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Effects of olfactory and gustatory stimuli on the biomechanics of swallowing

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ABSTRACT

We have previously documented increased amplitude of motor-evoked potentials (MEPs) from the submental muscles during volitional swallowing following simultaneous odor and tastant stimulation. The MEP denotes neural excitability from the motor cortex to the target muscle(s). However, it is unknown if changes in the MEP transfer to the swallowing muscles to facilitate improved swallowing. Thus, we sought to evaluate changes in the biomechanics of swallowing following stimulation protocols that are known to influence neural excitability. Sixteen healthy participants were exposed to low and high concentrations of lemon odor and tastant. The odor and tastant concentrations which produced the highest amplitude of submental electromyography (EMG) were then combined for simultaneous stimuli presentation. Outcome measures included EMG from the submental muscles, as well as lingual and pharyngeal manometry. Poststimulation results showed decreased midglossopalatal pressure at 30 min and decreased duration at anterior and midglossopalatal pressure and increased EMG duration at 60 min. This study strengthens the justification for the use of flavor in managing patients with dysphagia as long-term changes were present in the poststimulation period.

1. Introduction

Combined olfactory and gustatory stimulation (flavor) has been shown to increase neural excitability in healthy participants, as measured by the amplitude and latency of motor-evoked potentials (MEPs) recorded from the submental muscles [1]. Increased MEP amplitude has been associated with neuroplastic changes in the unaffected hemisphere of nondysphagic poststroke patients compared to patients with dysphagia following stroke who showed no changes in their unaffected hemisphere [2,3]. Although we have reported increased MEP amplitude following simultaneous odor and tastant stimulation, changes in neural excitability do not directly imply functional changes in swallowing. Similarly, an absence of change in neural excitability would not necessarily suggest an absence of functional change in swallowing.

Submental muscles, comprised of the anterior belly of digastric, mylohyoid, and geniohyoid muscles, are involved in the superior and anterior excursion of the hyolaryngeal complex, which is an important biomechanical event to facilitate opening of the upper esophageal sphincter (UES) for bolus transfer [4]. Surface electromyography (sEMG) of the submental muscles is a noninvasive method to study swallowing function [5–7]. Although normal swallowing function is highly variable across individuals, EMG can be used to compare within-subject swallows [6]. Several studies have evaluated EMG of the submental muscles following sour taste stimulation. The submental muscles were found to contract earlier when sour taste was used, compared to a no-taste condition [8]. Contractions of the submental muscles were stronger and the onsets were closer across the three muscles when sour bolus was presented compared to a control condition [9]. EMG recordings of submental muscle contraction were greatest when recorded during swallowing of sour taste, compared to sweet, salty, or bitter [10]. When mechanical, cold, and/or sour stimulation was presented to the anterior faucial pillars, there was a shorter latency in the first swallowing activity when all three conditions were combined, compared to no stimulation, but no changes in the duration of submental contraction were detected [11]. Conversely, another study identified no differences in submental EMG recordings when either high or low concentration of sour food was ingested [12].

Prior to swallowing, the tongue generates pressure which propels a bolus into the pharynx by squeezing the tongue to the palate in an anterior to posterior movement [13]. The pattern of pressure generation in the oral cavity has been systematically studied using pressure transducers secured in a base plate, similar to a denture, which a volunteer wears [14,15]. This method guarantees that the transducers are in situ at all times, ensuring the reliability and stability of the recorded pressures; however, it requires custom-fitted

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hardware. Measures of pressure data in healthy participants, as well as in patients with head and neck cancers, have also been reported to be reliable and stable when using a commercially available lingual pressure bulb (Kay® Digital Swallowing Workstation, Kay Elemetrics Corporation, Lincoln Park, New Jersey, USA) [16,17]. The normal swallowing pattern in healthy individuals was not altered with the presence of the lingual bulb in the mouth [18]. Using this system, lingual pressure was increased when 10-ml chilled sour boli were presented compared to water [19]. It is possible that retronasal odors may have also contributed to the higher lingual pressures seen in that study. Furthermore, bolus volume or temperature, or both, may have contributed to the increased pressure.

