

## Research Article

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# The Influence of Selection Criteria for Healthy Subjects on PLAI Measurements

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## Abstract

The PLAI measurement method's specificity varies depending on the following parameters and findings from various diagnostic tests in healthy individuals: otoscopy, audiometry, tympanometry, the Valsalva and Toynbee maneuvers, and pneumatic otoscopy.

This prospective, single-center study aims to identify the examinations that must be performed on individuals who report normal hearing and no ear complaints to achieve a specificity of 90% or higher for the PLAI method.

**Materials and Methods:** A total of 75 healthy volunteers (145 ears) were recruited, all of whom reported normal hearing and no history of ear problems. Each participant underwent a pure-tone audiometry, bilateral video endoscopy, the Valsalva and Toynbee maneuvers, pneumatic otoscopy, PLAI measurement of both ears, 226-Hz tympanometry, otoacoustic emissions, and ipsilateral stapedia reflex measurement. In participants with one-sided absent stapedia reflexes, both ipsilateral and contralateral reflexes were measured using Medwave®, and monosyllabic word tests were performed.

**Results:** The PLAI data analysis indicates that the distribution and dispersion of the measured values change slightly as more tests are conducted on healthy subjects and as more data from ears with pathological findings are excluded from the study. The results clearly demonstrate that as the number of tests used to classify the "healthy ear" condition increases, the PLAI method's diagnostic specificity also increases. Based on the study's questionnaire, the specificity can be enhanced from 77% to 96%. To achieve this, besides the questionnaire, pure-tone audiometry, otoscopy, clinical Eustachian tube function tests, tympanometry, and stapedia reflex measurement must also be included.

**Conclusion:** This prospective, single-center study of healthy individuals shows that at least pure-tone audiometry, otoscopy, and the Valsalva maneuver must be performed to rule out obvious pathologies of the tympanic membrane and middle ear, as well as Eustachian tube dysfunction, thereby reducing the dispersion of the PLAI measurements and achieving a diagnostic specificity of 90%.

## Introduction

Broadband absorbance measurement and classic single-frequency tympanometry provide parameters for further classification of normal hearing, including an assessment of middle

ear function. Wideband absorbance (WBA) is the proportion of energy absorbed into the middle ear compared to the incident energy presented from the probe. Many studies have demonstrated

a significant advantage of this technology over conventional single-frequency tympanometry in newborns for detecting middle ear effusion [1-5]. Additionally, several studies have suggested that it may be a more accurate predictor of conductive conditions than 226-Hz tympanometry in adults [4] and 1000-Hz tympanometry in infants aged 0–6 months [1]. Moreover, WBA testing has been demonstrated to be more effective than tympanometry in detecting specific pathologies, including otitis media with effusion (OME) and otosclerosis [4, 6].

To understand the measurements taken during middle ear diagnostics, knowing the values for healthy subjects with normal hearing is essential [7, 8]. Sole reliance on air-conduction audiometric data to determine normal hearing does not always indicate normal middle ear function [9].

The method for PLAI measurements, implemented in the Medwave® device, has been described in detail elsewhere [10]. However, a summary is provided here.

PLAI measures acoustic admittance across a frequency range of 100–3000 Hz without inducing pressure changes in the outer ear canal. By stimulating the ear with a multifrequency acoustic signal, the PLAI device estimates the energy transmitted versus reflected by the middle ear [7, 8, 11, 12]. The resonance frequency and bandwidth of the resonant system reflect the mass and stiffness of the tympanic membrane and the ossicular chain complex, as well as damping factors in the middle ear and the effective geometry and volume of the external auditory canal [10].

Importantly, the resonance frequency represented is not that of the middle ear itself but that of the admittance, which changes due to alterations in the tympanic membrane and/or the middle ear.

The specificity of the PLAI measurement method varies depending on the parameters considered and the findings from

various diagnostic tests in healthy subjects, including otoscopy, audiometry, and tympanometry, the Valsalva and Toynbee maneuvers, and pneumatic otoscopy [7, 8, 13–15].

The availability of additional diagnostic tests facilitate the classification of the PLAI measurement. An algorithm is needed that incorporates findings from these diagnostics alongside the PLAI during evaluation.

This study hypothesizes that the specificity of the PLAI method increases and the dispersion of normal data decreases when some or all of these tests are performed and normal findings are obtained. For this purpose, specificity is calculated after each test section and in combination. The study aims to identify the examinations that must be performed on individuals who report normal hearing and no ear complaints to achieve a specificity of 90% or higher for the PLAI method.

## Materials and Methods

### Participants

Based on the sample size analysis, 75 healthy volunteers (145 ears) were recruited, all of whom reported having normal hearing and no history of ear problems. These included staff members of the KMG Clinic, students from the School of Surgical Assistants, companions of patients who had come to the ENT Clinic, as well as patients who had come to the clinic for other conditions affecting the nose and throat. Table 1 presents the demographic data. Inclusion criteria were age between 18 and 80 years, subjectively normal hearing, no ear complaints, and no prior middle ear or nasal surgery. Each ear was examined separately.

Overall, the study included 75 participants with a mean age of  $39.8 \pm 15.6$  years (47 women and 28 men). A total of 145 ears were examined (72 right ears and 73 left ears).

**Table 1:** Demographic data of participants (category 0).

Characteristics	Participants without any ear complaints and with normal hearing (medical history)
Number	
N/N <sub>ears</sub>	75/145
Age of participants <sup>a</sup>	
Min–Max <sup>a</sup>	18–80
Mean $\pm$ Std. deviation <sup>a</sup>	39.8 $\pm$ 15.6
Gender	
Female	47 (62.7)
Side	
Right ear	72 (49.7)

<sup>a</sup> in years.

## Procedures

The following criteria were used to examine the dispersion of the PLAI measurements depending on the selection criteria for healthy study participants: (1) normal hearing in pure-tone audiometry and otoacoustic emissions (OAE); (2) otoscopy (normal tympanic membrane and external auditory canal); (3) normal tube ventilation and exclusion of fixation of the ossicular chain (positive Valsalva and Toynbee maneuvers and normal pneumatic otoscopy), (4) tympanometry (tympanogram type A only); and (5) normal ipsilateral stapedial reflexes.

All 75 participants underwent pure-tone audiometry, bilateral video endoscopy, the Valsalva and Toynbee maneuvers, pneumatic otoscopy, the PLAI measurement of both ears, 226-Hz tympanometry, otoacoustic emissions, and ipsilateral stapedial reflex measurement.

