

Article

Consequences of Stapes Surgery on Tongue Morphological Characteristics in Narrow Band Imaging, Gustatory Function and General Sensation: A Prospective Tertiary Center Study

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Abstract: (1) Background: Chorda tympani (CT) manipulation during stapes surgery affects its functions. We hypothesized that this alters tongue morphology and sensory functions. (2) Methods: Patients undergoing stapes surgery were tested 1 day preoperatively, 1 and 6 months postoperatively. Narrow band imaging contact endoscopy (NBI) was used to determine the number of fungiform papillae (N_{papillae}) and the total score of blood vessel morphology (NBI_{total}). The taste was tested with taste strips. General sensation was tested with a static two-point discrimination. Tests were performed on ipsilateral and contralateral side of the tongue. (3) Results: 52 otosclerosis patients were included in the study. There was a statistically significant decrease of NBI_{total} ($p = 0.005$), N_{papillae} ($p = 0.009$), sensation of sweet ($p = 0.003$), salty ($p = 0.035$), sour ($p = 0.036$), and bitter taste ($p = 0.013$) within the test side during the follow-up. A statistically significant impact on presence of dysgeusia for sweet was found 1 month postoperatively ($p < 0.005$). Postoperative decrease in two-point discrimination score did not reach a statistical significance ($p = 0.056$). (4) Conclusions: CT manipulation affects fungiform papillae density, vascular patterns and taste sensation. The general sensation of the tongue is not influenced by CT manipulation.

Keywords: hearing loss; facial nerve; taste buds; otologic surgical procedures; microscopy; endoscopes; ageusia; neurophysiology; denervation; atrophy



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1. Introduction

Stapes surgery remains the procedure of choice for hearing rehabilitation in otosclerosis [1], even though this disease was described almost 400 years ago [2] Otosclerosis is a rather common cause of adult conductive hearing loss, with a clinical prevalence estimated at 0.3% to 0.38% [3].

During the stapes surgery, the chorda tympani (CT) nerve is manipulated, leaving the operated patient with potential consequences.

CT is considered as a “taste nerve”; however, other functions have also been discovered. The CT nerve is said to be responsible for non-gustatory information such as texture, temperature [4], and pain [5], for conveying the general sensations from the tongue [6] and for having parasympathetic fibers for submandibular and sublingual glands that stimulate salivary flow [5].

In humans, taste receptor cells are found on the tongue, palate, and other parts of the pharynx and epiglottis [7]. They are innervated by the branches of facial, glossopharyngeal and vagal nerves [8]. Taste receptor cells form taste buds that are organized in macroscopically visible structures named papillae. Papillae are named regarding their shape: foliate

(can be found on the back lateral sides of the tongue), vallate (the largest among papillae, found on the dorsum of the posterior tongue) and mushroom-shaped fungiform papillae (FP) (located at the first 2/3 of the tongue). FP have complex sensory functions, including responses to chemical, thermal and mechanical stimuli [9].

FP of anterior 2/3 of the tongue are innervated by CT, a branch of the facial nerve [7]. However, according to animal studies, they could be dually innervated, including the trigeminal nerve fibers [10]. As described by Purohit et al., 2007 [11], the CT also innervates the contralateral side of the anterior 2/3 of the tongue, explaining the normal taste and FP morphology after ipsilateral CT injury. On the other hand, the number and morphology of papillae are expected to reflect the viability of the appropriate nerve. Sollars et al., 2000 [12] reported that neural damage leads to a decrease in the number of papillae. For that reason, the degeneration process of papillae after severing the CT nerve remains unclear [13].

Taste perception is reported to be influenced by the density and morphology of papillae. Consequently, the visualization of FP is essential [14,15], being the most anatomically accessible. This has been in the focus of scientific interest for many years, yet the techniques have evolved. In vivo techniques allow the visualization of papillae number [12], structure, and vascularization [14]. Some techniques of papillae quantification require the usage of food dye [16]. Very few studies concentrate on postoperative FP morphology [17]. Only one study [18] applies narrow-band imaging (NBI), a noninvasive technique to visualize FP vascular patterns of the anterior 2/3 of the tongue. NBI is a relatively new method of blood vessel visualization that illuminates the structures in narrowed red, green, and blue spectrum bands with interference filters. That increases the contrast between the epithelial surface and the subjacent vascular network [19]. It has been essential in evaluating different organ systems and has an enormous added value in assessing a lesion's malignant potential [20]. To obtain detailed images of epithelium, NBI can be performed with the contact endoscopy technique [21,22].

The alterations in taste are not the only complaints after the stapes surgery. A tingling sensation and tongue numbness are common postoperative findings. The CT nerve is believed to confer the general sensory fibers to the anterior 2/3 of the tongue. A static two-point discrimination test proved to be a sensitive and specific tool for the tongue general sensation assessment [6].

Although there is a lot of evidence of CT function impairment in relation to stapes surgery, many questions remain unanswered. For that reason, this prospective clinical study aims to determine the effect of CT manipulation during stapes surgery on tongue morphological characteristics and sensory functions in 6 months postoperative follow-up. The study hypothesizes that CT manipulation affects tongue FP vascular patterns and density of FP, the taste, and the general sensation of the tongue.

