5 were also fitted by the generalized Maxwell model with the five parameters determined as $G_0 = 31.8$ kPa, $G_1 = 15.4$ kPa, $G_2 = 16.0$ kPa, $\tau_1 = 8.9$ ms, and $\tau_2 = 121$ μ s. The results are shown in Fig. 6(c) where the theoretical complex modulus curves were compared with the mean experimental data.

4 Discussion

4.1 Feasibility of FTS. In this study, the FTS principle was applied to expand the effective frequency to a much higher level (at least 3760 Hz) compared with the real testing frequency (up to 40 Hz). The feasibility of FTS needs to be checked and validated. There are three requirements for the FTS principle to hold as commented by Ward [27] (1971): (1) perfect matching of the curve shapes at adjacent regions, (2) same shift factor value for all viscoelastic parameters, and (3) the shift factor obeying the WLF equation. The results obtained in this study show that the complex shear modulus–frequency curves of the human SAL specimens satisfied all three requirements for the FTS principle. Thus, the FTS principle is feasible to measure dynamic properties of the SAL at the auditory frequency range. This method also has the potential to test other middle ear soft tissues, such as the ligament and round window membrane, which are mainly composed by collagen fibers and can be considered as homogenous materials as well.

4.2 Comparison With Published Data. To date, there is no report about dynamic properties of SAL in the literature. The results reported in this study fill the gap in biomechanics of the middle ear tissues. However, the mechanical properties of human SAL under quasi-static conditions have been studied by Gan et al. [13]

Table 3 The viscoelastic parameters of human SAL samples

Specimen	SAL-1	SAL-2	SAL-3	SAL-4	SAL-5	SAL-6	SAL-7	Mean $\pm$ SD
G_0 (kPa)	27.6	26.0	26.7	36.4	33.8	28.1	42.6	31.6 ± 6.2
G_1 (kPa)	15.2	16.7	9.4	13.1	17.7	21.8	16.4	15.8 ± 3.9
G_2 (kPa)	19.0	17.0	10.5	19.0	17.2	18.4	17.3	16.9 ± 3.0
τ_1 (ms)	13.4	10.1	8.7	12.3	5.6	6.0	8.2	9.2 ± 3.0
τ_2 (μ s)	174	181	100	134	92.8	69.7	64.5	117 ± 47
r^2	0.9829	0.9818	0.9741	0.9745	0.9888	0.9876	0.9773	0.9810 ± 0.0059

Fig. 6 The theoretical fitting of generalized Maxwell model to the experimental complex modulus for (a) sample SAL-1, (b) sample SAL-6, and (c) the mean experimental complex modulus

using pulling tests in MTS. Their study demonstrated that human SAL is a typical viscoelastic material with hysteresis and stress relaxation behavior. The SAL showed nonlinear stress-strain relationship and the shear modulus changed from 3.6 to 220 kPa when the shear stress increased from 2 to 140 kPa. In the present study, the complex shear modulus of the SAL specimens was obtained over the frequency range from 1 to 3760 Hz. The storage shear modulus at 1 Hz can be considered comparable with the shear modulus obtained from the quasi-static test. The mean storage shear modulus of seven SAL specimens at 1 Hz was 31.7 kPa obtained from this study, which was among the range from 3.6 to 220 kPa reported by Gan et al. [13].

Considering that the SAL is a nonlinear material and the shear modulus varies with the stress or strain level, we further compared the results from these two studies at the same strain level. In this study, the displacement amplitude of 0.1 mm was used in dynamic tests, and thus, the maximum shear strain was about 1.25 calculated by Eq. (2) with the thickness of 0.08 mm. Gan et al. reported the mean shear stress of 17.5 kPa, and the mean shear modulus of 35.2 kPa when the shear strain was 1.25. The value of 35.2 kPa

was close to the result (31.7 kPa) reported in this study. The difference between the results from two studies was probably caused by either the individual variation of human SAL specimens or the different experiment setup and testing condition.

The complex shear modulus G' and G'' of human SAL obtained in this study can be converted into the complex modulus E' and E'' in extension and applied into the FE model of the human ear. There is a simple relation between the shear modulus and elastic modulus as expressed by

$$E' = 2(1 + \nu)G' \quad E'' = 2(1 + \nu)G'' \quad (11)$$

where ν is the Poisson's ratio of SAL. The value of 0.3 was widely used for ν in the published FE models of the ear [15,28,29] and was adopted for converting shear modulus G' and G'' to tensile modulus E' and E'' . The calculation indicated that the mean storage modulus E' was 82.4 kPa at 1 Hz and 160.9 kPa at 3760 Hz. The mean loss modulus E'' was 2.86 kPa at 1 Hz and 16.9 kPa at 3760 Hz. In most published FE models of human ear summarized by Zhao et al. [29], the constant elastic modulus was used for SAL and the large variation existed for the value from 0.065 to 5.5 MPa.

4.3 Comparison of the Viscoelastic Model and Rayleigh Type Damping of SAL. The complex modulus of human SAL obtained in this study can be applied into the FE model of the human ear and help improve the accuracy of the model. In most published FE models of the human middle ear, a Rayleigh type damping was applied for the soft tissues, including the SAL [29]. Rayleigh type damping parameter c is defined in terms of material mass and stiffness for a structure (e.g., ear) as follows:

$$c = \alpha M + \beta K \quad (12)$$

where α and β are the mass and stiffness coefficients, respectively. M represents the mass and K represents the stiffness components

Fig. 7 (a) Damping of the SAL calculated using Rayleigh type damping coefficient with $\beta = 0.000075$ and (b) damping of the SAL calculated from the viscoelastic model determined from the dynamic test in this study

for the ear. As an example, values of 0 and 0.000075 were used for α and β coefficients, respectively, in Gan et al.'s FE models [15,28]. A constant elastic modulus and the Rayleigh type damping in FE models make the loss modulus or damping of the SAL proportional to the frequency, which makes the damping too high at the higher frequencies as shown in Fig. 7(a). Recently, Zhang and Gan [30] reported the FE model of human ear with viscoelastic properties for the middle ear soft tissues. The damping or loss modulus was not proportional to the frequency but calculated by equations, such as Eqs. (5)–(7) in this study. Based on material parameters obtained in this study, the damping reaches its maximum value around 1 kHz and then decreases (Fig. 7(b)). The application of the viscoelastic properties for the middle ear soft tissues can avoid too large damping at high frequency and improve the results at the higher frequencies as shown in Zhang and Gan's [30] model when compared with the previous model of Gan et al. [15].

5 Conclusion

This is the first study to report the dynamic properties of human SAL over the auditory frequency range (up to 3760 Hz). The dynamic tests were conducted at frequencies from 1 Hz to 40 Hz at three different temperatures: 5°C, 25°C, and 37°C and the FTS principle was applied to expand the frequency range to a much higher level. The generalized Maxwell model was used to describe the viscoelastic behavior of the SAL. The storage shear modulus G' and the loss shear modulus G'' in the frequency domain were obtained from seven SAL specimens. The mean storage shear modulus was 31.7 kPa at 1 Hz and 61.9 kPa at 3760 Hz. The mean loss shear modulus was 1.1 kPa at 1 Hz and 6.5 kPa at 3760 Hz. The data reported here contribute to the biomechanics of the middle ear tissues and help improve the accuracy of the FE model for the human ear.

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