The pharynx contracts in a superior to inferior direction to transfer the bolus into the esophagus [20]. Adequate pharyngeal pressure during swallowing clears the pharynx of residue [21]. If inadequate pressure is generated, postswallow residue in the pharynx can enter the airway when the airway re-opens to resume breathing [22,23]; therefore, measurement of pharyngeal pressure provides a valuable indicator of successful swallowing. Pharyngeal pressure can be measured by solid state manometry [20]. Many studies have looked at pharyngeal pressure following other behavioral interventions [24–29] but no study has evaluated the immediate effects of odor or taste on pharyngeal pressure during swallowing. Moreover, to our knowledge, no poststimulation data exists to document the effects of sensory stimulation on the biomechanics of swallowing over a long time course.

The current study is a follow-up to our MEP research which has shown increased MEP amplitude during swallowing following simultaneous odor and tastant stimulation, indicating that the neural substrates involved in swallowing are modulated following sensory intervention. The current study aimed to determine if the same stimulation would change biomechanical swallowing function by way of changes in the contraction of the submental muscles, the pressures in the oral cavity and pharynx, and/or the dynamics of the UES. We hypothesized that there would be an increase in the amplitude of submental surface EMG, lingual and pharyngeal pressures, and the negative pressure in the UES when flavor is presented compared to no stimulation.

2. Methods

A repeated-measures within-subject study design was used to evaluate changes in the biomechanical aspects of swallowing. Ethical approval was obtained from the regional Health and Disability Ethics Committee.

2.1. Participants

Sixteen healthy participants aged 19–47 years (mean 27.5, SD 7.8) were recruited. They reported no previous history of neurological problems or dysphagia and were not taking medication that could affect swallowing. They were all asked not to ingest caffeine, alcohol, or spicy food during the hour prior to the procedures to ensure our stimuli were not contaminated by chemical residues of food in the mouth.

2.2. Stimuli

Low (25%) and high (100%) concentrations of lemon concentrate (Country Gold lemon juice, Steric Trading Pty Ltd, Villawood, NSW, Australia) were utilized in this study. Tap water was used as control. The odor was presented as a mist via nasal cannula attached to a nebuliser (DeVilbiss PulmoMate® compressor/nebuliser, Model 4650I, Sunrise Medical, Somerset, Pennsylvania, USA) and taste was presented by placing filter paper strip (Genuine Whatman Filter Paper No. 5, W & R Balston, Maidstone, Kent, UK) impregnated with the stimulus on the tongue.

2.3. Procedures

Participants provided written informed consent prior to the procedures. Additionally, they were also asked to complete a brief medical questionnaire to confirm that they met the inclusion and exclusion criteria to participate in the study. Prior to data collection, the tongue array and pharyngeal manometer were calibrated following the manufacturer's recommendation.

The participants were seated comfortably in a chair and the surface under the chin was cleaned vigorously with an alcohol swab. A triode surface electrode 5.4 cm in diameter (disposable pregelled electrode pads, standard silver/silver chloride EMG electrodes, Multi Bio Sensors, El Paso, Texas, USA) was placed under the chin, between the spine of the mandible and the superior border of the thyroid cartilage. The two active electrodes were positioned in the midsagittal plane and the ground electrode was positioned laterally. The differential EMG signal of the submental muscles was amplified, band-pass filtered (50–220 Hz), rectified, low-pass filtered at 3 Hz, and digitized at 1000 Hz. The EMG recording system is part of the Kay® Digital Swallowing Workstation. The averaged and rectified EMG waveforms were checked to ensure that clear EMG recordings were achieved.

Next, the manometer was inserted transnasally. We used a solid state pharyngeal manometer 2.1 mm in diameter, with three pressure transducers measuring 2×5 mm, which were oriented toward the posterior pharyngeal wall, to record pressures in the pharynx and UES. As the catheter reached the posterior aspect of the participant's nasal cavity, the participant was asked to look briefly to the ceiling to reduce the nasopharyngeal angle so that the catheter could be inserted into the pharynx. Then, with the head back to neutral position, he/she was handed a glass of tap water and asked to rapidly drink the water through a straw. In doing so, the distal portion of the catheter was swallowed into the esophagus. The participants were asked to swallow until the catheter was pulled down 30 cm as measured from the tip of the nose. It was then slowly pulled out again until it was in the appropriate location to measure the information needed for this study. When positioned correctly, the first, second, and third sensors recorded pressures from the oropharynx, hypopharynx, and UES, respectively, during swallowing [30]. The M wave [31,32] was observed in the third sensor during swallowing, indicating its correct placement within the UES. When the catheter was correctly placed, it was taped securely to the external nose with adhesive tape.

