### **ORIGINAL RESEARCH**

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# Three-dimensional wideband absorbance immittance findings in young adults with large vestibular aqueduct syndrome

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### Abstract

Objective: To investigate the effect of large vestibular aqueduct syndrome (LVAS) on middle ear sound transmission using wideband absorbance immittance (WAI).

Methods: WAI results from young adult LVAS patients and normal adults were compared.

Results: Averaged energy absorbance (EA) at ambient and peak pressure in the LVAS group showed differences to the normal group. Under ambient pressure, the average EA of the LVAS group was significantly higher than the normal group at frequencies 472-866 Hz and 6169-8000 Hz (p < .05) and lower at frequencies 1122-2520 Hz (p < .05). Under peak pressure, absorbance was increased at frequencies 515–728, 841, and 6169-8000 Hz (p < .05) and decreased at 1122-1374 Hz and 1587-2448 Hz (p < .05). An investigation into the effect of external auditory canal pressure on EA across frequencies in the pressure-frequency domain, showed that EA differed significantly in the low-frequency region of 707and 1000 Hz from 0 to 200 daPa and 500 Hz at 50 daPa (p < .05). There was also a significant difference in EA between the two groups at 8000 Hz (p < .05) in the pressure range -200-300 daPa.

Conclusion: WAI is a valuable tool to measure the effect of LVAS on middle ear sound transmission. LVAS has a significant effect on EA at low and mid frequencies under ambient pressure, while the frequencies affected are mainly at low frequencies when positive pressure is presented.

Level of Evidence: Level 3a.

### KEYWORDS

energy absorbance, large vestibular aqueduct syndrome, wideband absorbance immittance

#### 1 | INTRODUCTION

Large vestibular aqueduct syndrome (LVAS) is characterized by an enlarged vestibular aqueduct (EVA) and sensorineural hearing loss (SNHL). In the majority of patients, the SNHL is bilateral, fluctuating, and progressive. A minority of patients have a stable SNHL. LVAS is diagnosed primarily on the basis of computed tomography (CT) or magnetic resonance imaging (MRI), that shows an abnormal VA.<sup>1</sup>

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Hearing loss in LVAS is generally progressive and fluctuating, with approximately 34% of LVAS patients suffering sudden drops in hearing thresholds after minor head trauma, barotrauma, or noise trauma.<sup>2</sup> The proposed mechanism underlying sudden drops in hearing thresholds is that head trauma induces a rapid increase in intracranial pressure, which could pass through the EVA by the cerebrospinal fluid (CSF) to the endolymphatic ducts. A sudden pressure change in the endolymphatic duct could influence blood supply to the cochlea and Corti's organs and lead to hearing loss.<sup>3</sup>

It is generally agreed that enlargement of the skeletal structure of the vestibular aqueduct may lead to abnormal transmission of CSF pressure to the inner ear.<sup>4</sup>

A theory describing the pressure balance between endolymph and perilymph postulates that the endolymphatic sac transmits CSF pressure changes to the endolymph, while at the same time, the cochlear aqueduct transmits CSF pressure to the perilymph.<sup>5</sup> Ultimately, if CSF pressure increases, then both endolymphatic and perilymphatic pressure will increase in response. Therefore, there are a couple of further rationales to explain the influence of increased CSF pressure on hearing fluctuations: (1) impairment of hair cell function by altering endolymphatic pressure; and (2) impact of increased perilymphatic pressure on movement of the stapes footplate.

In addition, to the mechanisms proposed above, several studies have highlighted the hearing loss caused by endolymphatic hydrops in LVAS patients. Sone et al.<sup>6</sup> used gadolinium-based MRI technique to investigate endolymphatic hydrops in LVAS patients. Their findings revealed in all ears a displacement of Reissner's membrane, or larger area of the cochlear duct than area of the vestibular scala. Also the area ratio of endolymph to the sum of endolymph and perilymph was greater than 1/3, indicating inner ear endolymphatic hydrops in these LVAS patients.<sup>6</sup> Scarpa et al.<sup>7</sup> also suggested that endolymphatic hydrops could lead to increased perilymphatic pressure, with decreased stapes mobility and increase in air conduction thresholds.