After each examination, participants who exhibited pathological findings were excluded from further data analysis. The first group was designated as Category 0 and included 145 ears from the 75 study participants. Following pure-tone audiometry and OAE testing, participants with either hearing loss of 20 dB or more or abnormal OAE measurements were excluded. This reduced the data set to 129 participants in Category 1. During video otoscopy, all pathological findings—such as retractions of the tympanic membrane or calcifications—known to influence the PLAI measurements were excluded [13]. In one participant, debris from the external auditory canal was removed, and all measurements were repeated. This left 111 ears (Category 2) for further data analysis. For Category 3, all ears in which the Valsalva and/or Toynbee maneuvers were negative were excluded. Pneumatic otoscopy ruled out malleus fixation in all 145 ears and an adhesive process in cases of mild retraction. Pathological tympanic membrane findings had already been excluded during video otoscopy. For Category 4, only type A tympanograms were used for further analysis; all measurements of other tympanogram types were excluded. Using stapedial reflex measurement, those ears were excluded in which no ipsilateral stapedial reflexes were detectable in the affected ear despite normal findings in Categories 1–4. This left 72 ears for the evaluation of Category 5 (Figure 1). In cases where very high or very low resonance frequencies were measured, or where OAEs or stapedial reflexes were absent, the tests were repeated within 10 minutes to rule out measurement errors.

Data for the prospective study were collected between February and March 2026.

## PLAI measurements

The hand-held probe of the Medwave® device is inserted into the external auditory canal without manipulating ear canal pressure, meaning that sealing the ear canal is unnecessary.

A multifrequency stimulus (65 dB HL) is delivered over a range of approximately 100–3000 Hz. Particle velocity is measured at two closely spaced points in the external auditory canal. Complex admittance is derived from the ratio of sound pressure to particle velocity as a function of frequency [11, 12]. The resulting admittance curve displays a resonance peak that is highly correlated with middle ear properties [16] and, consequently, with middle ear

pathology [13, 16].

The hand-held probe connects to a touchscreen device. Medwave® has established age-specific reference values for PLAI measurements in healthy ears [17]. Core parameters are extracted from the frequency-domain curve rather than from a pressure-admittance loop. Current clinical implementations typically extract a compact set of scalar parameters from the admittance magnitude. For clinical application, the resonance frequency ( $F_{res}$  or  $F$ ) and peak of admittance (Peak) are routinely reported. For research purposes, additional factors were derived from the measurements and used for analyses, including acoustic ear volume ( $V_{ea}$ ), bandwidth height ( $B_{nd}$ ), and quality factor from bandwidth ( $Q$ ).

### 1 Pure tone audiometry and otoacoustic emissions

All audiological assessments were conducted in an audiometric, sound-attenuated room using calibrated signals and equipment according to accepted ISO standards. For classification of participants, pure-tone audiometry was performed to measure air-conduction (AC) and bone-conduction (BC) thresholds. The PTA4 was measured across the frequencies of 0.5, 1, 2, and 4 kHz, and the air-bone gap was calculated.

### 2 Otoscopy-based evaluation of the outer ear canal and tympanic membrane

For otoscopy, a 0° endoscope was used.

### 3 Otoscopy-based evaluation of the Valsalva and Toynbee maneuver and pneumatic otoscopy

For pneumatic endoscopy, a 0° endoscope was converted into a pneumatic otoscope, as described by Pau and Strenger [18, 19].

### 4 Conventional 226-Hz tympanometry

Tympanometry was performed using the Madsen® Zodiac (type 1096; Otometrics) at 226 Hz, over a pressure range of -400 to +200 daPa. The following measures were used for calculation: tympanometric peak pressure (TPP), tympanic width (TW), equivalent ear canal volume (ECV), and static peak compliance (SC).

### 5 Tympanometry-based stapedial reflex measurement

Ipsilateral stapedial reflex measurement was performed using the Madsen® Zodiac (type 1096; Otometrics).

In participants with one-sided absent stapedial reflexes, both ipsilateral and contralateral reflexes were measured using Medwave®, and monosyllabic word tests were performed.

For PLAI-based stapedial reflex measurement, a baseline PLAI measurement is recorded at moderate intensity. Subsequently, a stapedial reflex-eliciting stimulus, either ipsilateral or contralateral, is delivered [17]. A second PLAI measurement is recorded during or after the reflex, thus resulting in a frequency shift toward higher frequencies, changes in peak values, and alterations in bandwidth and band admittance height.

Speech intelligibility tests were conducted in the free field using the Freiburg monosyllabic word test—in quiet and in noise—through headphones at 60 dB, 80 dB, and 100 dB SPL. The loudspeakers were positioned 1 m away from the participant's head. In quiet, i.e., under S0 condition, speech intelligibility was

measured at 65 dB SPL. In noise, i.e., under S0N0 condition, speech intelligibility was measured with a fixed noise level of 60 dB SPL, and speech level of 65 dB SPL, resulting in a signal-to-noise ratio (SNR) of 5 dB.

Ethical considerations

The study design was approved by the Ethics Committee of the General Medical Council of Mecklenburg–West Pomerania (A2026-0063).

The study was conducted in strict adherence to the revised version of the Helsinki Declaration. All subjects were informed about the study’s objectives, and they provided their written consent for the measurements and publication of the data.

