Tactile stimulation of the oropharynx elicits sympathoexcitation in conscious humans

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Muller MD, Mast JL, Cui J, Heffernan MJ, McQuillan PM, Sinoway LI. Tactile stimulation of the oropharynx elicits sympathoexcitation in conscious humans. J Appl Physiol 115: 71–77, 2013. First published April 18, 2013; doi:10.1152/japplphysiol.00197.2013.—Tactile stimulation of the oropharynx (TSO) elicits the gag reflex and increases heart rate (HR) and mean arterial pressure (MAP) in anesthetized patients. However, the interaction between upper-airway defense reflexes and the sympathetic nervous system has not been investigated in conscious humans. In Experiment 1, beat-by-beat measurements of HR, MAP, muscle sympathetic nerve activity (MSNA), and renal vascular resistance (RVR) were measured during TSO and tactile stimulation of the hard palate (Sham) in the supine posture. In Experiment 2, TSO was performed before (pre) and after (post) inhalation of 4% lidocaine via nebulizer. Rate pressure product (RPP) was determined. Compared with Sham, TSO elicited the gag reflex and increased RPP (absolute change (Δ)36 ± 6 vs. 17 ± 5%), MSNA (Δ122 ± 39 vs. 19 ± 19%), and RVR (Δ55 ± 11 vs. 4 ± 4%). This effect occurred within one to two cardiac cycles of TSO. The ΔMAP (12 ± 3 vs. 6 ± 1 mmHg) and the ΔHR (10 ± 3 vs. 3 ± 3 beats/min) were also greater following TSO compared with Sham. Lidocaine inhalation blocked the gag reflex and attenuated increases in MAP (Δpre: 16 ± 2; Δpost: 5 ± 2 mmHg) and HR (Δpre: 12 ± 3; Δpost: 2 ± 2 beats/min) in response to TSO. When mechanically stimulated, afferents in the oropharynx not only serve to protect the airway but also cause reflex increases in MSNA, RVR, MAP, and HR. An augmented sympathoexcitatory response during intubation and laryngoscopy may contribute to perioperative cardiovascular morbidity and mortality.

sympathetic nervous system; afferent; pharynx; local anesthesia; blood flow

Many upper-airway defense reflexes (e.g., cough reflex, gag reflex, swallow reflex) originate in the mouth, pharynx, and/or larynx (3, 7, 25). These upper-airway reflexes are operable under physiological conditions and effectively route food and air to the proper anatomical locations. During an acute perturbation, such as choking or coughing, blood pressure (BP) homeostasis is likely to be affected. Tactile stimulation of the oropharynx (TSO) is also a potent stimulus that increases heart rate (HR), BP, and rate pressure product (RPP) in humans. Specifically, endotracheal intubation, laryngoscopy, and bronchoscopy are clinical procedures that increase myocardial oxygen demand (24, 45, 47, 48). Considering that many patients are at risk for cardiac ischemia and arrhythmia during these procedures (29, 30, 35, 41), attenuating HR, BP, and RPP in response to TSO has received much attention (1, 8, 16, 47, 53, 55). However, the basic physiology is unclear, because patients in these cited experiments were premedicated, sedated, and/or under general anesthesia when TSO was applied.

A number of physiological stressors (i.e., orthostasis, exercise, hypoxemia) elicit sympathoexcitation, which serves to redistribute blood flow and maintain perfusion to critical organs. Studies in anesthetized patients have shown that muscle sympathetic nerve activity (MSNA) and BP increase rapidly and robustly during intubation and laryngoscopy (9, 11, 43), providing evidence that TSO elicits vasoconstriction within skeletal muscle. Another vascular bed—the kidney—receives ~20% of cardiac output at rest, and alpha-adrenergic renal vasoconstriction occurs in responses to physiological stress (5, 27, 28, 40). In dogs, renal sympathetic nerve activity and BP both increase during intubation (42), but the renal vascular responses to TSO in humans are unknown. This may be clinically relevant, considering that perioperative renal failure (seemingly due to reductions in renal blood flow) is not uncommon.