Murakami et al. reported that increased inner ear pressure would influence middle ear transfer function.<sup>8</sup> Consequently, noninvasive measurement of middle ear transfer function could provide a tool to determine the condition of the inner ear. Macrae et al. found that acoustic impedance at the TM was raised in living human subjects when intracranial pressure increased.<sup>9</sup> Tympanic membrane displacement could be used to evaluate intracranial pressure.<sup>10,11</sup>

Compared to the measurements described above, wideband absorbance immittance (WAI) has a wider range of stimulation frequencies, between 226 and 8000 Hz, which can provide more information about the middle and inner ear status in terms of acoustic energy absorbance (EA) across different frequencies at different pressures. Because of its rapid and noninvasive nature, WAI has been used as a sensitive diagnostic tool for the assessment of middle ear conditions in ENT and Audiology clinics. So far, the literature indicates that WAI can be used in newborn hearing screening,<sup>12</sup> and diagnosis of middle ear disorders, such as otitis media,<sup>13</sup> otosclerosis,<sup>14</sup> and ossicular discontinuity.<sup>15</sup> For example, Merchant et al.<sup>16</sup> found that WAI could distinguish the middle ear effusion with 100% accuracy and

differentiate full effusion from partial effusion, where there was no change in conventional tympanograms. Importantly, the WAI results were associated with the physical characteristics of the effusion. Won et al.<sup>17</sup> observed that the absorbance of mucus middle ear effusion was lower than that of serous middle ear effusion, especially above the 2 kHz region. In recent years, characteristic manifestations of WAI have been observed in superior semicircular canal dehiscence (SSCD) syndrome,<sup>18</sup> Meniere's disease,<sup>19</sup> and LAVS.<sup>20</sup>

A previous study found that WAI results in children with LVAS showed an increase in absorbance at low and high frequencies and a decrease at mid-frequencies.<sup>20</sup> More recent research has shown that the WAI can measure EA under varying pressure and frequency conditions, and that the current commercial three-dimensional (3D) WAI logs a large dataset. This rich dataset providing more detailed information as to the dynamic behavior of the middle ear.<sup>21</sup>

In this study, we used WAI to explore the effect of EVA on middle ear transfer function in young adults with LVAS who present with severe or profound deafness SNHL.

# 2 | MATERIALS AND METHODS

### 2.1 | Participants

A total of 19 ears from 10 young adult patients (two females and eight males) with LVAS were recruited in the Department of Otolaryngology Head and Neck Surgery, Beijing Tongren Hospital, Capital Medical University, China.

Inclusion and exclusion criteria were as follows:

- 1. no history of other acquired ear diseases (such as otitis media);
- 2. no acute or chronic upper respiratory inflammation;
- normal type-A 226 Hz tympanometry with peak pressure between -50 and +50 daPa;
- 4. normal TM and Eustachian tube function during participation.

The age range was 16–35 years (mean 26.8 years, standard deviation: 5.3 years). All participants underwent a routine otological examination, followed by WAI (Titan IMP440, Interacoustics) and a CT scan. Vestibular Aqueduct diameters (vertical and axial width at the midpoint between labyrinth and operculum) were greater than 1.5 mm.

Fourteen healthy volunteers (twenty-six ears) with normal hearing thresholds who also met the inclusion criteria were recruited as the control group. Anyone who had previously suffered from hearing fluctuations was excluded. The age range was 18–42 years (mean: 26.3 years, standard deviation: 6.4 years). After a routine otologic examination, all subjects in the control group had the WAI test to verify the normality of their external ear canal and tympanic membrane.

In compliance with ethical standards for human subjects, written informed consent was obtained from all participants before proceeding with the study. This study was approved by the Institutional



**FIGURE 1** An example of the three-dimensional (3D) wideband absorbance immittance (WAI) image and data preprocessing. (A) An example of 3D WAI data obtained from a participant with normal middle ear function aged 26. (B) The two-dimensional (2D) frequency-absorbance plot at the peak pressure obtained from the same participant. (c) The 2D frequency-pressure image converted from the 3D WAI in (A). (D) The 2D frequency-pressure image converted from the 3D WAI in (A) after interpolating the pressure values on the Y-axis

Review Board in Beijing Tongren Hospital, Capital Medical University (No. TRECKY2014-030).