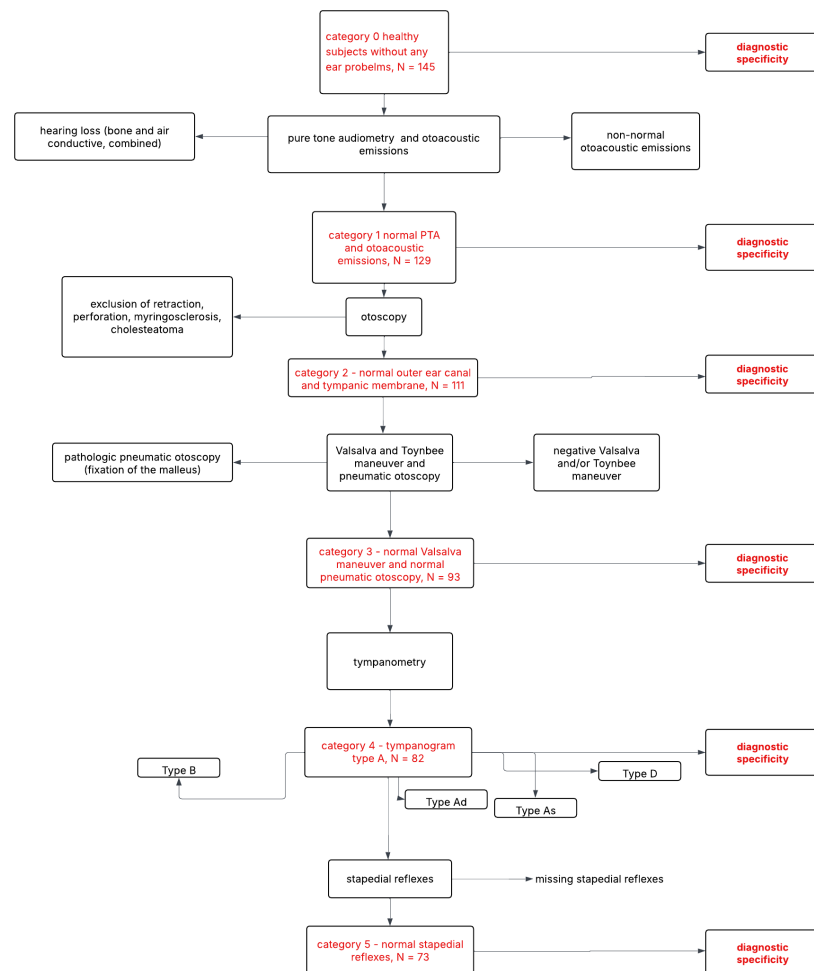
Statistical analyses

SPSS version 30.0 (SPSS Inc., Chicago, IL, USA) was used for statistical analyses. Data were evaluated using descriptive statistical methods and comparative tests. Numerical data were

expressed as mean ± standard deviation, and categorical variables were presented as percentages.

The diagnostic specificity (true negative rate) as the probability of a normal (negative) PLAI test result was calculated after each test section (Category 0-5), conditioned truly being negative. True negative means healthy people correctly identified as normal with PLAI.

Statistical tests (Kolmogorov–Smirnov test and Shapiro–Wilk test) indicated a non-normal distribution for all parameters (all p-values < 0.05). Nonparametric tests were performed for group comparisons whenever appropriate. Spearman correlation coefficients were used for correlational analyses. A p-value < 0.05 was considered statistically significant in all analyses. It was hypothesized that resonance frequency values change as hearing age increases. A nonlinear regression line (Epanechnikov kernel, incorporating 75% of all data points) was fitted to the results to describe the distribution of the data (Figures 2 and 3).

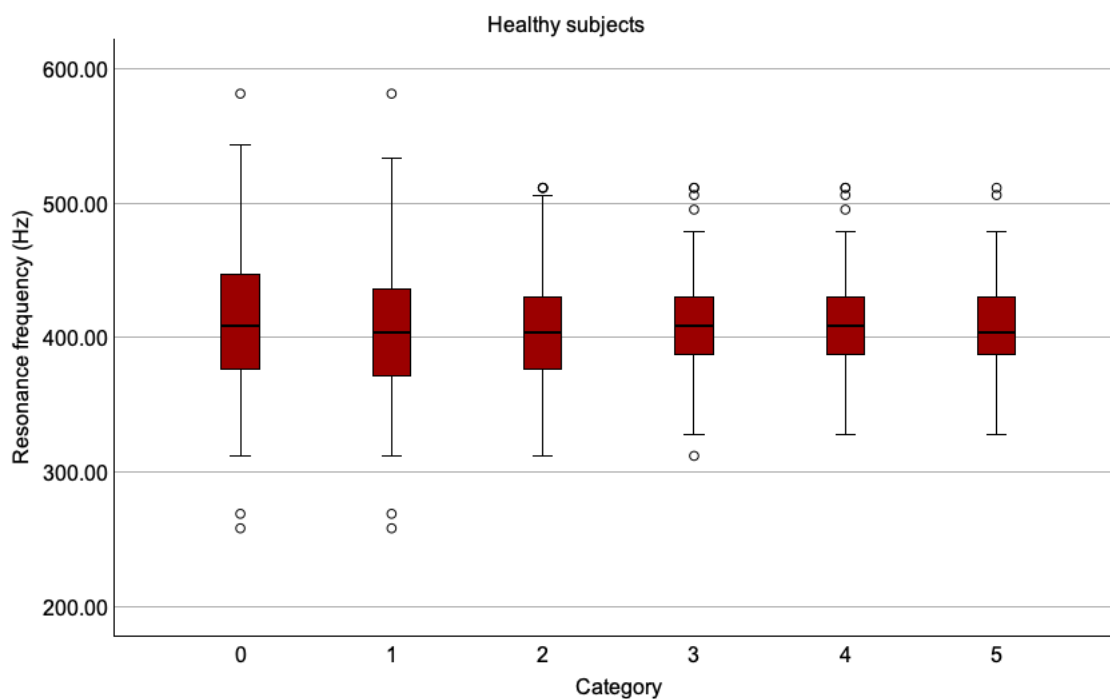


**Figure 1:** Flow chart for the selection criteria. Categories 0–5: 0 – PLAI diagnosis in participants without any ear complaints and with normal hearing (medical history); 1 – normal audiogram and otoacoustic emissions; 2 – otoscopy (normal outer ear canal and tympanic membrane); 3 – normal Valsalva and Toynbee maneuvers and normal pneumatic otoscopy; 4 – normal tympanometry (tympanogram type A); 5 – present stapedial reflexes.

## Results

PLAI data analysis shows that the distribution and dispersion of the measured values change slightly as more tests are conducted on healthy subjects and as more data from ears with pathological

findings are excluded from analyses (Table 2 and Figure 1). This effect is most noticeable in the resonance frequency. While the mean value fluctuates around 410 Hz across all categories, extreme values are filtered out as the number of examinations increases. The standard deviation decreases significantly.



**Figure 1:** Box plot presenting group medians and distribution of resonance frequency of admittance values among healthy subjects depending on the category.

Categories 0–5: 0 – PLAI diagnosis in participants without any ear complaints and with normal hearing (medical history); 1 – plus normal audiogram and otoacoustic emissions; 2 – plus otoscopy (normal outer ear canal and tympanic membrane); 3 – plus normal Valsalva and Toynbee maneuvers and normal pneumatic otoscopy; 4 – plus normal tympanometry (tympanogram type A); 5 – plus present stapedial reflexes.

