An acoustic pressure pipette aspiration method for isotropic materials

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Abstract: A measurement setup is presented as a method to determine the elasticity modulus of both artificial and real tissue as a function of frequency. The future goal at the end is to develop and validate the method to measure the elasticity modulus of ex vivo cadaver animal vocal folds over the human phonation frequency range. In the present study, we investigate an innovative acoustic pressure pipette aspiration technique to measure the material characteristic of an isotropic silicone specimen with similar characteristics as human vocal folds. With the aim of ensuring reliable results, we test our method on different mixtures of silicone rubber. Four silicone samples with different consistencies were tested over a frequency range of 50–500Hz. Influence of vibrometer laser spot location in an enclosed area on the sample surface was investigated by repeating the measurements ten times. The results show only small standard deviations demonstrating the potential of the acoustic pressure pipette aspiration technique for material characterization.

1. Introduction

The determination of mechanical material properties of synthetic as well as natural vocal fold material is an important task. The information is necessary for the verification of synthetic models, for numerical simulations and finally for the further understanding of the phonation process. In particular, silicone or polyurethane mixtures are used as substitute for vocal folds tissue on which the research of this study is aimed at. This work presents a method to measure the elasticity modulus of silicone specimen with different mixtures as a function of frequency. We focus is on the pipette aspiration method which was originally introduced to measure the stiffness and intracellular pressure of red blood cell membranes by Rand and Burton [1]. Therein, a pipette was placed onto the material sample and excited via a static low pressure, sucking it into the pipette. Aoki [2] applied it to the local elasticity measurement of soft tissue. In a recent investigation, Weiss [3-6] and Zörner [7] developed a new pipette aspiration setup to measure silicone sample elasticity. In the both experimental setups, the silicone sample was placed under the pipette and by means of a pistonphone, which was mounted on an electromechanical shaker and connected to a small gauge head by a flexible tube, a fluctuating pressure was generated within the pipette resulting in a vibration of the area enclosed by the pipette. The out-of-plane velocity at several measurement points on the specimen’s surface was measured by the laser scanning vibrometer. We enhanced the aspiration pipette method to a miniaturized pipette with acoustic pressure excitation which
allows to dynamically apply an acoustic pressure excitation to the surface of the material at frequencies in the human phonatory range.

2. Experimental setup of the enhanced aspiration pipette
For experimental investigations, four mixing ratio samples 1:1:0, 1:1:2, 1:1:3 and 1:1:4 were made of a two-part silicone composite, Ecoflex TM Platinum Cure Silicone Rubber, Part-A: Part-B: silicone thinner. The measurement of the four samples have been performed by the pipette aspiration with the experimental setup shown in (figure 1). In the original setup as reported in Aoki [2] a static low pressure was used to suck the material into the pipette and the resulting deformation was measured. By a simple analytical formula, the elasticity modulus is determined. We replace the static low pressure by using a loudspeaker to produce an acoustic pressure at a predefined frequency as displayed in figure 1.

![Figure 1. Picture of the measurement setup.](image)

**Figure 1.** Picture of the measurement setup.

The loudspeaker is mounted in 10 mm distance from the base of the pipette wall with a pipette fixed to it that is placed upon the sample. The loudspeaker is driven at ten frequency steps in the range of 50 – 500Hz with 50Hz intervals to cover the frequency range of normal vocal fold oscillations to obtain excitation pressure (sound) on the specimen surface. The acoustic pressure applied to the inner volume of the pipette enables to have an enclosed pressure on the surface of the silicone sample. The displacement of the vibration induced by the sinusoid fluctuating force generated by the acoustic pressure on the silicone sample surface inside the pipette walls is measured by a laser vibrometer. Thereby, the exciting pressure on the sample surface can be measured by a microphone and the resulting mechanical vibrations by a laser vibrometer. Since the wavelength of the sine wave with 500Hz maximum frequency is much longer than the dimension of the pipette, the pressure considered constant in all the pipette volume, therefore, the pressure on the surface of specimen is equal with the pressure sensed by the microphone positioned in the pipette. The sound pressure received by the microphone is the exciting pressure to calculate the module of elasticity. The displacement of the sample due to the acoustic pressure is in the range of one micrometer which demands a very accurate measurement method. Therefore, an OVF-5000 Xtra, the newest Polytec modular laser vibrometer is applied. A
helium-neon laser exploits the Doppler Effect to derive the velocity. The displacement is a sinusoidal movement and obtained by integrating over velocity, which is done by a Labview automation program. A sketch of the whole setup is depicted in (figure 2 (a)). Due to the small displacement, external influences must be kept to a minimum or remedied. Hence, the measurement setup and laser vibrometer sensor head were placed on a vibration isolated table to damp external vibrations and interferences.