2. Materials and Methods

The Slovenian National Medical Ethics Committee approved this prospective non-randomized clinical study (No. 0120-544/2019/6, date: 6 February 2020). It was registered on ref. [23] (NCT04881513). Written informed consent was obtained from each patient.

2.1. Enrollment

Patients undergoing stapes surgery for otosclerosis were enrolled between February 2020 and June 2021 during their regularly scheduled check-ups at five otologists (SB, IF, KJ, MH, and AM) at the tertiary otorhinolaryngology referral center.

2.1.1. Inclusion Criteria

- Age > 18 years;
- Patients with clinical otosclerosis diagnosed by the ENT specialist-otosurgeon.

2.1.2. Exclusion Criteria

- A recent modification of regular therapy [24];

- Recent onset of smoking [25];
- COVID-19 infection [26];
- History of taste disturbance at the enrolment;
- History of smell disturbance at the enrolment;
- History of facial palsy at the enrolment;
- Diseases of the oral cavity.

Patients were assessed by two-point discrimination, tongue morphology, and taste 1 day prior to surgery, 1 month (first check-up) and 6 months postoperatively (second check-up). Two-point discrimination was tested 6 h postoperatively, as well. The ipsilateral (i.e., ipsilateral to stapes surgery or test) and contralateral (i.e., contralateral to stapes surgery or control) sides of the tongue were tested. The stapes surgery was conducted under local anesthesia with an endaural approach.

2.2. Tongue Morphology Assessment with Narrow Band Imaging

The patient was instructed to keep the tongue out and still as much as possible. The FP blood vessels were visualized on the test and control side of the middle lateral area of the anterior 2/3 of the tongue using NBI contact endoscopy technique with a 0° otological endoscope (Olympus-Gyrus ACMI, Hamburg, Germany) and compatible equipment (Olympus Visera Pro PRO CLV S40 light source, and Olympus Visera Pro OTV-S7 PRO HD endoscopic camera system, Olympus, Tokyo, Japan). The videos of contact endoscopy were recorded using high-definition technique in an upscaled resolution of 1920 × 1080 pixels at 50 frames per second (Sony, Tokyo, Japan) symmetrically on the test and control side, avoiding the tongue tip. Endoscopy was performed by two independent observers (NBU and MH) with the standardized protocol.

Then, two independent observers (NBU and RŠ) performed analyses of recorded videos to determine blood vessel morphology and the number of FP. In unclear cases, the consensus was reached with a third researcher (SB). The morphological blood vessel evaluation was performed on both sides of the tongue from the recorded videos with the five-point scoring system (Table 1) [13,17].

Table 1. NBI Morphological Blood Vessel Evaluation Grading System.

Grade	Blood Vessel Morphology
Type A (4 points)	Clear loop and wooden branch shape
Type B (3 points)	Unclear loop and wooden branch shape
Type C (2 points)	Elongated blood vessels with wen (swelling)
Type D (1 point)	Granulated shape, dotted shape
Type E (0 points)	Unclear, disappeared

NBI—narrow band imaging. Adapted by Negoro et al., 2004 and Shimada et al., 2013 [13,17].

The number of fungiform papillae (N_{papillae}) was counted in the areas with the richest blood vessel morphology under the still picture from the recorded video (Figure 1). The total score of blood vessel morphology ($\text{NBI}_{\text{total}}$) was obtained by summing the points of papillae grades (Table 1) in the 20 mm² areas (using the special plasticized paper with the 20 mm² holes cut out (Figure 1A) of the richest vascular morphology graded. The mean score of blood vessel morphology (NBI_{mean}) was calculated by dividing the $\text{NBI}_{\text{total}}$ by the N_{papillae} in the 20 mm² areas. All the evaluations were done using the contact endoscopy technique exclusively for all the measurements and regarded the unoperated side as the control side.