To determine why the variation in the measured values does not decrease further despite the exclusion criteria—given that outliers remain that cannot simply be removed—the influence of age on the PLAI measured values was investigated. The admittance resonance frequency values for Categories 0 and 5 are presented as examples

(Figures 2 and 3). High and low values are observed at every age and cannot be explained by ear pathologies detectable using the measurement methods of Categories 1–5. In both Categories 0 and 5, a significant but weak correlation was found between the PLAI measures Peak, Q, and Bnd and age (Table 3).

**Table 2:** PLAI Diagnosis in participants without any ear complaints and with normal hearing (medical history), regardless of the diagnosis ( $N_{\text{ears}} = 145$ ).

PLAI Parameter						
Category	0	1	2	3	4	5
N	145	129	111	93	82	73
<b>F (Hz)</b>						
Mean $\pm$ SD	414 $\pm$ 57.1	408 $\pm$ 54.1	407 $\pm$ 45.7	410 $\pm$ 43.6	411 $\pm$ 41.5	409 $\pm$ 39.0
<b>Peak (<math>10^{-2}</math> mmho)</b>						
Mean $\pm$ SD	1.55 $\pm$ 0.32	1.55 $\pm$ 0.32	1.53 $\pm$ 0.29	1.52 $\pm$ 0.28	1.51 $\pm$ 0.26	1.48 $\pm$ 0.25
<b>Q</b>						
Mean $\pm$ SD	1.89 $\pm$ 0.52	1.89 $\pm$ 0.52	1.86 $\pm$ 0.45	1.84 $\pm$ 0.43	1.85 $\pm$ 0.41	1.79 $\pm$ 0.37
<b>Vea (mL)</b>						
Mean $\pm$ SD	1.47 $\pm$ 0.48	1.52 $\pm$ 0.48	1.50 $\pm$ 0.39	1.47 $\pm$ 0.37	1.46 $\pm$ 0.35	1.47 $\pm$ 0.33
<b>Bnd (<math>10^{-2}</math> mmho)</b>						
Mean $\pm$ SD	0.80 $\pm$ 0.20	0.80 $\pm$ 0.20	0.79 $\pm$ 0.19	0.78 $\pm$ 0.19	0.79 $\pm$ 0.18	0.76 $\pm$ 0.13

F – resonance frequency of admittance, Peak – peak of admittance, Q – quality factor derived from bandwidth, Vea – acoustic ear volume, Bnd – bandwidth height

Categories 0–5: 0 – PLAI diagnosis in participants without any ear complaints and with normal hearing (medical history); 1 – plus normal audiogram and otoacoustic emissions; 2 – plus otoscopy (normal outer ear canal and tympanic membrane); 3 – plus normal Valsalva and Toynbee maneuvers and normal pneumatic otoscopy; 4 – plus normal tympanometry (tympanogram type A); 5 – plus present stapedial reflexes.

**Table 3:** Correlational analyses of Categories 0 and 5 ( $N_{\text{ears}} = 145$  and 73, respectively).

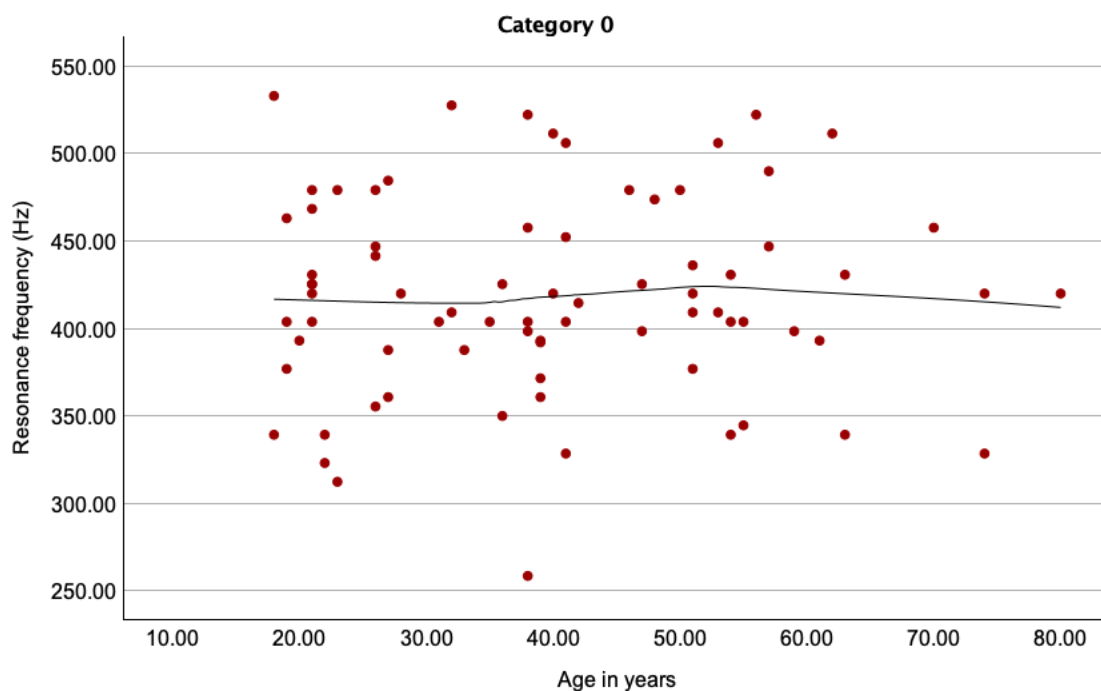
Category 0						
Spearman		F	Peak	Q	Vea	Bnd
Correlation Coefficient		0.064	0.232**	0.221**	-0.025	0.227**
<b>Sig. (2-tailed)</b>		0.447	0.005	0.008	0.768	0.006
<b>N</b>		145	145	145	145	145
Category 5		F	Peak	Q	Vea	Bnd
Spearman Correlation Coefficient		0.029	0.240*	0.233*	-0.027	0.245*
<b>Sig. (2-tailed)</b>		0.807	0.041	0.048	0.821	0.037
<b>N</b>		73	73	73	73	73

F – resonance frequency of admittance, Peak – peak of admittance, Q – quality factor derived from bandwidth, Vea – acoustic ear volume, Bnd – bandwidth height

