# A new extension to the 'Taste Strips' test\*

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## Abstract

**Objective:** Assessment of gustatory function in human subjects using the 'taste strips' test is an easy and validated procedure. The aim of this study was to extend this test in order to detect subjects with superior gustatory sensitivity.

**Methods:** The investigation included 134 subjects (29.5±12.6 years, range 18-84 years) with normal gustatory function. Four concentrations of sweet, sour, salty, and bitter were augmented with additional low concentrations (sweet: 25/12.5mg/ml sucrose; sour: 27/15mg/ml citric acid; salty: 6.4/2.6mg/ml sodium chloride, bitter: 0.15/0.06mg/ml quinine hydrochloride), resulting in a maximum extended taste score (ETS) of 24.

**Results:** The mean ETS was  $14.5 \pm 3.2$ . Specifically, it was  $4.5 \pm 1.2$  for sweet,  $2.8 \pm 1.0$  for sour,  $4.0 \pm 1.3$  for salty, and  $3.2 \pm 1.2$  for bitter. In contrast to the original version of the taste strips test, no ceiling effect was observed. Cluster analysis separated three groups of subjects by ETS, whereas test scores derived from the original four concentrations were insufficient to discriminate the subgroup with higher gustatory sensitivity.

**Conclusions:** The extended taste strips test seems to be a useful tool for the detection of patients with low gustatory thresholds for sweet, sour, salty, or bitter taste.

Key words: taste, human, taste buds, normative data, supertaster

## Introduction

In most patients, self-reported chemosensory function does not correlate reliably with psychophysical test results <sup>(1-3)</sup>. Moreover, many patients confuse taste and flavour perception, making it necessary to perform both smell and taste examinations <sup>(1-4)</sup>. Although a significant number of smell tests have been described, only a few valid taste tests have been published <sup>(5)</sup>. Quantitative assessment of gustatory function with taste strips is an established method for the assessment of taste function in routine clinical assessment and research <sup>(6)</sup>. This test was partly initiated by a working group on olfaction and taste at the German Society of Otolaryngology, Head and Neck Surgery, and has been used in a number of studies including umami taste <sup>(7-11)</sup>. About half of healthy subjects could correctly identify the lowest concentrations of sweet, sour, salty, and bitter. This ceiling effect (i.e., the limitation of measurement range in the upper range of gustatory sensitivity), seen in subjects with normal gustatory function, might not be of clinical significance. However, detecting a partial reduction in gustatory function in patients with superior taste performance might be of interest in certain cases. Changes in the upper range of gustatory function cannot be measured with the current test. Consequently, the aim of this study was to extend the validated taste strip test for the characterization of individuals with low gustatory thresholds for sweet, sour, salty or bitter taste. In order to assess a possible anatomical correlation for taste sensitivity, the number of fungiform papillae on the tongue was recorded for each participant by photo-documentation.

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## **Materials and methods**

The study was carried out at the Department of Otorhinolaryngology at the Medical University of Vienna and was approved by the local ethics committee. It was conducted according to the guidelines of the Declaration of Helsinki on Biomedical Research Involving Human Subjects. All subjects provided their written informed consent.

#### **Experimental subjects**

The investigation included 134 participants (70 males and 64 females, mean age  $\pm$  SD: 29.5  $\pm$  12.6, 18 to 84 years old) without gustatory dysfunction.

#### **Gustatory testing**

The subjects did not eat or drink anything except water, and refrained from smoking, for at least one hour prior to testing. The extended test consisted of 24 taste strips (and two blanks without taste) that were presented to the participants in a pseudo-randomized sequence.