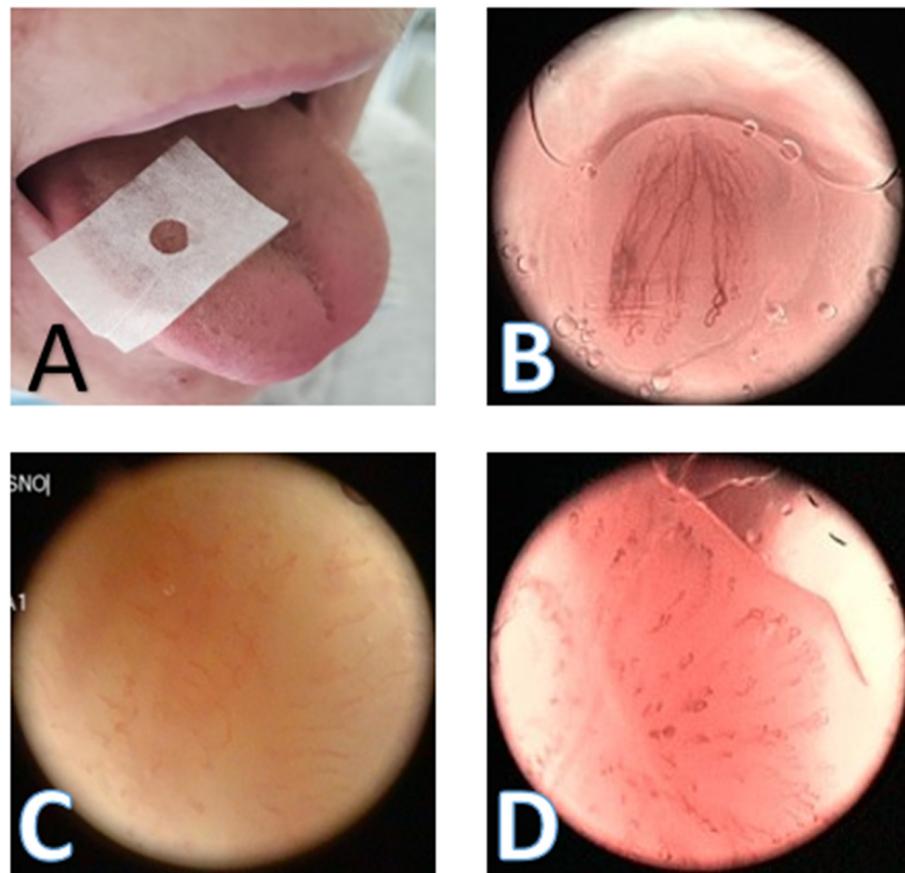


Figure 1. NBI contact endoscopy. (A) The tongue with the special plasticized paper with the 20 mm² holes positioned in the anterior 2/3 of the lateral tongue; (B) the NBI contact endoscopy picture of FP type A; (C) the contact endoscopy picture of FP type D without the use of the NBI; and (D) the contact endoscopy picture of FP type D with the use of the NBI; NBI—Narrow band imaging; FP—Fungiform papillae.

2.3. The Taste Assessment

The sense of taste was tested using taste strips (Burghart Messtechnik GmbH, Tinsdaler Weg 175, Wedel, Germany). The participants were instructed not to eat, chew bubble gum, or smoke, and to drink only water 1 h before testing. Each side of the tongue was tested separately. During the testing, the tongue was out. To avoid the activation of other gustatory nerves, patients were instructed to define only tastes they sensed with their tongues before swallowing. Taste strips were placed on one side of the tongue and moved gently anteroposteriorly (regarding the size of the tongue) for up to 5 s, as needed. The sweet, sour, salty, and bitter tastes were tested in four concentrations, beginning with lower concentrations in a randomized manner following the producer's instruction manual. Each correct answer yielded 1 point, with a maximum possible sum score of 4 points per taste tested. Tasteless strips were offered as well. After each strip, the patient rinsed the mouth with water. The anterior 1/3 of the tongue was avoided because of the possibility of dual innervation [10,27].

The dysgeusia was assessed with the same taste strips. The dysgeusia for sweet was present if a patient tasted sweet with any other taste strip (i.e., bitter, salty, or sour). Patients with no sensation of sweet taste, whether true or false (i.e., dysgeusia), were excluded from the analysis. The same method was applied to analyze bitter, salty, and sour dysgeusia.

2.4. Two-Point Discrimination

Patients were assessed for static two-point discrimination with the tongue out with a Discrim-a-gon 2-point Discriminator 12-1492 (D1) (3 B Scientific, Libertyville, IL, USA). The

two-disc set was used with the distances between the rods from 1 to 15 mm. The patient's tongue was touched for 3 s with the testing rods at 90 degrees to the tongue surface in the anterior 2/3 of the tongue in the lateral middle part symmetrically on both sides, avoiding the tongue tip and beginning with the rods that are more apart and proceeding to closer positioned rods (from 15 mm to 1 mm), taking care not to touch the tongue with three rods at the same time. Patients were asked to indicate whether they felt the touch of one or two rods by showing the number 1 or 2 with their fingers. When they showed one finger, we applied the one-rod-size up and down to confirm the result one more time. The smallest distance between the two rods that the patient sensed was taken as the definite result of tongue two-point discrimination testing. The procedure was then repeated on the other half of the tongue. The cooperation was checked from time to time by applying one rod to the tongue.

2.5. Surgical Procedure

After preparing the operative field, the mixture of local anesthetic (Bupivakain Grindeks 5 mg/mL sol., AS Grindeks, Krustpils iela 53, Riga, LV-1057, Latvia) and 0.4 mL of adrenaline (Suprarenin 1 mg/mL sol, Cheplapharm, Arzneimittel GmbH, Ziegelhof 24, Greifswald, Germany) was injected into the EAC meatus just lateral to the osseocartilaginous junction. The surgery was done using the operating microscope with the endaural approach. The tympanomeatal flap was elevated, and the bone covering the chorda tympani nerve was removed with the curette. With the use of the laser, the anterior and posterior crus of the stapes and the stapedius tendon were divided, and the footplate was perforated. Sometimes the footplate was perforated manually, using the perforator. The oval window was sealed with an autologous temporal muscle fascia or tragal perichondrium and a Schuknecht-type Teflon piston prosthesis (RN:2024-07-19, Medtronic Xomed, Jacksonville, FL, USA) was used. At the end of the surgical procedure, the surgeon graded the intraoperative CT manipulation from 1 to 5, as adapted from other authors [6]: (1) nonmanipulated, (2) minimal, (3) intermediate, (4) significant manipulation, and (5) transected.

2.6. Statistical Analysis

Statistical analysis was performed using Microsoft Excel for Windows and for Mac (Microsoft Corp., Redmond, WA, USA) and IBM SPSS (version 23, IBM Corp., Armonk, NY, USA). $p < 0.05$ was considered as statistically significant.