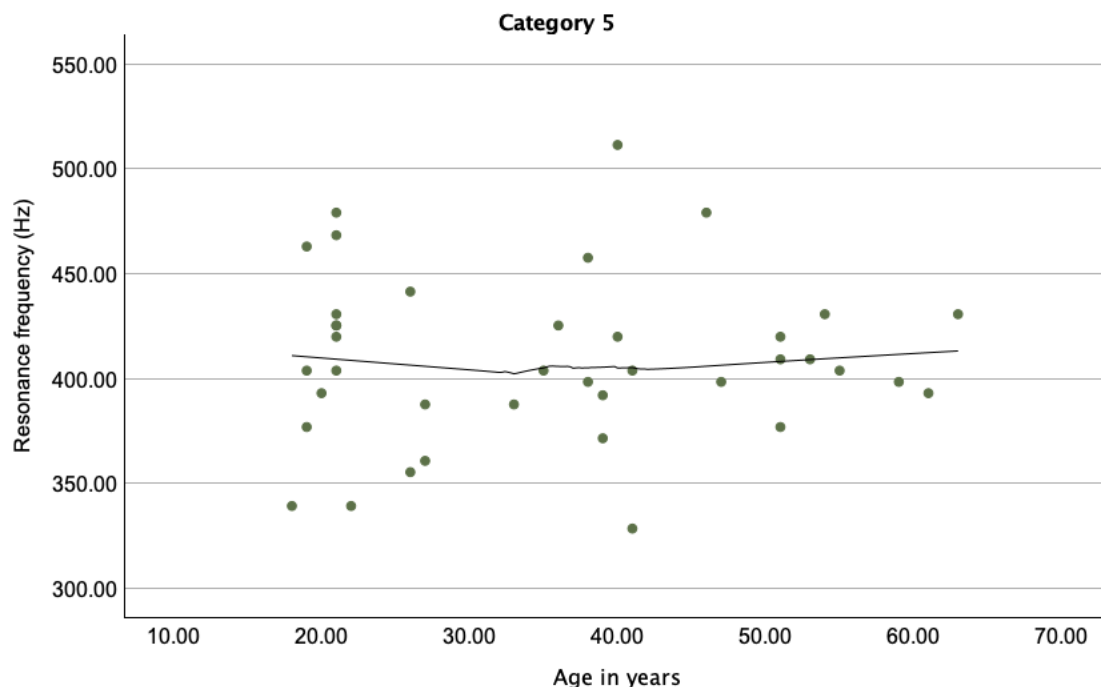
Categories 0–5: 0 – PLAI diagnosis in participants without any ear complaints and with normal hearing (medical history); 1 – plus normal audiogram and otoacoustic emissions; 2 – plus otoscopy (normal outer ear canal and tympanic membrane); 3 – plus normal Valsalva and Toynbee maneuvers and normal pneumatic otoscopy; 4 – plus normal tympanometry (tympanogram type A); 5 – plus present stapedial reflexes.

\* Correlation is significant at the 0.05 level (2-tailed).

\*\* Correlation is significant at the 0.01 level (2-tailed).



**Figure 2:** Resonance frequency of admittance in 145 healthy subjects of Category 0 depending on life age. Category 0: diagnosis in participants without any ear complaints and with normal hearing (medical history).



**Figure 3:** Resonance frequency of admittance in 145 healthy subjects of Category 0 depending on life age. Category 5: diagnosis in 73 participants without any ear complaints and with normal hearing (medical history) with normal findings in pure-tone audiometry and otoacoustic emission, tympanometry, the Valsalva and Toynbee maneuvers, pneumatic otoscopy, and stapedial reflex measurement.

For each category (0–5), the following calculation was performed to determine specificity.

**Table 4:** PLAI Diagnosis in participants without any ear complaints and with normal hearing (medical history), regardless of the diagnosis ( $N_{\text{ears}} = 145$ ).

PLAI Diagnosis						
$N_{\text{ears}}$	145	129	111	93	82	73
category	0	1	2	3	4	5
	Total/Percentage (%)					
No results	0/0	0/0	0/0	0/0	0/0	0/0
Perforation/soft	9/6.2	9/7.0	6/5.4	3/3.2	1/1.2	1/1.4
Normal	112/77.2	105/81.4	96/86.5	84/90.3	77/93.9	70/95.9
OME*/rigid	24/16.6	15/11.6	9/8.1	6/6.5	4/4.9	2/2.7
Total	145/100	129/100	111/100	93/100	82/100	73/100
Diagnostic Specificity	77.2	81.4	86.5	90.3	93.9	95.9

\*OME: Otitis media with effusion

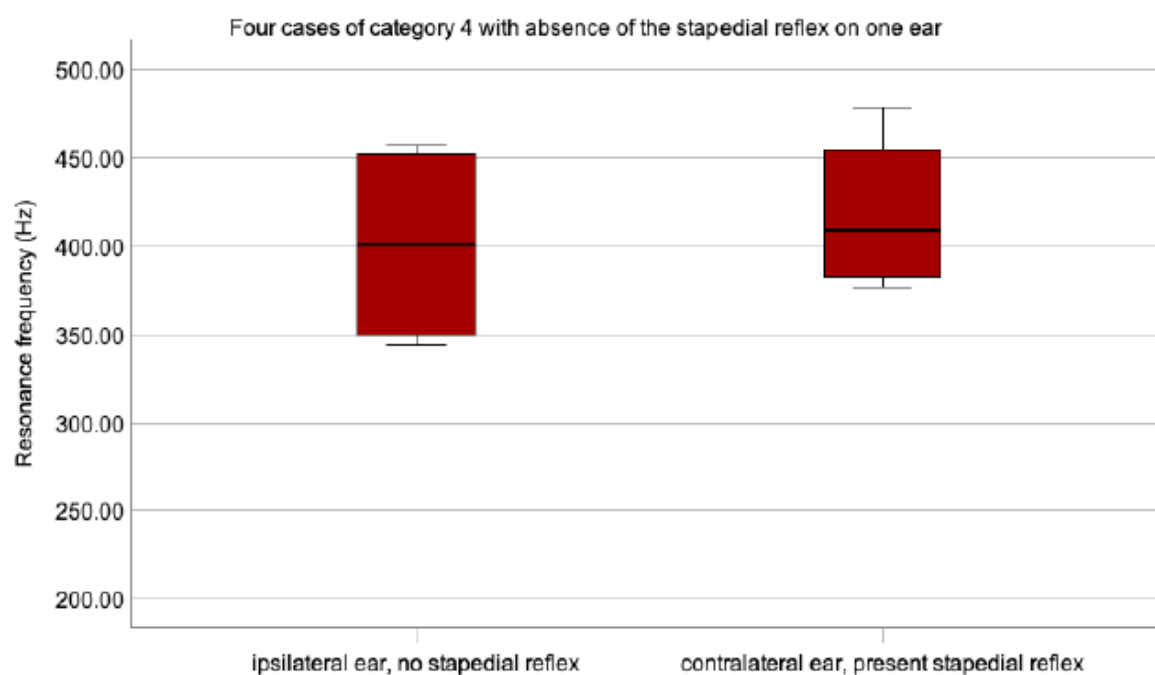
Categories 0–5: 0 – PLAI diagnosis in participants without any ear complaints and with normal hearing (medical history); 1 – plus normal audiogram and otoacoustic emissions; 2 – plus otoscopy (normal outer ear canal and tympanic membrane); 3 – plus normal Valsalva and Toynbee maneuvers and normal pneumatic otoscopy; 4 – plus normal tympanometry (tympanogram type A); 5 – plus present stapedial reflexes.