### 2.1.1 | WAI measurement

The equipment used in this study was a commercial Titan IMP440 middle ear impedance device (Interacoustics, Denmark, Version 3.4) which can be used to measure middle ear acoustic EA from 226 to 8000 Hz at a pressure sweep from -300 to +200 daPa. The equipment was calibrated by Interacoustics annually in clinic, and calibration of the probe performed using a 2cc standard cavity provided by Titan. WAI measurements were performed at least three times for each ear to acquire reliable results. All participants were seated and quiet during the entire measurement.

# 2.1.2 | 3D WAI data processing and twodimensional pressure-frequency WAI images analysis

The mean EA at ambient and peak pressures was compared between patients in the LVAS and control groups. In two-dimensional (2D)

pressure-frequency WAI image analysis, a similar data processing and analysis method used in the study by Grais et al.<sup>21</sup> To summarize, in data processing, the X-axis frequency range was 226–8000 Hz at 1/24 octave intervals for a total of 107 points, while the pressure axis had a range of -300 to 200 daPa with 10 daPa intervals, and a total of 51 evenly distributed pressure points after interpolation of unevenly sampled pressure data among different subjects. An example of the WAI results of a normal person is shown to illustrate conversion of the image (Figure 1). For detailed descriptions and explanations of WAI data preprocessing, please read the methodology section in the study by Grais et al.<sup>21</sup>

### 2.2 | Data analysis

Data on age, gender, pure tone threshold, static acoustic compliance, and EA were analyzed using SPSS 22.0. Mean EA was calculated separately for the LVAS and control groups at different pressures and frequencies. After testing whether the data were normally distributed, an independent sample t test was used for the normal data, and a non-parametric test used for data that was not. Differences between the two groups were considered significant when the probability value p was less than .05.

				РТА			WAI
Case	Age	Gender	Side	HL (dB)	ABG250 (dB)	ABG500 (dB)	Compliance (ml)
1	27	М	L	93.75	35	30	0.48
			R	102.5	NA	NA	0.36
2	32	М	L	66.25	45	30	1.0
			R	77.5	30	35	0.5
3	28	М	L	83.75	50	35	0.3
			R	72.5	35	20	0.25
4	35	F	L	98.33	70	30	0.67
			R	61.25	50	20	0.64
5	21	М	L	117.5	NA	NA	0.26
			R	106.25	NA	NA	0.22
6	30	М	L	106.25	NA	45	0.43
			R	102.5	NA	NA	0.88
7	28	М	L	110	NA	NA	0.69
			R	91.67	NA	NA	0.58
8	26	М	L	107.5	75	40	0.38
			R	105	50	30	0.4
9	20	М	L	82.5	35	25	NaN
			R	101.25	35	NA	0.39
10	16	F	R	82.5	60	25	0.4

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Note: Hearing level is the average air conduction level of four frequencies 500, 1000, 2000, and 4000 Hz. NA: Because the subject remains unresponsive at this frequency when the bone conduction output is maximal, this makes it impossible to calculate the air-bone gap.

Abbreviations: ABG250, the air-bone gap at 250 Hz; ABG500, the air-bone gap at 500 Hz; HL, hearing level; LVAS, large vestibular aqueduct syndrome; PTA, pure tone audiology; WAI, wideband absorbance immittance.

### 3 | RESULTS

# 3.1 | Demographic information and audiological characteristics of the LVAS group

Table 1 summarizes the subject information, including demographic data, affected ear side, and hearing status. Subject 10 had a left ear Cochlear Implant. As a result, 19 ears were included.

Mean age of the LVAS group was 26.8 years (SD: 5.2 years), and the control group 26.3 years (SD: 6.4 years). The ages of the two groups were not statistically different in terms of age after nonparametric testing (p = .188). However, there was a statistically significant difference in gender between the two groups (p = .0005). However, no significant difference in EA was found between genders in the previous study.<sup>22</sup> As a result, the significant difference in gender between groups was considered unlikely to affect any comparison of WAI results.