3. Results

In total, 52 patients (35% male, 65% female) aged 47 ± 9.4 years (48.2 ± 11.5 male, 45.7 ± 9.3 female) were enrolled in the study. Four patients skipped the first check-up ($Mdn = 33$ days) and four the second one ($Mdn = 181$ days).

Of them, 11 patients (21%: 5 female, 6 male) received regular therapy. One female patient received oral contraceptive, two levothyroxine sodium, and one bisoprolol and one patient received rosuvastatin, ezetimibe, bisoprolol, and acetylsalicylic acid. Two male patients received enalapril, two adalimumab, one secukinumab, and one patient received bupropion and pregabalin. In none of these patients did the treatment regimen change.

3.1. Tongue Morphology Assessment with NBI

Results of statistical analysis of NBI_{total} , $N_{papillae}$, and NBI_{mean} are summarized in Tables 2–4, Figures 2 and 3.

3.1.1. The Total Score of Blood Vessel Morphology

Summarized data are presented in Table 2 and Figure 2. Results of detailed between-group and within-group statistical analysis are provided in Supplementary File S1.

Due to outliers in the preoperative and second check-up group, the Friedman test was run. There was a statistically significant difference in NBI_{total} between the preop-

erative and second check-up ($p = 0.007$). No statistically significant difference was observed between preoperative and first check-ups and between first and second check-ups ($p > 0.05$). p — p -value.

3.1.2. The Number of Fungiform Papillae

Summarized data are presented in Table 3 and Figure 3. Results of detailed between-group and within-group statistical analysis are provided in Supplementary File S2.

Due to outliers in the first and second check-up group, the Friedman test was run. There was a statistically significant difference in the N_{papillae} between the preoperative and second check-up ($p = 0.017$). No statistically significant difference was observed between preoperative and first check-ups and between first and second check-ups ($p > 0.05$). p — p -value.

3.1.3. The Mean Score of Blood Vessel Morphology

Summarized data are presented in Table 4. Results of detailed between-group and within-group statistical analysis are provided in Supplementary File S3.

Table 2. The NBI_{total} .

Group	Check-Up ^a			p
	0	1	2	
Test	<i>Mdn</i> = 12 12.28 ± 4.577	<i>Mdn</i> = 12 10.97 ± 4.215	<i>Mdn</i> = 9	0.005 ^b
Control	<i>Mdn</i> = 12 11.85 ± 4.171	<i>Mdn</i> = 12 11.79 ± 4.824	<i>Mdn</i> = 10	0.801 ^b
p	0.661 ^c	0.426 ^c	0.147 ^b	

^a—data are presented as medians and/or mean ± SD; ^b—Friedman test; ^c—independent samples t -test; 0—preoperative check-up; 1—first postoperative check-up (1 month postoperatively); 2—second postoperative check-up (6 months postoperatively). p — p -value. A total of 39 patients were included in the within-group and 39 in the between-group analysis of NBI_{total} . There was a statistically significant decrease of the total score of blood vessel morphology (NBI_{total}) within the test side during the follow-up. Cell shaded grey shows a statistically significant difference.

Table 3. The N_{papillae} .

Group	Check-Up ^a			p ^b
	0	1	2	
Test	5	4	4	0.009
Control	4	5	4	0.564
p ^b	0.315	0.980	0.154	

^a—data are presented as medians; ^b—Friedman test; 0—preoperative check-up; 1—first postoperative check-up (1 month postoperatively); 2—second postoperative check-up (6 months postoperatively); p — p -value. A total of 39 patients were included in the within-group and between-group analysis of the number of fungiform papillae (N_{papillae}). There was a statistically significant decrease of N_{papillae} within the test side during the follow-up. Cell shaded gray shows a statistically significant difference.

Table 4. The NBI_{mean} .

Group	Check-Up ^a			p ^b
	0	1	2	
Test	2.8	2.5	2	0.515
Control	3.0	3.0	3	0.915
p ^c	0.462	0.396	0.434	

^a—data are presented as medians; ^b—Friedman test; ^c—Mann Whitney U test; 0—preoperative check-up; 1—first postoperative check-up (1 month postoperatively); 2—second postoperative check-up (6 months postoperatively); p — p -value. A total of 39 patients were included in the within-group and between-group analysis of mean score of blood vessel morphology (NBI_{mean}).