Lingual swallowing pressures were measured with a three-bulb lingual pressure array placed onto the palatal vault by means of oral adhesive (Stomahesive® strips, ConvaTec, Princeton, New Jersey, USA). The lingual pressure device is a component of the Kay® Digital Swallowing Workstation and measures glossopalatal pressures corresponding to the anterior, middle, and posterior part of the tongue. However, as some participants could not tolerate the posterior sensor, which when the array was secured onto the palate was approximately between the junction of the hard and soft palate, it was removed. Thus, data was recorded only from the anterior and middle sensors. Consistency in placement was established by placing the anterior sensor 5 mm posterior to the incisive papilla [14]. Each sensor was 13 mm in diameter and the spacing between sensors was 8 mm. All data were recorded concurrently with a sampling rate of 1000 Hz.

When the participant was ready, he/she executed five relaxed dry (saliva) swallows, which were taken as baseline measures. Stimuli were then randomly presented: control odor, low odor, high odor, control tastant, low tastant, and high tastant. The odor stimuli were presented continuously for 1 min, then paused for 15 s to avoid

adaptation [33]. The odor was presented again for another minute, and the cycle repeated until all data were recorded. A fresh taste stimulus was used after three swallows to ensure adequate taste stimulation. Participants were asked to breathe normally during stimulus presentation, and to swallow their saliva approximately once every 30 s. The instructions given were: "You may now swallow whenever you are ready", after the paper strip was placed on the tongue or the nebuliser was switched on for 10 s to ensure that the odor stimulus has reached the nostrils. Participants completed five repetitions of a dry swallow with each of the stimulus. The concentrations of odor and tastant that best stimulated a participant's swallowing when presented on its own (based on the largest EMG amplitude) were then combined for the simultaneous presentation of odor and tastant. The high concentration stimuli were used if no differences were detected. Using the same method as when the olfactory and gustatory stimuli were presented independently, five dry swallows were recorded during the combined odor and tastant stimulation, which was denoted as time = 0 min. Five dry swallows were again recorded at 30-, 60-, and 90-min poststimulation, as was done in our MEP study upon which this research was based [1]. Data were saved on the computer for offline analyses (Kay® Digital Swallowing Workstation, Kay Elemetrics Corporation, Lincoln Park, New Jersey, USA). Confidentiality was assured by assigning a coded numerical identification for each participant.

2.4. Data analyses

Preliminary analyses of the mean EMG amplitudes were completed on the low and high concentrations of odor and tastant for each participant. The concentration that produced greater EMG amplitude was selected for simultaneous presentation of both stimuli. Data from the combined odor and tastant stimulation were subjected to two separate repeated-measures ANOVAs to evaluate immediate (during stimulation compared to baseline) and late (at 30-, 60-, and 90-min poststimulation compared to baseline measures) effects of sensory stimulation on swallowing biomechanics. Data were analyzed with SPSS 17.0 (SPSS Inc, Somers, New York, USA).

Initial analysis included sex as a covariate; if it was not significant, the analysis was recalculated without sex. Pharyngeal manometry analyses were done separately for the pharyngeal pressures (the first and second sensors) and the sensor in the UES. Additionally, the time difference between the peak pressures at the first and second sensors was also analyzed (the peak-to-peak timing). Oral pressures and EMG data were analyzed separately in two additional analyses. $p<0.05$ was taken as significant. For all analyses, Greenhouse–Geisser correction was reported if Mauchly's test of sphericity was significant, suggesting that the assumption of sphericity was violated. *t*-tests comparing baseline measures with poststimulation data were also carried out even if the ANOVAs showed no significant differences, as data from our MEP study showed significant changes at 30-, 60-, and 90-min poststimulation [1].