3. Results
The bottom end of the pipette was placed on the surface of the specimen perpendicularly (figure 3 (a)). Due to the sinusoidal sound pressure in the pipette, the surface area enclosed by the pipette walls is aspirated into the pipette and pushed in (–z) direction on specimen (figure 3 (b)). The displacement wave (L) at the center of the pipette cross-section is optically measured as a function of the acoustic pressure (∆P). In this study, we assumed that the silicone samples used are isotropic and homogenous and has linear elasticity. Based on these assumptions and using the pipette aspiration technique, the linear equation (1) taken from Aoki [2] used to calculate Elasticity modulus E. The coefficient C is corresponding to the effect of the variations in pipette wall thickness (b/a), specimen thickness (h/a) and specimen radius (R/a) which form Aoki [2] plots (figure 4) we consider the coefficient 1.1 according to our pipette and silicone sample dimensions.

\[ \frac{L}{a} = C \frac{\Delta P}{E} \]

\[ E = C \frac{\Delta P}{L/a} \] (1)

![Figure 3](image-url) Pipette and sample: (a) Pipette Aspiration; (b) Schematic of the deformed sample under pipette.

![Figure 4](image-url) Effect of pipette wall thickness, specimen size and radius extracted from Aoki [2].
The elasticity modulus of all four samples summarized with respect to the frequencies at which they were excited are shown in figure 5. The results clearly show the change of the elasticity modulus over the considered frequency steps. For the specimen with 1:1:0 mixing ratio, an elasticity modulus of about 37 kPa is determined at an excitation frequency of 50 Hz (step 0) while in the same step, 25 kPa, 11 kPa and 8 kPa are shown by the 1:1:2, 1:1:3 and 1:1:4 sample respectively. The elasticity modulus raise to 210 kPa at an excitation frequency of 500 Hz (step 9) in the 1:1:0 specimen. The highest and the lowest elasticity modulus belongs to the 1:1:4 sample with maximum 450 kPa at 500 Hz and minimum 8 kPa at 50 Hz.

![Figure 5](image)

**Figure 5.** The Elasticity moduli of the four samples.

We measured the modulus of elasticity with various pressure amplitudes and it remained independent from the pressure amplitude diversity as it demonstrated in figure 6. In the chart (a), the elasticity modulus of the 1:1:3 specimen calculated based on disparate pressure amplitudes from around 1 Pa to 12 Pa for each frequency steps. While the chart (b) illustrated module of elasticity with unique pressure amplitude, 3 Pa for all frequency steps.

Like the previous studies which took into account, rather than the thickness and radius of the pipette walls which we assumed their effects by the coefficient (C), the elasticity modulus shows very high dependency to the force imposed on the silicone surface by the pipette walls. Moreover, since the
displacement is in range of nanometers or micrometers depend on the excitation pressure amplitude, and the sound pressure measured by the microphone should not be distorted, the environmental factors like as vibrations, temperature, noise and humidity are essential to get precise results.

Figure 6. Elasticity modulus with (a) disparate pressure amplitudes (b) unique pressure amplitude.

We changed the position of the laser spot between ten different locations in the area of a circle with a diameter of 3 mm on the center of the silicone surface in the pipette-enclosed area and measured the elasticity module for each of the laser spot positions for the ten frequency steps. Elasticity modules for the ten different laser spot positions are shown in (figure 7). The mean value and the standard deviation of the values in the first step and the last step of the 1:1:0 graph is (34.203 kPa, 2.75925 kPa) and (214.053 kPa, 5.525 kPa) respectively. The mean value and standard deviation of the 1:1:2, 1:1:3 and 1:1:4 elasticity modulus graphs over the 500 Hz frequency are (316.405 kPa, 37.795.5 kPa), (344.162 kPa, 53.585 kPa) and (562.654 kPa, 52.291 kPa) respectively. Since in this case, we are measuring off the center, the producing measurement error is very small except in the first step of 1:1:0 specimen and in some of the ending steps in all the other samples. Therefore, this method has a good quality of the measurement.

Figure 7. Measured elasticity module for 10 different laser center points of the for silicone sample.

It is clear that they represent a higher deviation at the higher elasticity modulus, in these steps we calculate high elasticity modulus values because of a small excitation in the first step of 1:1:0 specimen and a small displacement amplitude in the ending steps of the rest of the specimens. Moreover, the
equipment, measurement errors or the environmental conditions like as vibrations, noises, temperature, etc. might be interfere in the results and be the cause of the high deviations.

4. Conclusion
We have presented a measurement method to determine the elasticity modulus at specific frequency, for silicone materials as used for artificial vocal folds. Therewith, a sample is placed under a pipette, which is used to channel the acoustic pressure produced by a loudspeaker. With the help of a Labview program, where the measured excitation pressure as well as the displacement of the silicone surface is used as input data, the elasticity modulus is determined. The obtained elasticity modulus clearly shows an increase over frequency in all the three silicone mixtures. The elasticity moduli were found for the four different silicone mixing ratios in the range of 8kPa to 450 kPa. Extreme miniaturization and simplicity of the used measurement technique are the big advantages of this method with compare to the previously studied pipette aspiration method using pistonphone. Further work to develop this method needs to be done by comparing the results with numerical simulation and the silicone sample static elasticity measurement.

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