The four standard concentrations of sweet, sour, salty, and bitter taste were augmented with two additional low concentrations of each (sweet: 25/12.5 g/ml sucrose; sour: 27/15 g/ml citric acid; salty: 6.4/2.6 g/ml sodium chloride, bitter: 0.15/0.06 g/ml quinine hydrochloride), resulting in a maximum extended taste score (ETS) of 24 (six concentrations of four taste qualities). Testing started with the lowest concentrations, in order to minimize adaptation and habituation processes. The strips were placed in the middle of the anterior part of the tongue and the mouth was closed (whole-mouth testing). The subjects were asked to respond according to a list of five possible answers (sweet, sour, salty, bitter, no taste). After presentation of each taste strip, the mouth was rinsed with tap water. Each correct answer was awarded one point. Consequently, the ETS ranged from 0 to 24. The time needed for testing was approximately 12 minutes. To obtain test-retest reliability, the same procedure was administered a second time (after a minimum interval of one hour and a maximum interval of 25 hours) in 33 subjects (11 males and 22 females, mean age of 29.0  $\pm$  10.3 years, range 19 to 62 years).

According to previous authors we divided our population into different age groups (18-40 years, 41-60 years and >60 years) <sup>(8)</sup>. The majority of the study population was between 18 and 40 years old (n=118, 63 males and 55 females) meanwhile other age groups were considerably smaller (41-60 years: n=10, 4 males and 6 females; >60 years: n=6, 3 males and 3 females).

#### **Photo-documentation**

High-resolution digital cameras were used for documentation of taste papillae (Panasonic Lumix TZ41/LG P970). In a similar way to the method of Shahbake and colleagues, the surface of the

anterior tongue was colored with blue food dye before taking a picture <sup>(12)</sup>. The pictures were edited with photo editing software (Adobe Photoshop CS2 2005), and an area of approximately 1 cm<sup>2</sup> (according to quarter 5 and 7, as described by Shabake et al.) on the tongue was marked. Subsequently, the number of fungiform papillae in this area was counted. To obtain reliability of the captured results, the papillae were counted separately by two authors (O.I. and D.U.) and the mean values were calculated. If the quality of the photo-documentation was rated insufficient by at least one examiner, data were excluded from the study analysis.

#### **Statistical analysis**

IBM SPSS Statistics 21 (Chicago, IL, USA) was employed for statistical evaluation. For group comparisons we used t-tests for independent samples. Correlation analyses were performed using Spearman statistics. Hierarchical cluster analysis was performed on the raw dataset (extended test with a maximum score of 24). A further cluster analysis was conducted using a reduced data set with four concentrations (short test with a maximum score of 16 according to the previous test version). The betweengroups linkage method was used to identify relatively homogeneous groups of cases. The squared Euclidean distance served as a criterion for the appropriate number of clusters generated. Post-hoc cluster validation was performed by a between-group analysis using the Kruskal-Wallis H-test and by post-hoc analysis using the Mann Whitney U-test. The alpha level was set at 0.05.

#### Results

#### Mean taste score

The mean ETS  $\pm$  SD was 14.5  $\pm$  3.2. It was 4.5  $\pm$  1.2 for sweet, 2.8  $\pm$  1.0 for sour, 3.9  $\pm$  1.3 for salty, and 3.2  $\pm$  1.2 for bitter taste (Table 1). Female subjects scored a significantly better mean ETS than male subjects (females 15.2  $\pm$  3.2, males 13.9  $\pm$  3.1, p = 0.01).

The percentage of correctly identified taste strips for the highest concentrations was 99.3 for sweet, 96.3 for sour, 89.6 for salty and 94.0 for bitter. For the lowest concentrations it was 35.1 for sweet, 11.2 for sour, 32.1 for salty, and 6.7 for bitter. Results for all concentrations are listed in Table 2.

The new test yielded significant negative correlations between the taste scores and the age of the subjects [r134 = -0.379 for the ETS (p < 0.001), r134 = -0.301 for sweet (p < 0.001), r134 = -0.299 for salty (p < 0.001), r134 = -0.212 for sour (p < 0.05), and r134 = -0.249 for bitter (p < 0.001) taste].

The majority of participants was between 18-30 years old, therefore normative values were calculated for this age group separately (Table 3). Within this group female subjects scored a significantly better mean ETS than male subjects (females  $15.6 \pm$ 2.9, males  $14.2 \pm 2.7$ , p = 0.01). Table 1. Mean values ( $\pm$  SD) of the test results. The maximum score was 6 for each taste quality and 24 for the ETS. Test results of all subjects are listed in the table. P-values describe significant differences between males and females.