3. Results

Sex was not significant in all initial ANOVAs analyses, thus analyses were recalculated without sex. A figure of sEMG, lingual pressure, and pharyngeal manometry waveforms captured concurrently is shown in Fig. 1. Although the phases of swallowing cannot be explicitly defined by these methods, one can infer, based on an understanding of swallowing biomechanics, the end of oral phase and the start of pharyngeal phase.

3.1. Electromyography of the submental muscles

EMG amplitude and duration at baseline, during combined odor and tastant stimulation, and at 30-, 60-, and 90-min poststimulation are tabulated in Table 1. EMG amplitude and duration during simultaneous odor and tastant presentation were not different from baseline. Repeated-measures ANOVAs for EMG amplitude and duration across time were also not significant. However, *t*-tests showed increased EMG duration 60 min poststimulation compared to baseline [$t(15)=2.13$, $p=0.050$].

3.2. Lingual pressures

The amplitude and duration of lingual pressures at baseline, during combined odor and tastant stimulation (immediate effect), and at 30-, 60-, and 90-min poststimulation (late effects) are tabulated in Table 2. For immediate effects, the analyses for pressure amplitudes and durations were significant for interaction between the tongue sensor (anterior vs middle) and condition (baseline vs during stimulation) [$F(1, 15)=26.3$, $p<0.0001$ and $F(1, 15)=53.7$, $p<0.0001$, respectively]. The durational analysis for the main effect of tongue sensor (anterior vs middle) was also significant [$F(1, 15)=5.5$, $p=0.033$]. Further, *t*-tests showed increased pressure and duration of glossopalatal contact at anterior tongue when simultaneous odor and tastant stimulation was presented compared to baseline [$t(15)=2.6$, $p=0.022$ and $t(15)=2.9$, $p=0.012$, respectively].

In contrast to the immediate effect, the repeated-measures analyses showed no effect for the poststimulation data, or late effects. However, *t*-tests showed decreased pressure at midglossopalatal contact 30 min poststimulation compared to baseline [$t(15)=3.2$, $p=0.006$] and decreased duration for anterior and midglossopalatal contact at 60 min poststimulation compared to baseline [$t(15)=2.3$, $p=0.035$ and $t(15)=2.2$, $p=0.048$, respectively].

3.3. Pharyngeal pressures

For immediate effects, repeated-measures ANOVAs for the peak pharyngeal pressures were significant for the main effect of condition (baseline vs stimulation) and the interaction between condition and the sensor (the first and second sensors) [$F(1, 15)=5.01$, $p=0.041$ and $F(1, 15)=8.21$, $p=0.012$, respectively]. Further *t*-tests showed decreased contact pressure at the second sensor during stimulation compared to baseline [$t(15)=3.2$, $p=0.006$; Table 3). No pressure differences were recorded from the sensor in the UES. Repeated-measures for durational measures for the first and second sensors were also different [$F(1, 15)=21.0$, $p<0.0001$]. The contact duration at the first sensor was longer than the second sensor (Table 3). No differences in duration were detected for the sensor in the UES or in the peak-to-peak timing.

For late effects, differences were found in the amplitude of contact pressure at the first and second sensors [$F(1, 15)=4.5$, $p=0.050$]. Pressures recorded at the second sensor were higher than the first sensor (Table 3). No pressure differences were computed for the sensor in the UES. Repeated-measures ANOVA for the durations and the sensors (first and second) showed a significant main effect of sensor and time [$F(1, 15)=21.3$, $p<0.0001$ and $F(3, 45)=3.38$, $p=0.026$, respectively]. Pressure durations at the first sensor were higher than the second sensor (Table 3). *t*-tests comparing the durations of the first and second sensors at baseline with 30-, 60-, and 90-min poststimulation showed no differences. No durational differences were detected in the UES and peak-to-peak timing.