# 3.2 | Characteristics of WAI in the LVAS and control groups (absorbance at ambient pressure and peak pressure)

Figures 2 and 3 show the absorbance at ambient and peak pressure in both groups. The results indicate that the EVA influenced middle ear



**FIGURE 2** Mean absorbance at ambient pressure against frequency for control and large vestibular aqueduct syndrome (LVAS) groups. Error bars are ±1 standard error of the mean (SE)

transmission function. The mean absorbance of the LVAS group at frequencies around 1000–2500 Hz was lower than the control group under peak and ambient pressure, indicating less efficient sound energy transmittance. Mean absorbance under ambient and peak pressure were higher than the control group below 1000 Hz and above 6000 Hz.

**TABLE 1**Demographic informationand audiological characteristics of theLVAS subjects that participated in thepresent study





**FIGURE 3** The mean absorbance at peak pressure against frequency for control and large vestibular aqueduct syndrome (LVAS) groups. Error bars are ±1 SE

Further statistical analysis showed that average absorbance under ambient pressure in the LVAS group was lower than the control group at frequencies 1122–2520 Hz and higher at frequencies 472–866 Hz and 6169–8000 Hz. Similar results were obtained at peak pressure with average absorbance increased at frequencies 515–728 Hz, 841 Hz, 6169–8000 Hz, and decreased at frequencies 1122–1374 Hz and 1587–2448 Hz. Differences in WAI at the frequency regions indicated above were significant (Table 2).

# 3.3 | 2D frequency-pressure WAI images analysis

Difference in absorbance between the two groups at different frequencies and pressures can be easily seen in 2D images. Figure 4A,B shows the average results of EA in the LVAS and control groups at

	Absorbance at ambient			Absorbance at peak		
Frequency (Hz)	LVAS	Control	p-Value	LVAS	Control	p-value
257	0.13	0.11	NS	0.13	0.12	NS
500	0.25	0.19	<.05	0.26	0.21	NS
749	0.15	0.13	<.05	0.14	0.17	NS
1000	0.50	0.51	NS	0.51	0.54	NS
1498	0.48	0.59	<.05	0.50	0.58	NS
2000	0.56	0.67	<.05	0.57	0.67	<.05
2520	0.67	0.75	<.05	0.67	0.74	NS
2997	0.75	0.75	NS	0.74	0.75	NS
4000	0.56	0.59	NS	0.55	0.59	NS
5993	0.45	0.36	NS	0.45	0.36	NS
8000	0.53	0.33	<.05	0.53	0.33	<.05

TABLE 2Comparison of absorbanceacross frequencies 250-8000 Hz atambient and peak pressures between theLVAS and control groups

Note: NS indicates not significant (p > .05).

Abbreviation: LVAS, large vestibular aqueduct syndrome.



**FIGURE 4** The comparison of two-dimensional (2D) frequency-pressure wideband absorbance immittance (WAI) images between normal and large vestibular aqueduct syndrome (LVAS) group. (A) Mean of absorbance contour plot at different frequencies and pressures in normal ears. (B) Mean absorbance contour plot at different frequencies and pressures in the ears with LVAS

different pressures and frequencies. It can be seen that the highest absorbance in both groups is mainly concentrated at frequencies between 2500 and 4000 Hz and at pressures of -50 to 150 daPa. However, the maximum average absorbance was higher in the control group than in the LVAS group, and its distribution area was also larger. There were some other differences in the EA between the two groups. At frequencies higher than 6000 Hz, the LVAS group had a higher EA than the normal group, regardless of pressure. At ambient pressure, the LVAS group had a lower mean EA than the normal group in the frequency range around 1000–2500 Hz. When the external auditory canal pressure was positive, and at 750–1000 Hz EA was higher in the LVAS group than in the normal group.