	Score Mean±SD						
	Total (n = 134)	Male (n=70)	Female (n=64)	p-value			
Total	14.5 ± 3.2	13.9 ± 3.1	$15.2 \pm 3.2$	0.017			
Sweet	4.5 ± 1.2	4.3 ± 1.2	4.8 ± 1.2	0.01			
Sour	2.8 ± 1.0	2.8 ± 1.0	2.8 ± 1.0	0.998			
Salty	3.9 ± 1.3	3.8 ± 1.4	4.2 ± 1.2	0.123			
Bitter	3.2 ± 1.2	2.9 ± 1.3	3.4 ± 1.0	0.025			

#### **Cluster analysis**

The cluster analysis showed that the extended test version is able to differentiate the subjects into three distinct groups: One group was in the range of hypogeusia with a median score of 11 and a maximum score of 12; a second group of normogeusic subjects with a median score of 14 up to a maximum score of 16; and a third group with superior gustatory function with a median score of 18 up to a maximum score of 20. On the other hand using the shorter dataset with four concentrations, only two distinct groups of subjects were detected (see Figure 1 and Table 4). Post-hoc validation showed a significant overall difference between the three groups using the extended test version (df = 2; p < 0.001). Post-hoc analysis showed that cluster 1 differed significantly from cluster 2, and also cluster 2 from cluster 3 (Z = -8.0 and -8.6; all p < 0.001). The same was true for the two clusters generated from the short test version (Z = -5.5; p < 0.001).

#### **Test-Retest reliability**

Test-Retest reliability was assessed in 33 subjects. For the taste strips, the correlation coefficient for test and retest of the total ETS was r33 = 0.74 (p < 0.001); for individual taste qualities it was r33 = 0.58 for sweet (p < 0.001), r33 = 0.78 for salty (p < 0.05), r33 = 0.76 for sour (p < 0.001), and r33 = 0.49 for bitter (p < 0.01).

#### Correlation of papillae count and the ETS

The number of fungiform papillae could be reliably counted in 105 patients. The average number of papillae in the counted fields was  $13.1\pm3.8$  (range 6 to 25). Analyses revealed significant correlation between the number of papillae and the total ETS (r105=0.28, p<0.001). For individual taste qualities, only sweet taste reached a significant level of correlation (r105=0.312, p<0.001).



Figure 1. Cluster analysis. Results from the cluster analysis for the short and extended test version: Subjects' total scores in the extended version showed a ceiling effect due to the small number of subjects scoring at 20 out of possible 24 (n=3).

The cluster analysis provided 3 groups of subjects in the extended version. Using the short test version, the cluster analysis did not discriminate the subgroup with higher gustatory sensitivity. Therefore, only 2 different groups of subjects were detected. Post hoc analysis also showed significant different scores between the clusters within each test version (see text). Data are presented as median (Q0.5; line), quartieles (Q0.25, Q0.75; boxes), and the 10th/90th percentiles (Q0.10 and Q0.90; whiskers).

#### Discussion

The present investigation provides the first data from an extended test of gustatory function using the established taste strips test. The aim was to create a tool for the assessment of taste acuity in subjects with superior sensitivity to sweet, sour, salty, and bitter taste. Recent research has shown that the assessment of subjects with enhanced gustatory function might be clinically relevant. Hypersensitivity of taste receptors seems to play a role in the pathophysiology of chronic inflammatory diseases, e.g., chronic rhino-sinusitis (13-15). Subjects with high sensitivity to bitter taste are supposed to have an increased local immune response due to increased nitric oxide production leading to enhanced mucociliary clearance and killing of bacteria <sup>(16)</sup>. Recent findings have revealed an antagonistic role of sweet and bitter taste receptors in the human respiratory epithelium during defense against microbial infection <sup>(17)</sup>: it was shown that activation of sweet taste receptors inhibited the effects of bitter taste receptor-mediated secretion of antimicrobial peptides. These studies demonstrate the need for an easy and valid test of gustatory function that might prove the clinical influence of taste sensitivity on pathophysiological mechanisms involved in upper respiratory epithelium.