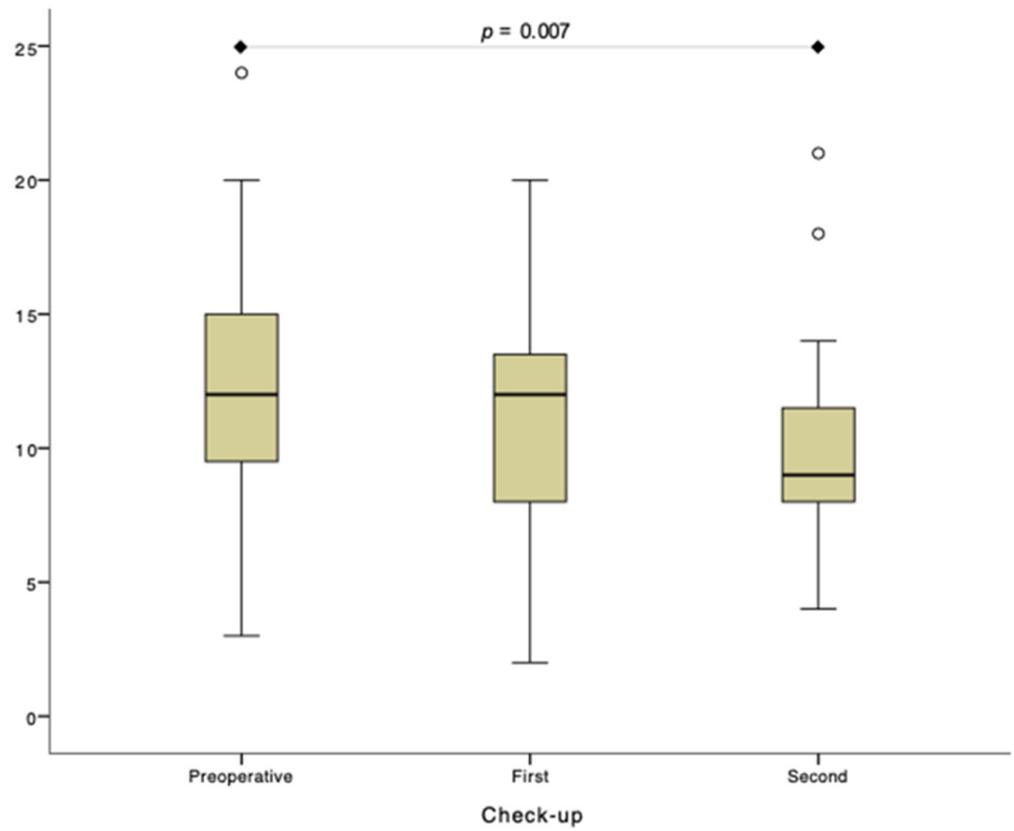


Figure 2. The total score of blood vessel morphology analysis (NBI_{total}) within the test side. First check-up was performed 1 month and second check-up 6 months postoperatively.

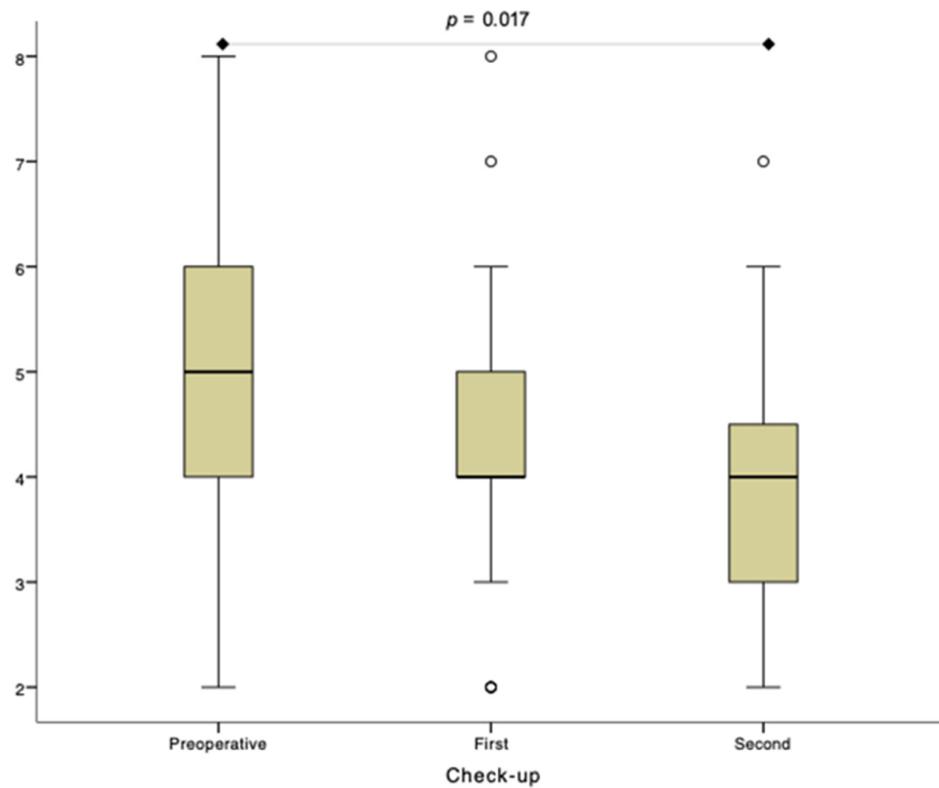


Figure 3. The number of fungiform papillae (N_{papillae}) analysis within the test side. First check-up was performed 1 month and second check-up 6 months postoperatively.

3.2. The Taste Assessment

Summarized data are presented in Table 5. Results of detailed between-group and within-group statistical analysis are provided in Supplementary File S4.

Table 5. Taste Strips Sum Score.

Taste	Group	Check-Up ^a			<i>p</i> ^b
		0	1	2	
Sweet	Test	4	2	3	0.003
	Control	4	3	4	0.878
	<i>p</i> ^c	0.941	0.010	0.044	
Salty	Test	2	1	1	0.035
	Control	2	3	2	0.159
	<i>p</i> ^c	0.853	0.077	0.045	
Sour	Test	3	1.5	2	0.036
	Control	2.5	3	3	0.786
	<i>p</i> ^c	0.900	0.023	0.034	
Bitter	Test	3	1.5	2	0.013
	Control	3	3	3	0.281
	<i>p</i> ^c	0.447	0.007	0.260	

^a—data are presented as median values; ^b—Friedman test; ^c—Mann Whitney U test; 0—preoperative check-up; 1—first postoperative check-up (1 month postoperatively); 2—second postoperative check-up (6 months postoperatively); *p*—*p*-value. A total of 32 patients were included in the within-group and between-group taste strip analysis. Cells shaded gray show statistically significant differences.