To allow comparison between the LVAS group and the control group regarding whether there is a significant difference in the EA of the 2D frequency-pressure profile and to better visualize the distribution of this difference, we resampled the X- and Y-axes using a 1/2 octave frequency band and a pressure step size of 50 daPa. Based on this, the *p*-value frequency-pressure graph was plotted (Figure 5). The absorbance of the two groups was significantly different in the blue area. Based on the half-octave and the 50 daPa profile, the total number of points was is  $11 \times 11 = 121$ , and the total number of non-significant points was 97 indicating that 19.83% of the area in the 2D frequency-pressure images shows a significant difference in absorbance. Table 3 summarizes the main differences between the LVAS and control groups in the low-, medium-, and high-frequency regions at negative, ambient, and positive pressures. In general, for the high-frequency region of 8000 Hz, there was a significant difference in EA



**FIGURE 5** The significantly different regions in the wideband absorbance immittance (WAI) data

between the two groups regardless of the force applied by the probe to the external auditory canal.

There was no significant difference in EA between the two groups of subjects when the probe applied negative pressure to the external ear canal. When the external ear canal pressure was ambient, there were significant differences in the low-frequency region at 500 and 707 Hz, and in the mid-frequency region at 1414 and 2000 Hz. When the external ear canal pressure was positive, significant differences existed in the low-frequency region at 500, 707, and 1000 Hz at 50 daPa.

### 4 | DISCUSSION

The common audiological features of LVAS are fluctuating progressive SNHL, together with ABG at low frequencies. Several studies suggest that this phenomenon is related to energy shunting from the third window of the inner ear into the cochlea, decreased impedance of the scala vestibule, increased inner ear pressure or endolymphatic hydrops.<sup>6,23,24</sup>

Since the VA connects CSF to the perilymph, pressure from the CSF can be transmitted to the perilymph when the VA is enlarged and not functioning properly. Reduced resonance frequencies can be observed in LVAS and Ménière's disease.<sup>24,25</sup> This also indicates an increased inner ear pressure in patients with LVAS. Increased inner ear pressure may also affect the function of the ossicular chain through the vestibular window. Abnormal pressure transmission breaks the impedance balance between endolymph and perilymph. Allen et al. observed a mismatch in impedance at frequencies below 2000 Hz in patients with otosclerosis, with a resultant decrease in absorbance.<sup>26</sup> The results in this study show a decrease in absorbance around middle ear resonance frequencies, that is, a significantly lower absorbance in the LVAS group than the control group at frequencies between 1100 and 2500 Hz. According to Wang et al., otosclerosis patients had significantly lower absorbance between 1260 and 2520 Hz than normal subjects.<sup>27</sup> Similarly decreased mid-frequency absorbance in patients with otosclerosis implies that increased inner ear pressure has an effect on the middle ear.

On the other hand, the EVA acting as a third window increases absorption of acoustic energy.<sup>23</sup> Tonndorf et al. referred to the vestibular aqueduct, cochlear aqueduct, vascular and neural channels as the third window, which they suggested was playing a role in transmitting acoustic energy under both physiological and pathological conditions.<sup>28</sup>

TABLE 3	The significantly	different regions betw	ween LVAS group and the	control group
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	Low-frequency region	Mid-frequency region	High-frequency region
Negative pressure	All insignificant	All insignificant	Significant in 8000 Hz between 0 and $-300\ \text{daPa}$
Ambient pressure	Significant in 500 and 707 Hz	Significant in 1414 and 2000 Hz	Significant in 8000 Hz
Positive pressure	Significant in • 500 Hz at 50 daPa • 707 Hz between 0 and 200 daPa • 1000 Hz between 0 and 200 daPa	All insignificant	Significant in 8000 Hz between 0 and 200 daPa

Abbreviation: LVAS, large vestibular aqueduct syndrome.