The study's stated objective was to determine whether diagnostic specificity could be increased through the rigorous selection of healthy subjects.

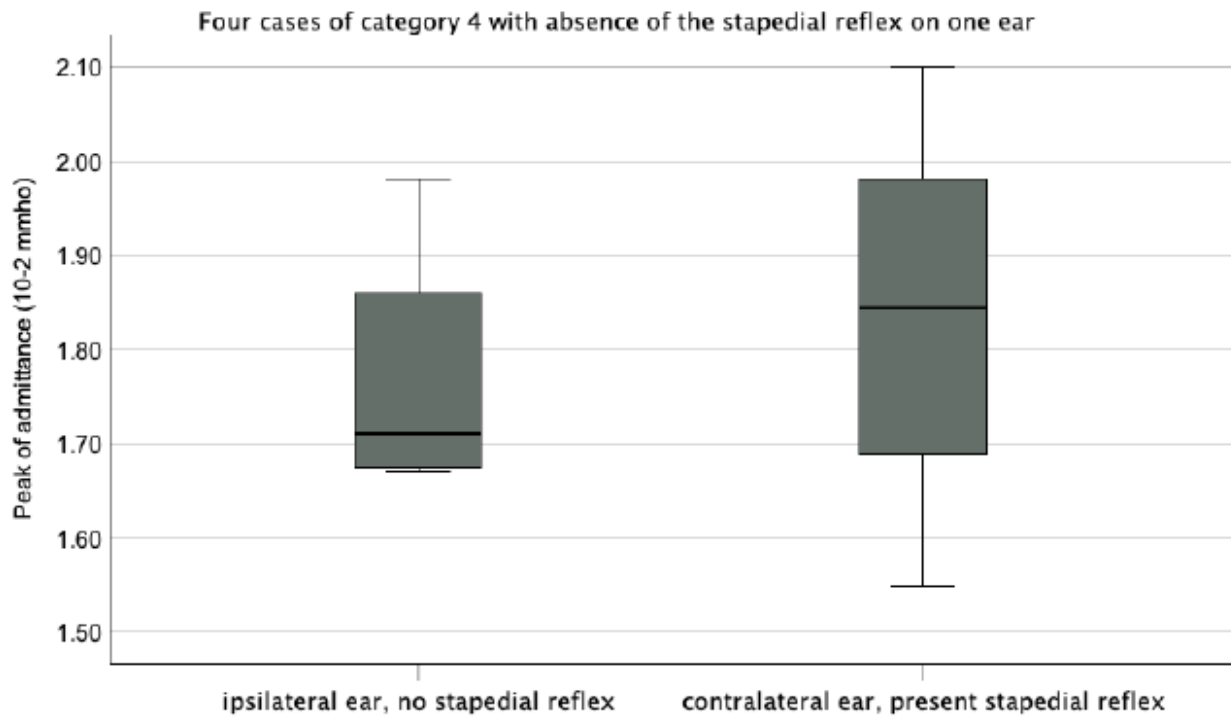
The results presented in Table 4 clearly show that as the number of tests used to classify the "healthy ear" condition increases, the PLAI method's diagnostic specificity also increases. Specificity can be enhanced from 77% to 96% based on the study's questionnaire (Category 0). To achieve this, besides the questionnaire, pure-tone audiometry, otoacoustic emissions, otoscopy, clinical Eustachian tube function tests, tympanometry, and stapedial reflex measurement must also be included.

A side effect of the study became apparent during the recording of stapedial reflex measurement results. In nine ears, no stapedial reflexes could be elicited despite normal pure-

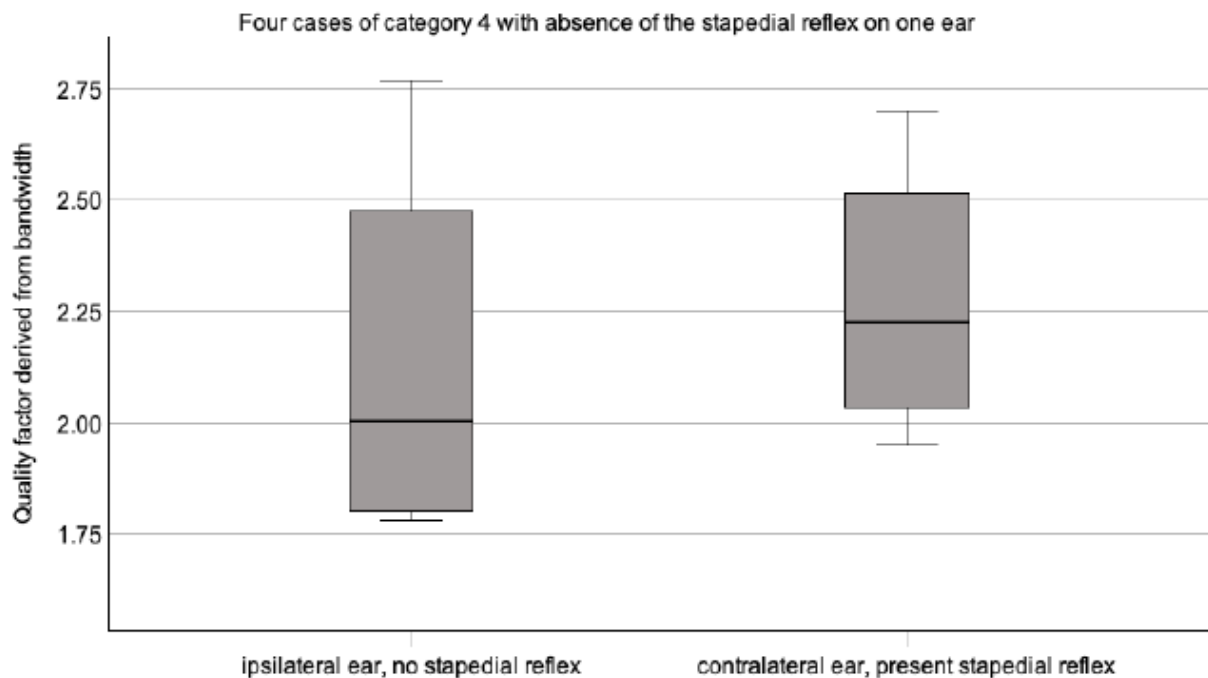
tone audiometry, unremarkable OAE measurements, normal tympanometry, and normal clinical Eustachian tube function tests, along with an intact and normally appearing tympanic membrane. However, the exclusion of these data had no significant effect on the distribution of the PLAI measurement results (Table 2), thus increasing diagnostic specificity by only 2% (Table 4). Five of the nine subjects with a unilateral absence of stapedial reflexes had at least one pathological finding in the opposite ear. Therefore, for the remaining four subjects, aged between 33 and 55 years of Category 4 with a unilateral absence of the stapedial reflexes, the PLAI values of both ears were examined (Figures 4A–E). In all four subjects, the stapedial reflexes of one ear could not be elicited either ipsilaterally nor contralaterally in the frequency range between 0.5 and 4 kHz, whereas the reflexes on the opposite side could be elicited both ipsilaterally and contralaterally.