The need for lower concentrations in the assessment of taste perception was also suggested in a recent study by Doty and

### Wolf et al.

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Taste	Correct	No taste	Sweet	Sour	Salty	Bitter
Sweet 1	99.3	0.7		0	0	0
Sweet 2	95.5	0.7		1.5	1.5	0.7
Sweet 3	94.0	1.5		3.0	1.5	0
Sweet 4	77.6	11.9		4.5	4.5	1.5
Sweet 5	53.7	26.1		6.7	6.7	6.7
Sweet 6	35.1	49.3		5.2	6.7	3.7
Sour 1	96.3	0	0		0.7	3
Sour 2	84.3	0.7	0.7		10.4	3.7
Sour 3	59.0	14.2	0.7		17.2	9
Sour 4	16.4	47.0	4.5		17.2	14.9
Sour 5	12.7	52.2	6.0		13.4	15.7
Sour 6	11.2	58.2	6.7		15.7	8.2
Salty 1	89.6	0.7	0	4.5		5.2
Salty 2	85.1	0.7	0.7	6.6		6.7
Salty 3	81.3	2.2	0.7	10.4		5.2
Salty 4	65.7	5.2	3.7	20.9		4.5
Salty 5	44.0	25.4	3.4	17.2		9.7
Salty 6	32.1	42.5	5.2	9.7		10.4
Bitter 1	94.0	4.5	0	0.7	0.7	
Bitter 2	89.6	6.0	0	2.2	2.2	
Bitter 3	69.4	23.1	1.5	3.0	3.0	
Bitter 4	39.98	44.8	2.2	7.5	6.7	
Bitter 5	19.4	56.7	6.0	5.2	12.7	
Bitter 6	6.7	73.1	6.7	8.2	5.2	

Table 2. Results for each taste strip in percentages. Results listed for each of the 24 concentrations of taste strips from 116 subjects. 1=highest, 6=lowest concentration. Correct: percentage of correctly identified taste strips. No taste: percentage of taste strips, which had been mistakenly identified as having no taste. The four columns Sweet, Sour, Salty, Bitter indicate the percentage of confusion with other taste qualities.

colleagues, investigating the gustatory side effects of eszopiclone, a hypnotic drug. Here, the authors observed better correlation of intensity estimates with plasma or saliva drug levels by using taste strips with lower concentrations <sup>(18)</sup>.

Different tests of gustatory function using a variety of techniques [e.g., taste tablets <sup>(19)</sup>, filter paper discs <sup>(20)</sup>, cotton swabs <sup>(21)</sup>, liquids <sup>(22-24)</sup>, edible wafers <sup>(25)</sup> and impregnated taste strips <sup>(6)</sup>] were introduced over the past few decades. Assessment of gustatory function with impregnated taste strips is an easy, cost-

effective and reliable method. Moreover, the process allows testing of specific areas on the tongue (e.g., for detection of chorda tympani nerve injury in middle-ear surgery or glossopharyngeal nerve injury after tonsillectomy) and has a long shelf life. While some authors prefer taste tests based on forced-choice paradigm other authors prefer non-forced choice testing procedures. Major advantage of the forced choice test is the avoidance of subject's response bias <sup>(8)</sup>. The advantage of non-forced choice models is the possibility to distinguish between percepTable 3. Normative values of subjects between 18-40 years. Normative values derived from 118 healthy volunteers between 18-40 years for the extended taste strips. Maximum score was 6 for each taste quality and 24 for the ETS. The 10th and 90th percentile can be used to separate normo-geusic from hypogeusic and hypergeusic subjects 30,31. Test results of the population between 18-40 years, males and females are listed in the table.