SSCD is an audiovestibular disease caused by a third window. The abnormal third window in this case affects the reflection and absorption of acoustic energy.<sup>18</sup> Since both SSCD and LVAS are structurally inner ears with irregular channels, they may have similar effects in terms of acoustic energy absorption. Indeed, Nakajima et al.<sup>29</sup> observed decreased reflectance at frequencies 750-1000 Hz in patients with SSCD. Their results in keeping with the results found in patients with LVAS in this study, that is, absorbance at low frequencies increased. In the study by Rosowski et al.,<sup>30</sup> tympanic membrane vibration in patients with SSCD was greater than the average value of normal ears at frequencies below 1500 Hz. It was suggested that this might be related to the decreased impedance caused by dehiscence and be influenced by the size of the dehiscence. This mechanism may also explain the increased absorbance at low frequencies found in our experiment. The EVA diverted some of the acoustic energy and made the energy transmission to the inner ear easier. Similar results were observed in patients with endolymphatic hydrops. After performing WAI in 32 ears diagnosed with endolymphatic hydrops by MRI. Kobayashi et al.<sup>31</sup> found that low-frequency (560-600 Hz) absorbance was significantly higher for ears with significant endolymphatic hydrops compared to those with mild hydrops or no hydrops. Endolymphatic hydrops may have shunted a portion of the incoming acoustic energy and reduced the energy flow to the cochlea.

Overall, the effect of LVAS on middle ear acoustic absorbance appears complex. The EVA not only transmits pressure from the CSF to the inner ear, but also shunts sound energy to the inner ear. This may also be related to the frequency of the sound. Cheng et al.<sup>32</sup> concluded that the acoustic impedance of SCD increases with frequency. Therefore, at low frequencies, a small impedance allows the sound to be shunted through the SCD, and conversely for high frequencies, an excessive impedance prevents the sound from being shunted.<sup>32</sup> Since the EVA and SCD are both third windows, this change in impedance may also apply to LVAS.

An increase in high-frequency absorbance is also observed in the present study, in agreement with Wang et al.<sup>27</sup> Their results showed that Chinese patients with otosclerosis had higher EA than normal controls above 6350 Hz.<sup>27</sup> Although WAI measurements cover a frequency range from 226 to 8000 Hz, the significance of high frequencies for diagnosis is not clear. In a study of middle ear disease on high-frequency reflectance, Merchant et al.<sup>33</sup> found that reflectance at frequencies above 4-6 kHz was not sensitive in the diagnosis of middle ear disease. This variation with high frequencies may be related to standing waves. Since the ear canal needs to be sealed with a microphone probe when performing WAI testing, standing waves may form in the ear canal due to the reflection by the tympanic membrane. These standing waves can affect the results of the test and lead to errors. In the study of standing wave errors, Richmond et al.<sup>34</sup> observed that variance due to the standing wave effects was greatest at about 4 kHz and smaller at 8 kHz. When Lawton and Stinson<sup>35</sup> estimated the acoustic energy reflectance at the tympanic membrane using standing wave patterns in the ear canal, they identified significant individual differences above 7 kHz. Motallebzadeh et al.<sup>36</sup> also considered that the high-frequency resolution of WAI is associated

with resonance of the middle ear cavity and standing-waves in the ear canal. In this study variation under high frequencies may also be related to unreliability of the high-frequency results. Using evanescent modes for reflectance measurements, Nørgaarda et al.<sup>37</sup> found that absorbance above 3000 Hz did not accurately reflect the function of the middle ear. In compensated evanescent mode, it is observed that absorbance at high frequencies is susceptible to the effect of residual ear canal length and the angle of the inserted ear-probe.<sup>38</sup> Keefe<sup>39</sup> also observed variance in reflectance above 4000 Hz using a causal constraint procedure to measure acoustic reflectance. Sundgaard et al.<sup>40</sup> thought that WBT results above 4000 Hz were susceptible to noise, which may explain this unreliability. These results indicate a degree of uncertainty with the high-frequency acoustic reflectance results. Therefore, WAI results at high-frequency regions in patients with LVAS need to be further investigated in the future studies.