4. Discussion

This is the first study to investigate immediate and late changes in the biomechanics of swallowing following simultaneous odor and taste stimulation using both EMG and pressure measurements. Patterns of change in the peripheral biomechanics were found which—to some extent—parallel patterns of neural change documented in our previously published study of MEPs associated with sensory

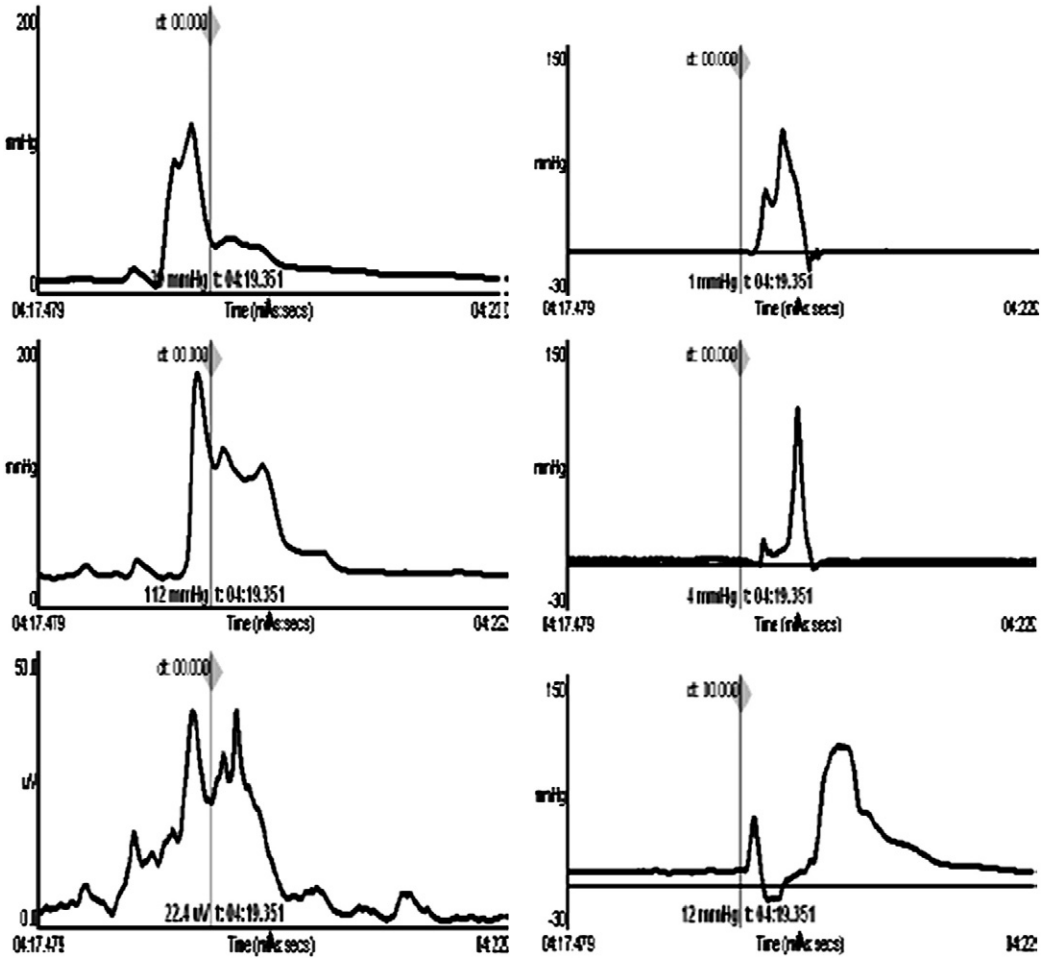

Fig. 1. The averaged and rectified waveforms of submental EMG (lower left), anterior and middle lingual pressures (upper and middle left, respectively), and pharyngeal manometry (right, with oropharynx, hypopharynx, and UES pressures sequentially from top to bottom) recorded from one participant. The vertical line indicates the likely boundary between the oral and pharyngeal phases of swallowing. Note the peak of lingual pressures during oral phase of swallowing to the left side of the vertical line and the occurrence of pharyngeal pressure changes during swallowing to the right side of the vertical line. Lingual pressure is apparent during oral phase of swallowing to facilitate bolus transfer into the pharynx and is maintained during the pharyngeal swallow. Submental sEMG and midlingual activation is apparent during both oral and pharyngeal phases of swallowing.

stimulation. A summary of significant findings from our MEP and biomechanical studies are presented in Table 4.