Dysgeusia Analysis

Dysgeusia analyses for each taste, side, and check-up are depicted in Tables 6 and 7.

Table 6. The Test Side.

Sweet	1		2		<i>p</i>	
	True	False	True	False	0 vs. 1	0 vs. 2
0	21 (100%)	0	16 (67%)	8 (33%)	<0.005	0.130
True						
False	2 (20%)	8 (80%)	1 (20%)	4 (80%)		
Salty	1		2		<i>p</i>	
	True	False	True	False	0 vs. 1	0 vs. 2
0	1 (9%)	10 (91%)	4 (50%)	4 (50%)	0.063	1.000
True						
False	6 (55%)	5 (45%)	5 (56%)	4 (44%)		
Sour	1		2		<i>p</i>	
	True	False	True	False	0 vs. 1	0 vs. 2
0	0	5 (100%)	4 (57%)	3 (43%)	0.553	0.375
True						
False	6 (25%)	18 (75%)	7 (32%)	15 (68%)		
Bitter	1		2		<i>p</i>	
	True	False	True	False	0 vs. 1	0 vs. 2
0	9 (56%)	7 (44%)	8 (50%)	8 (50%)	0.159	0.249
True						
False	4 (29%)	10 (71%)	3	10		

Data are presented as numbers and proportions in parentheses. 0—preoperative check-up; 1—first postoperative check-up (1 month postoperatively); 2—second postoperative check-up (6 months postoperatively); True—true sensation of a taste; False—dysgeusia of a taste; *p*—*p*-value. Cell shaded gray shows statistically significant differences.

Table 7. The Control Side.

Sweet		1		2		p	
0	True	False	True	False	0 vs. 1	0 vs. 2	
True	18 (78%)	5 (22%)	20 (87%)	3 (13%)	0.226	0.314	
False	5 (56%)	4 (44%)	6 (67%)	3 (33%)			
Salty		1		2		p	
0	True	False	True	False	0 vs. 1	0 vs. 2	
True	3 (25%)	9 (75%)	10 (83%)	2 (17%)	1.000	1.000	
False	2 (33%)	4 (67%)	6 (75%)	2 (25%)			
Sour		1		2		p	
0	True	False	True	False	0 vs. 1	0 vs. 2	
True	6 (40%)	9 (60%)	5 (31%)	11 (69%)	0.252	0.685	
False	3 (19%)	13 (81%)	3 (91%)	13 (81%)			
Bitter		1		2		p	
0	True	False	True	False	0 vs. 1	0 vs. 2	
True	13 (68%)	6 (32%)	10 (53%)	9 (47%)	0.173	1.000	
False	6 (43%)	8 (57%)	7 (54%)	6 (46%)			

Data are presented as numbers and proportions in parentheses. 0—preoperative check-up; 1—first postoperative check-up (1 month postoperatively); 2—second postoperative check-up (6 months postoperatively); True—true sensation of a taste; False—dysgeusia of a taste. *p*—*p*-value.

3.3. Two-Point Discrimination

Summarized data are presented in Table 8. Results of detailed between-group and within-group statistical analysis are provided in Supplementary File S5.

Table 8. Two-Point Discrimination.

Group	Check-Up ^a				p ^b
	0	6 h	1	2	
Test	6	6	6	7	0.056
Control	6	6	6	6	0.801
<i>p</i> ^c	0.688	0.824	0.992	0.635	

^a—data are presented as median values in millimetres; ^b—Friedman test; ^c—Mann Whitney U test; 0—preoperative check-up; 6 h—check-up 6 h postoperatively; 1—first postoperative check-up (1 month postoperatively); 2—second postoperative check-up (6 months postoperatively); *p*—*p*-value. A total of 40 patients were included in the within-group and between-group analysis of two-point discrimination measurement.

3.4. CT Manipulation

The data about the CT manipulation during stapes surgery are presented in Table 9. The statistical analyses were performed to test the assumptions prior to the planned correlation tests. Multiple scatter graphs were created to plot the grade of CT manipulation as an ordinal independent variable and one continuous dependent variable (difference between two check-ups in NBI_{total}, N_{papillae}, NBI_{mean}, taste strips sum score for each taste, and two-point discrimination). Visual inspection of these scatter graphs did not show any monotonic or non-monotonic relationship of the data. For that reason, statistical correlation tests were not performed. The detailed data are available in Supplementary File S6.

Table 9. The Grade of Chorda Tympani Manipulation.

Grade of CT Manipulation	No (%)
minimal	23 (44)
intermediate	17 (33)
significant	10 (19)
transected	1 (2)
no data	1 (2)

CT—chorda tympani. Data are presented as number of patients (Nr) and proportions (%) within group of 52 analyzed patients.