In comparison to the absorbance-frequency curves at ambient pressure and peak pressure, the 3D and corresponding 2D pressurefrequency WAI graphs contained more information, especially regarding the effect of pressure. The results of this study suggest that sound transmission in the middle ear changes at different pressures. Regardless of the pressure in the external auditory canal, EA showed a gradual decrease as pressure increased in both groups of participants. Feeney et al. also found that both +200 daPa and -200 daPa ear canal pressures caused a decrease in absorbance from 250 to 3000 Hz.<sup>41</sup> However, discrepancies in the effects of positive and negative pressure were observed when comparing the EA performance of the two groups at different pressures. As shown in Figure 5, the regions where significant differences exist are mainly concentrated in the range of positive pressure, but when the pressure is less than -150 daPa, there is no significant difference between the absorbance of the two groups except for high frequencies. Dirckx et al.42 observed a corresponding displacement of the tympanic membrane in cadavers when the external ear canal pressure changed. Koike et al.<sup>43</sup> had a similar result using a finite element model and demonstrated that when the external ear canal pressure is negative, that is, when the external ear canal pressure is less than the middle ear pressure, the tympanic membrane will be displaced outward. Not only the tympanic membrane but also the change in pressure in the external auditory canal affects the displacement of the stapes, causing the oval window membrane to shift toward the inner ear or toward the middle ear, resulting in a change in the displacement of the perilymph in the fistula.44

Therefore, we speculate that the reason for the uneven distribution of significant differences may be that external ear canal pressure affects the mobility of the ossicular chain and tympanic membrane. Pressure on the ear canal may lead to stiffening of the middle ear structures through tension.<sup>45</sup> As the positive pressure in the external ear canal given by the probe gradually increases, the tympanic membrane and the ossicles are applied with increasing force toward the inner ear.<sup>25</sup> Therefore, they gradually lose their mobility and become tightly connected. At this time, the force from the inner ear is better transmitted without the cushioning effect of the middle ear, which may provide a better expression of the impact on sound absorbance from WAI. In contrast, when the pressure in the external ear canal is negative, the tympanic membrane and the ossicular chain receive an outward pulling force.<sup>43</sup> When this force is excessive, the distance between them will increase, and they will have better mobility toward the tympanic membrane. In this case, the force from the inner ear is also directed outward, whereas it is cushioned by the activity of the tympanic membrane and the ossicular chain. That is, the effect of the inner ear on sound absorbance becomes insignificant under negative pressure conditions. However, even with positive pressure, when excessive pressure in the external ear canal is applied, and the force caused by the pressure far exceeds the force transmitted from the inner ear. Some small effects caused by changes in inner ear pressure may be masked from the presentation, such as differences in midrange frequencies that are not observed at pressures greater than 50 daPa.

To investigate whether there was a correlation between WAI results and pure tone hearing threshold results (air conduction threshold, bone conduction threshold, and air-bone gap) in the LVAS group subjects, we observed that the EA at some frequencies were significantly correlated with the PTA results, especially between the bone conduction (BC) hearing threshold at 250 Hz and the absorbance at 257, 500, 1000 Hz. However, the specific association between WAI and pure tone hearing thresholds is currently unclear, and this part of the results requires more evidence to make sense of it. Therefore, this part of the outcomes was not presented in this study.

The limitations of the present study were the small sample size and the gender differences between the two groups of subjects. Although Shahna et al.<sup>46</sup> concluded that gender had an effect on WAI results, there are studies suggesting that gender has no significant effect.<sup>22,47</sup> Even if gender has an effect, the affected frequency regions are various in different studies. In the study by Malanz et al.,<sup>48</sup> gender influenced absorbance at 280–790 Hz and 2830–4490 Hz, but Shahna et al.<sup>46</sup> found the affected frequencies to be 4000 and 5000 Hz. Considering the uncertainty of the effect of gender differences on WAI results and the sample size, we did not segregate by gender. To clarify this issue, we will expand our sample size to investigate the effect of gender differences on WAI outcomes in Chinese populations in further studies.

### 5 | CONCLUSION

The present study shows significant differences in absorbance at several frequency regions between the LVAS and normal groups, indicating that EVA influences middle ear sound transmission and that this influence can be detected with the WAI. 3D-WAI could provide more information about middle ear transfer function, especially the impact of pressure. The differences in EA between the two groups are mainly in the positive pressure in low-frequency regions, the ambient pressure in low, mid-frequency regions as well as high-frequency regions. The absorbance value shows differences across frequency intervals, suggesting that the mechanism by which EVA affects middle ear sound transmission is complex.

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### **CONFLICT OF INTEREST**

The authors declare that there are no conflicts of interest regarding the publication of this article.

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