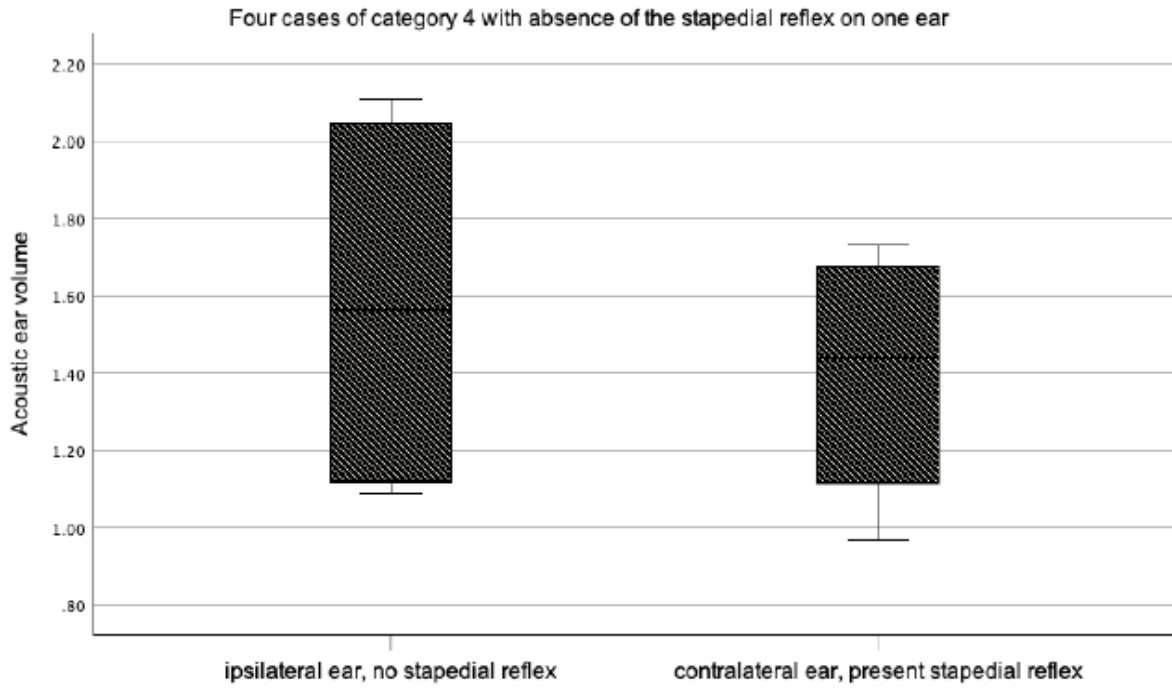
**Figure 4A:** Resonance frequency of admittance value of participants of category 5 with absent stapedial reflexes in one ear and normal stapedial reflexes in the opposite ear.



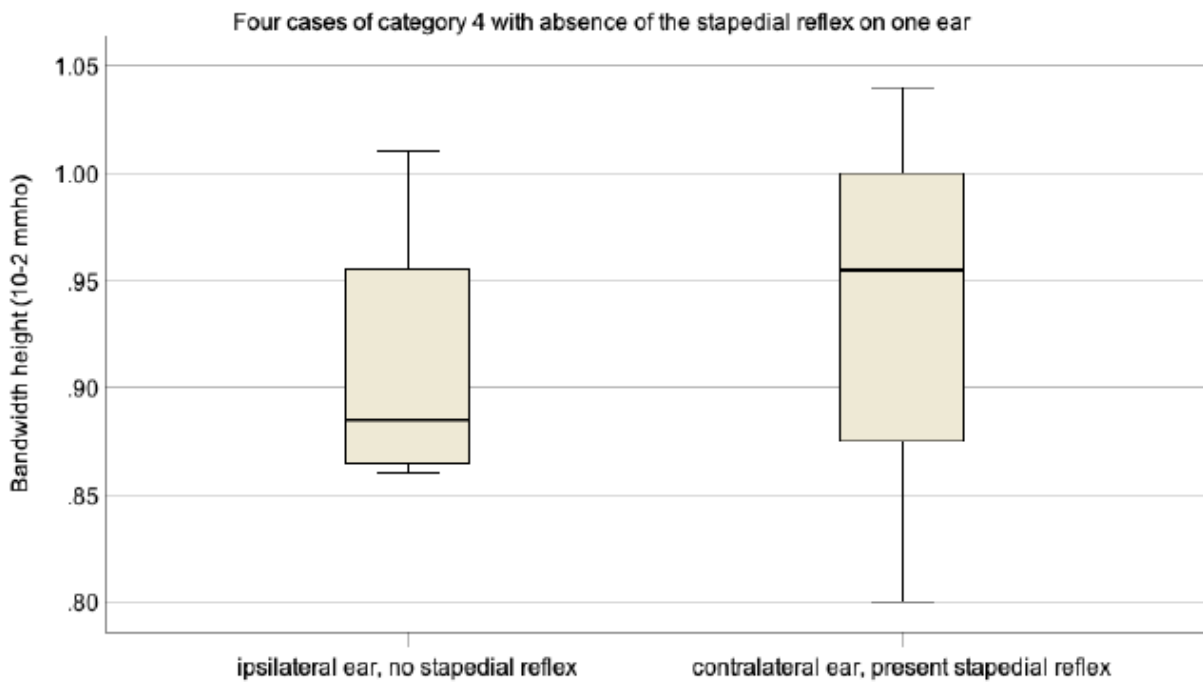
**Figure 4B:** Peak of admittance values of participants of category 5 with absent stapedial reflexes in one ear and normal stapedial reflexes in the opposite ear.