4. Discussion

This prospective tertiary center clinical study evaluated the influence of stapes surgery in a group of adult otosclerosis patients on FP density and blood vessel morphology with NBI contact endoscopy, taste function with taste strips, and tongue general sensation with static two-point discrimination in 6 months follow-up. Stapes surgery was proved to cause the decrease in the N_{papillae} , the NBI_{total} and sensation of the sweet, sour, salty, and bitter taste. Increased likelihood of dysgeusia for sweet taste was detected. Stapes surgery did not influence the static two-point discrimination scores. Therefore, the hypothesis that CT manipulation affects tongue FP vascular patterns and density of FP and the taste was confirmed. We rejected the hypothesis that the general sensation of the tongue was influenced by CT manipulation during stapes surgery.

4.1. NBI

Studies confirm the change of FP blood vessel morphology after the middle ear surgery [13]. Our research has proved the usefulness of contact endoscopy in tongue papillae assessment in combination with the NBI technology. To our knowledge, this is the first prospective NBI contact endoscopy study to compare the FP blood vessel morphology before and after surgery to be applied to both sides of the tongue of the same patient. Unlike some other NBI studies [18], we have used contact endoscopy exclusively for all the measurements. We have found that observational distances of 2, 5, 10, and 20 mm used by Shimada et al. [18] are difficult to obtain. Furthermore, they have a negative impact on the accuracy of the measurements. Contact endoscopy provides more detailed views and annulates the influence of tongue movements on the picture quality. We have also found the 0° otoendoscope to be the most suitable instrument for the FP blood vessel assessment as it is small and easy to handle. Instead of taking pictures, we have filmed a larger portion of the tongue and analyzed the areas with the richest vasculature.

Our study has shown that stapes surgery negatively impacts the number of FP and the blood vessel morphology, which is consistent with Saito et al. (2001, 2014). Regarding some studies, taste buds disappear 50 days after severing the CT nerve [11]. Whitehead and colleagues showed that the hamster's taste buds persisted 330 days after transection of the CT [28]. In contrast, in our group of patients, the number of FP decreased statistically significantly on the operated side during follow-up of 6 months (Figure 3 and Table 3). That could be explained by the human taste buds being more sensitive to CT manipulation than those of hamsters and the turnover of hamster taste buds being much slower. As there was no change in the FP number during the 1-month postoperative follow up, we can conclude that human FP persisted for at least 30 days after the CT nerve manipulation. That could indicate the human FP turnover time is between 1 and 6 months, which is consistent with the study by Perea-Martinez et al. [29] that estimates the half-life of cells in mammalian taste buds to be 8–22 days depending on the cell type. There was no decrease in NBI_{mean} , which could be explained by the reduction in the number of the papillae and the total score of blood vessel morphology. As the number of papillae has not changed on the other

non-operated, our study does not confirm the contralateral CT influences on the anterior tongue as reported by others [10].

4.2. Taste

The taste strips are an excellent clinical tool for taste assessment [30]. Taste dysfunction after middle ear surgery is said to be dependent on the degree of CT manipulation [30]. Ciofalo et al. (2015) presented the results of 30 otosclerosis patients whose taste assessment on both sides of the tongue revealed no changes in the taste scores on the contralateral side of the tongue and the declining taste scores on the operated side in the early postoperative period. They have found the improvement of taste scores 6 to 12 months after surgery and, unfortunately, did not collect the data about postoperative dysgeusia [31]. Mueller et al. found a significant change in the taste strip scores on the operated side in patients with the considerable manipulation of the CT nerve, with no significant differences in patients with minor manipulation or on the contralateral side of the tongue [30]. These results are consistent with ours since we have found statistically significant differences in the test strips scores for all tastes after the stapes surgery.

On the other hand, the taste scores on the control side remained unchanged throughout the follow-up period. In our study, the only patient with the sectioned nerve had lost all the taste sensitivity on the sectioned nerve side. Some authors found the possible preservation of taste during the whole-mouth testing, regardless of the CT manipulation, which is explained by releasing CT's inhibitory effect on the glossopharyngeal nerve [32,33]. Regardless of the sectioning of the CT nerve, Celis-Blaubach et al., 1970 [34] found the preserved ability to taste the acid in 43.5% of cases, which was explained by the possibility of acid fibers following a different pathway. To avoid the influence of other nerves, we have used the separate taste testing of both halves of the tongue, avoiding the tongue tip region, and instructed the patients to report the tastes felt with the tongue before swallowing the saliva.

Some authors speculate that a part of the taste afferent fibers innervating the anterior part of the tongue may bypass the tympanic cavity [35] and lead to the preservation of taste function [31] after the middle ear surgery. Our study indicates that there are probably no afferent CT taste fibers that would be bypassing the tympanic cavity or innervating the contralateral side of the tongue as well.

There are only a handful of studies exploring the characteristics of post-stapedectomy dysgeusia. Regarding a review of the stapes surgery complications by Bartel et al., 2021 [36] the rate of postoperative dysgeusia is said to be 10%, with no detailed description of the dysgeusia type. Molinari et al. [37] found only 3/48 (6.25%) of patients having a subjective (no objective taste testing performed) persistent dysgeusia 6 months after endoscopic stapes surgery, regardless of the preservation of the CT nerve. In microscopic surgical approaches, the percentage of dysgeusia is said to be even higher, up to 44.1% [38]. Regarding the postsurgical dysgeusia type, the most pronounced early postoperative changes were documented in a sweet and bitter taste, with the late recovery primarily of bitter taste [39]. Other studies found the most alterations in the sweet and salty taste qualities [40]. Guder et al. [41] found taste impairment on the ipsilateral side of the tongue after 3 days and the improvement within 3 months of surgery, not explicitly concentrating on individual tastes. We have found that stapes surgery has a statistically significant impact on the presence of dysgeusia for sweet taste up to 1 month postoperatively.