Our earlier MEP study documented no immediate effects of paired olfactory and gustatory stimulation [1]. However, the current study found immediate biomechanical changes during flavor stimulation. These changes included increased pressure and duration of tongue-to-palate contact at the anterior tongue and decreased contact pressure at the second pharyngeal sensor (in the hypopharynx) when simultaneous odor and tastant stimulation was presented compared to baseline. Other studies have documented increased submental muscle contraction or lingual pressure when sour taste was presented [8–10,19]. In the current study, there was a trend toward increased submental muscle contraction following simultaneous odor and tastant stimulation compared to baseline but it was not significant (Table 1). A larger sample size may have revealed the differences.

Table 1
Mean (SD) amplitude and duration for submental EMG.

	Amplitude (μV) (SD)	Duration (s) (SD)
Baseline	51.0 (17.3)	1.31 (0.28)
Combined stimulation	55.9 (23.7)	1.46 (0.33)
30 min post	53.4 (18.1)	1.39 (0.33)
60 min post	51.5 (17.8)	1.41 (0.32)*
90 min post	52.8 (18.0)	1.34 (0.27)

* $p<0.05$ compared to baseline.

At baseline, the midglossopalatal contact produced greater pressure than its anterior counterpart, comparable to another study [13]. However, a higher pressure was recorded in the anterior tongue during stimulation compared to midglossopalatal contact, as reported by other researchers [19]. We hypothesized that increased activation of the facial and glossopharyngeal nerves, which carry taste information from the oral cavity and pharynx, would subsequently activate more sensory neurons in the nucleus tractus solitarius (NTS). Moreover, flavor stimulation may have activated other brain areas, such as the insula, which also feeds sensory information into the NTS [34]. Information from the NTS is conveyed to the motor neurons in the nucleus ambiguus (NA), which contains motor neurons involved in swallowing (cranial nerves IX and X). Consequently, there would be

Table 2
Mean (SD) amplitude and duration for glossopalatal pressures.

	Anterior glossopalatal		Midglossopalatal	
	Pressure (mm Hg) (SD)	Duration (s) (SD)	Pressure (mm Hg) (SD)	Duration (s) (SD)
Baseline	150.3 (81.1)	1.50 (0.21)	184.7 (66.9)	1.48 (0.27)
Combined stimulation	187.4 (98.6)*	1.73 (0.27)*	162.2 (79.7)	1.49 (0.39)
30 min post	134.7 (107.1)	1.41 (0.34)	156.8 (64.4)*	1.42 (0.35)
60 min post	147.9 (102.1)	1.35 (0.23)*	161.1 (72.9)	1.37 (0.32)*
90 min post	150.6 (102.3)	1.29 (0.37)	161.6 (66.7)	1.43 (0.33)

* $p<0.05$ compared to baseline.

Table 3
Mean (SD) amplitude and duration for pharyngeal pressures.

	First sensor		Second sensor	
	Pressure (mm Hg) (SD)	Duration (s) (SD)	Pressure (mm Hg) (SD)	Duration (s) (SD)
Baseline	92.2 (22.4)	0.48 (0.09)	111.1 (34.0)	0.36 (0.12)
Combined stimulation	92.4 (29.3)	0.45 (0.11)	94.0 (23.5)*	0.35 (0.12)
30 min post	93.4 (28.8)	0.46 (0.11)	104.2 (32.8)	0.36 (0.11)
60 min post	92.6 (25.8)	0.46 (0.10)	105.4 (36.8)	0.36 (0.13)
90 min post	90.4 (24.1)	0.48 (0.10)	113.3 (32.3)	0.37 (0.13)

* $p<0.05$ compared to baseline.

more motor neurons activated in the NA; the neural signals may then be conveyed via monosynaptic or interneuronal connections [35,36] to other cranial motor nuclei involved in swallowing (cranial nerves V, VII, and XII). A similar hypothesis has been suggested previously by others [8,10,19,37].