Percentile	ETS (n =118)	Sweet (n =118)	Sour (n =118)	Salty (n =118)	Bitter (n =118)	ETS, males (n =63)	ETS, females (n =55)
5th	9	3	1	2	1	10	9
10th	11	3	2	2	2	11	11
50th	15	5	3	4	3	14	16
90th	19	6	4	6	5	18	19
95th	19	6	4	6	5	19	19

Table 4. Cluster analysis with descriptive score data. Results revealed that the extended test version is able to separate three groups of subjects. The original test version separated only two groups of subjects.

Extended Test Version						
Cluster number	N	Mean ± SD	Minimum	Maximum	Median	
1	32	$10.2 \pm 1.9$	4	12	11	
2	61	14.3 ± 1.0	13	16	14	
3	41	18.1 ± 1.0	17	20	18	
Total	134	14.5 ± 3.2	4	20	14	
Short Test Version						
Cluster number	N	Mean ± SD	Minimum	Maximum	Median	
1	11	7.1 ± 1.2	4	8	7	
2	123	12.9 ± 1.7	9	16	13	
Total	134	12.4 ± 2.3	4	16	13	

tion of tastes and correct identification of tastes. Moreover, due to limited items (i.e., taste qualities) in taste tests, the chance hit rate might be too high without blanks, as in the non-forced-choice paradigm. Consequently, the taste strips test was validated as a non-forced-choice paradigm <sup>(6)</sup>.

The established taste strips test shows a ceiling effect in subjects with low gustatory thresholds that was not found with the extended taste strips test. By using six instead of four concentrations of each taste quality, the present results yielded no subject with the maximum score of <sup>(24)</sup>. The lowest concentrations of the extended taste strips could be correctly identified by 7 to 35% of the population compared to 36 to 53% in the established taste strips test. The absence of a ceiling effect makes the extended taste strips test more sensitive in the assessment of taste function in subjects with superior gustatory acuity.

The first data of the extended taste test helped us define an ETS and establish normative data for a healthy population between 18 and 30 years. The new test supported gender differences in gustatory sensitivity reported earlier <sup>(26,27)</sup>. Women significantly

outperformed men on the ETS, sweet and bitter taste score. Our findings also supported earlier evidence that taste becomes less acute as humans age <sup>(6,8)</sup>.

Cluster analysis showed that the extended test is able to separate three groups of subjects with significant differences in gustatory function. Using the shorter data set with four concentrations only, only two distinct groups of subjects were detected. The results of the retest in 33 subjects showed good reliability regarding the ETS and the scores for each taste.

Fungiform papillae density can be used as an indicator for taste function <sup>(21,28)</sup>. Interstingly, Shabake et al. showed the correlation between certain areas on the tongue and the number of papillae on the rest of the anterior tongue <sup>(12)</sup>. In accordance to these results our results yielded a correlation of the ETS with the number of papillae in these certain areas. Consequently, also the new extended test version seems to correlate with the number of papillae on the tongue and taste function.

The results for individual taste qualities showed that the sweet taste score correlated significantly with a higher number of

papillae, whereas salty, bitter, and sour taste did not. One explanation might be that sweet taste perception is most sensitive on the tip of the tongue. Our results agree with previous observations showing that the number of taste pores correlates most strongly with the perception of sweet taste <sup>(28)</sup>. Conversely, recent research addressing the role of fungiform papillae density in relation to taste sensitivity to the bitter substance propylthiouracil did not show any relationship between bitter taste sensitivity and density of fungiform papillae, which is consistent with our findings <sup>(29)</sup>.

test is a suitable method for investigating taste performance in subjects with superior gustatory performance. It offers the additional ability to assess gustatory function in these subjects without any obvious ceiling effect.

# Authorship contribution

CM, BR, AW: Study design and concept; AW, CM, CU, OI: Data ourcing; CM, BR, AW: Investigation of data, statistical analysis, manuscript draft and revision.

# **Conflict of interest**

No conflict of interest exists.

# Conclusion

#### Taken together, our findings show that the extended taste strips

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