4.3. General Sensation

Somatosensory receptors of the tongue have small receptive fields, which is why they have a superior two-point discrimination ability compared to other regions of the body [42]. The tongue tip is one of the most sensitive tactile sites of the body [9]. Previous studies advocated that there might be a relation between FP density and tactile perception [43]. The studies addressing the tongue general sensation after the stapes surgery are scarce. We have found the decrease in the static two-point discrimination testing after otosclerosis

surgery that was statistically insignificant ($p = 0.056$), as Perez et al., 2006 [6] showed when considering the late-stage follow-up period. Inconsistently, in our patient with the transected nerve, the sensory function regarding the static two-point discrimination test remained unchanged in the 6-month postoperative period. It is interesting that the same findings considering the patient with the transected nerve were found by Perez et al., 2006 [6].

One possible explanation could be that the CT nerve carries only a small number of sensory fibers to the tongue. That's why some patients complain of tongue numbness or tingling sensations for some weeks after surgery, and then eventually those sensations disappear. Perez et al. (2006) also found the static two-point discrimination recovered during the follow-up period [6]. The main sensory nerve of the anterior 2/3 of the tongue is the trigeminal nerve. There are said to be CT and trigeminal nerve interactions that can occur at the periphery or at the central level [44]. Results of our study do not support the theories of CT having the fibers for the tongue general sensation. Due to these inconsistencies and the small body of literature that includes animal studies [12,28], retrospective studies [45], small groups [13,41], and unilateral tongue analysis [18], there is an urge for multicentric prospective randomized controlled clinical trials.

4.4. CT Manipulation

The data about the influence of the degree of CT manipulation on morphological and functional changes of the tongue FP are scarce. Mueller et al., 2008 [29] have found more prominent taste change in the group of patients with significant CT manipulation during surgery, while Guder et al., 2012 [41] found the changes in taste even after minor CT manipulation. We did not find any relationship between the degree of CT manipulation during surgery and the severity of taste disturbance after stapes surgery, the change in number and morphology of FP or the change in two-point discrimination score. Our data were too scattered. The reason could lie in the subjectiveness of the grading system.

4.5. Perspective of CT Preservation

The preservation of the CT nerve could lower the incidence of the morphology and FP function change after stapes surgery. The endoscopic approach is said to enable the surgeon to reach better views of the operation field and avoid the manipulation of the CT nerve [38,46]. Injury to the nerve was said to be more than three times more likely to happen with the microscopic approach [47]. In our department, all surgeons use the microscopic approach in stapes surgery and should be stimulated to start applying other CT preservation techniques. On the other hand, Szmuda et al., 2020 [48] explored the new techniques of intraoperative nerve preservation. They have tested the preoperative visualization of the facial nerve by magnetic resonance-based diffusion tensor imaging-fiber tracking with neuronavigation system integration in patients with cerebellopontine angle tumors. Similar methods could be used in planning the stapes surgery in order to preserve the facial nerve and its branches, including CT.

4.6. Advantages and Limitations

Additional advantages of our study are its prospective design and relatively long and regular follow-up period, despite the epidemiological restrictions. Another advantage is taking the contralateral side of the tongue as a control, which annulates the effect of less-known between-subject differences that could influence the taste, tongue general sensation, and FP number and morphology [47]. Moreover, there were no within-subject (i.e., between sides) differences of these measurements at the enrolment.

The disadvantages are a relatively low number of patients included in the final analysis, which is also a consequence of the COVID-19 pandemic, which resulted in patients being rescheduled or lost to follow-up because of COVID-19.

5. Conclusions

That prospective, 6-month follow-up clinical study confirms the negative impact of stapes surgery on sweet, salty, sour, and bitter taste and tongue morphology with taste strips and narrow-band imaging contact endoscopy (NBI). This trial provides the sound basis for the routine clinical use of NBI in papillae evaluation in other medical conditions. It offers physiological insights into chorda tympani functions.

Supplementary Materials: The following supporting information can be downloaded at <https://www.mdpi.com/article/10.3390/app12073248/s1>, Supplementary File S1: Results of detailed between-group and within-group statistical analysis of the total score of blood vessel morphology. Supplementary File S2: Results of detailed between-group and within-group statistical analysis of the number of fungiform papillae. Supplementary File S3: Results of detailed between-group and within-group statistical analysis of the mean score of blood vessel morphology. Supplementary File S4: Results of detailed between-group and within-group statistical analysis of the taste assessment. Supplementary File S5: Results of detailed between-group and within-group statistical analysis of the two-point discrimination. Supplementary File S6: Chorda tympani manipulation analyses.

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Informed Consent Statement: Informed consent was obtained from all subjects involved in the study.

Data Availability Statement: Data supporting reported results can be found at the Department of Otorhinolaryngology and Cervicofacial Surgery, University Medical Centre Ljubljana, Zaloska 2, 1000 Ljubljana, Slovenia.

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