A previous taste study used 10-ml chilled sour bolus [19], where bolus volume or temperature, or both, may have influenced the results. Thus, the present study used filter paper strips impregnated with lemon concentrate at room temperature to ensure that the volume and temperature effects were controlled.

The current study found decreased contact pressure at the hypopharynx during stimulus presentation. Pressure at this site has been shown to correlate negatively with oral and pharyngeal transit times and pharyngeal response time [21] and with submental muscle contraction [30]. Our findings are comparable to the previous reports, as we recorded lower hypopharyngeal pressure and increased anterior glossopalatal contact pressure and duration during stimulus presentation compared to baseline. The decreased hypopharyngeal pressure has been suggested to be due to the close proximity of the second sensor to the UES [38]. Similarly, a transient negative subatmospheric pressure has been recorded in the hypopharynx during dry swallows, which was suggested as resulting from expansion of pharynx during swallowing [39].

Our MEP study [1] suggested late changes in submental muscle contraction. We proposed that the mechanism of long-term potentiation (LTP), a function of neural plasticity [40], was responsible for changes in MEP amplitude poststimulation. LTP is an increase in synaptic strength transmission which leads to more efficient neural communication. Persistent LTP activity will lead to long-term neural change which may contribute to recovery in patients with dysphagia. The late effects seen in the current study were detected in the submental muscles and the glossopalatal measures but no pharyngeal changes were seen. Changes in the cortical areas involved in swallowing have been reported to begin long before changes were seen at the periphery [41]. Although the submental muscles are involved in the opening of the UES, changes in the UES may not have been detectable during the course of our data collection.

The changes in submental EMG and lingual pressures were not significant at 90 min poststimulation, in contrast to our MEP data [1] where changes were detected up to 90 min poststimulation. However, unlike MEPs, which reflect neural excitability and transmission, biomechanical data are highly influenced by variations in voluntary behaviour which may have obscured a small, but real, biomechanical effect at 90 min.

Poststimulation changes in submental EMG and lingual pressures following flavor stimulation have not been previously reported. The submental EMG and lingual pressures are two measures which have been shown to have a low correlation [42]. Therefore, an increase in one measurement does not mean that there should be an increase in the other. In the current study, we found increased EMG duration but decreased glossopalatal contact duration compared to baseline. However, no significant increase in EMG amplitude was recorded compared to baseline. Our relatively small sample size may have limited our ability to detect differences but there was a trend of increased amplitude and duration of the EMG. The poststimulation results showed decreased midglossopalatal pressure and contact duration and decreased anterior glossopalatal contact duration. Decreased durations may be explained by increased efficiency in the oral phase, which appeared as faster oral transit time compared to baseline [43]. The decreased pressure at midglossopalatal contact could be explained by the existence of negative tongue pressure when the tongue moved away from the palate [14,44], which could not be measured via our method.

In the current study, changes in the biomechanics of swallowing were identified primarily during the volitional oral stage of swallowing. This may provide further evidence that the different stages of swallowing are controlled by different neural pathways, or utilize different levels of cortical involvement, or both. A similar hypothesis has been proposed by others [45]. More work is needed to further explore this hypothesis.

In conclusion, the simultaneous presentation of odor and tastant—that is, flavor—can change the biomechanical aspects of swallowing which are under volitional control. As these changes were evident even after the stimulus was removed, its use in therapy could be of great value, particularly for patients with cognitive deficits who have problems following instructions in a standard rehabilitation program. Follow-up research to investigate the effects of flavor on swallowing function in the elderly and in patients with dysphagia would lend support to the use of sensory stimulation in managing patients with dysphagia.

Table 4
Summary of significant findings from MEP and biomechanical studies following simultaneous odor and tastant stimulation.

	Amplitude measures		Temporal measures	
	Immediate effect	Late effect	Immediate effect	Late effect
Submental MEP*		Increased		
Submental EMG				Increased
Anterior glossopalatal pressure	Increased		Increased	Decreased
Midglossopalatal pressure		Decreased		Decreased
Oropharyngeal pressure				
Hypopharyngeal pressure	Decreased			
UES				

* Data from previous study [1].

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