**Figure 4C:** Quality factor from bandwidth values of participants of category 5 with absent stapedial reflexes in one ear and normal stapedial reflexes in the opposite ear.



**Figure 4D:** Acoustic ear volume value of participants of category 5 with absent stapedial reflexes in one ear and normal stapedial reflexes in the opposite ear.



**Figure 4E:** Bandwidth height value of participants of category 5 with absent stapedial reflexes in one ear and normal stapedial reflexes in the opposite ear.

There is a trend toward higher resonance frequencies; higher peak admittance values, bandwidth height, and quality factor derived from bandwidth; and lower acoustic ear volumes in ears with stapedial reflexes compared to the opposite ear without stapedial reflexes (Figures 4A–E).

## Discussion

The present study of healthy subjects shows that at least pure-tone audiometry, OAEs, otoscopy, and the Valsalva maneuver must be performed to rule out obvious pathologies of the tympanic membrane and middle ear, as well as Eustachian tube dysfunction, to reduce the dispersion of PLAI measurements. For the relatively new method of pressure-free admittance measurement, a diagnostic specificity of 90% was achieved. A reduction in the variability of PLAI measurements, particularly in the resonance frequency of admittance, is additionally achieved when 226-Hz tympanometry and stapedial reflex measurement are utilized. By further excluding participants who do not show normal findings with these two measurement methods, the specificity of the PLAI method can be increased to 96%.

In clinical practice, single-frequency tympanometry is generally assumed to indicate that deviations from a type A tympanogram are associated with pathology of the tympanic membrane or the middle ear. For example, the diagnostic sensitivity and specificity for diagnosing OME range from 54% to 97% and 29% to 100%, respectively [20–24]. A type B tympanogram is considered diagnostic of OME, with a mean sensitivity and specificity of 83% and 81.7%, respectively [25]. However, the accuracy of tympanometry is poor [26, 27]. Cinamon and Sade demonstrated in a model with a larger “mastoid” that tympanometry measurements can be unreliable, with errors ranging from 20% to 40%. Even in a model with a small mastoid and slightly negative middle ear pressure, they found an error of more than 400% [26]. They concluded that tympanometry does not accurately reflect middle ear pressure, especially in patients with chronic otitis media, which is characterized by small and often un-aerated mastoids, resulting in small negative pressures. Previous studies have shown that even in individuals with healthy middle ears, tympanometric findings may be observed that are attributed to pathologies of the tympanic membrane or the middle ear [7, 8, 15].

Against this backdrop, the use of new methods in middle ear diagnostics is both sensible and desirable. Before testing new methods for middle ear diagnostics, however, a healthy control group must be defined to establish normal values. It has been shown that even with the PLAI measurement method, the healthy control group exhibits significant variations that cannot be explained and, furthermore, make it difficult to distinguish between healthy individuals and conditions such as otosclerosis and middle ear effusion [13, 16]. By using specific tests during patient selection—such as pure-tone audiometry, otoscopy, the Valsalva and Toynbee maneuvers, tympanometry, and stapedial reflex testing—it is possible to reduce the variability in PLAI measurements and achieve a diagnostic specificity of 95%.

Eustachian tube dysfunction, detected by abnormal Valsalva

and Toynbee tests, explains altered PLAI values, particularly in the resonance frequency of admittance. In contrast, changes in PLAI values due to absent stapedial reflexes require further explanation. The four subjects with a unilateral stapedial reflex that cannot be elicited have normal hearing (including the authors M.A. and T.J.), in whom this phenomenon has been known for years. The afferent pathway of the reflex arc is completely normal in the affected ear. The stapedial reflex cannot be elicited in either the ipsilateral or contralateral ear using various tympanometric techniques. Facial function and the function of the chorda tympani are intact bilaterally in all four subjects. Bilateral normogeusia is present.

Several pathologies of the middle and inner ear, as well as other otoneurological disorders and systemic illnesses, may alter the physiology of the stapedial muscle [28]. Among these, mild middle ear issues—such as slight negative pressure or subtle middle ear abnormalities—and early retrocochlear or neural factors should be mentioned. In the latter case, although less common in the absence of hearing loss, subclinical issues involving the facial nerve or auditory nerve can prevent the reflex arc from functioning properly. Another possible cause of an absent stapedial reflex is reflex decay, in which the reflex is initially present but then rapidly disappears. Errors in the measurement of the stapedial reflex, such as incorrect placement of the test probe or ambient noise, can also result in an inability to register the reflex. Technical or methodological errors can be ruled out in the four ears of the participants involved in this study. Assuming that in four out of 73 ears, where a pathology affecting both ears could be definitively ruled out, the stapedial reflex on one side was absent in approximately 5% of cases despite normal hearing and unremarkable middle ear test results. While the stapedial reflexes are typically present in individuals with normal hearing, they are absent in up to 10% of individuals without any underlying pathology [29]. Pinotti et al. recommend AEP examination in normal-hearing individuals with absent stapedial reflexes to exclude auditory synaptopathy. In contrast, an Iraqi study examined 400 ears from 200 healthy children aged 6 and older and adults aged 19 and older [30]. In this study, ipsilateral and contralateral stapedial reflexes were measured, with no participant reporting absent stapedial reflexes in the frequency range between 0.5 and 4 kHz on either side. Age-related changes in acoustic stapedial reflex (ASR) thresholds are known [31, 32]. While some authors have shown a decrease in ASR thresholds with age [32], others have demonstrated an increase [31], mainly at 4000 Hz.

To determine the effect of the absence of unilateral or bilateral stapedial reflexes on PLAI measurement results, more data must be collected from healthy subjects with normal hearing who lack these reflexes. Additionally, it would be helpful to assess ventilation of the middle ear and mastoid using computed tomography to determine whether temporal bone ventilation affects PLAI measurements.

## Conclusion

This prospective, single-center study of healthy individuals shows that at least pure-tone audiometry, otoscopy, and the Valsalva maneuver must be performed to rule out obvious pathologies of the tympanic membrane and middle ear, as well as Eustachian tube

dysfunction, to reduce the dispersion of PLAI measurements and achieve a diagnostic specificity of 90%.

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## Conflict of Interest

The authors declare that there are no conflicts of interest.

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