The purpose of this study was to characterize the integrated neurovascular responses to TSO in conscious, unmedicated humans (Experiments 1) and to block this response using local anesthesia of the upper airway (Experiments 2 and 3). In these experiments, we tested two separate but related hypotheses. First, TSO will cause greater changes in BP, HR, MSNA, and renal vascular resistance (RVR) compared with tactile stimulation of the hard palate (Sham). Second, inhalation of 4% topical lidocaine prior to (pre) TSO will block the gag reflex and attenuate increases in HR and BP. Herein, we demonstrate that stimulation of mechanically sensitive afferents in the oropharynx elicits an increase in MSNA, along with acute hypertension, tachycardia, and renal vasoconstriction; these effects can be abolished with local anesthesia of the upper airway.

METHODS

All study protocols were approved in advance by the Institutional Review Board of the Penn State Milton S. Hershey Medical Center and conformed to the Declaration of Helsinki. A total of 23 healthy, unmedicated, young (range 22–33 years) subjects volunteered to participate and provided written, informed consent. Due to the unpleasant nature of gag-reflex testing, we attempted to recruit different groups of subjects for each study. In total, two subjects participated in both Experiments 1 and 2, and two different subjects participated in both Experiments 2 and 3. The sample size for Experiment 1 was determined based on power analyses of MSNA and RVR in response to TSO compared with Sham (i.e., power > 80% for each measure).

Experiment 1: Effect of TSO on MSNA and RVR

Thirteen individuals (eight men, five women; 25 ± 1 years, 1.76 ± 0.03 m, 74.3 ± 4.0 kg, 24.0 ± 0.7 kg/m²) participated in Experiment 1. All experiments were conducted in the supine posture. Subjects
pressed medical air was supplied at 4–8 l/min, and subjects breathed
consistently with previous lidocaine-inhalation studies (4, 22, 54), com-
prising afferents that initiate sympathetic reflexes (10, 13, 36). Con-
combiner, has been administered locally and systemically to block

Fig. 1. Representative beat-by-beat recordings of muscle sympathetic nerve activity (MSNA), blood pressure (BP), heart rate (HR), and respiratory movement (Resp) in the same subject. The 1st arrow denotes opening the mouth, and the 2nd arrow denotes tactile stimulation of the oropharynx (TSO; eliciting the gag reflex). Within 1–2 cardiac cycles after TSO, there was a rapid increase in MSNA, BP, and HR. Trials 1 and 2 were separated by 5 min.

MSNA

Trial 1

140

120

100

80

60

40

0

10 seconds

BP

Trial 2

140

120

100

80

60

40

0

10 seconds

HR

Resp

Inspire

10 seconds

Expiration

10 seconds

Expiration

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were first instrumented with a finger BP cuff (Finometer; Finapres
Medical Systems, Amsterdam, The Netherlands) to monitor beat-by-
beat BP, a three-lead ECG (Cardiopac5; GE Healthcare, Waukesha,
WI) to measure HR, and a custom-designed pneumotrace to detect
respiratory movement. Baseline recordings of MSNA (n = 6) or RVR
(n = 8) were obtained for 5 min. For testing, the subject was
instructed to “close the eyes, open the mouth, and stick out the
tongue.” A registered nurse (same investigator for all trials) then
applied stimulation to either the oropharynx (TSO; eliciting the gag
reflex) or the hard palate of the mouth (Sham; no gag reflex) in a
counterbalanced fashion. TSO was conducted using a wooden tongue
blade or cotton swab, per standard clinical practice (7, 22), and was of
very short duration (i.e., <2 s). Clinical intubation and laryngoscopy
are typically 10–30 s in duration (21, 30, 47) and elicit the same
airway defense reflex as TSO. The subject was not aware which
stimulus was about to occur. A recovery period followed for 3–5 min,
and once parameters returned to baseline, the opposite stimulus
was applied. The goal was to administer one trial of TSO and one trial of
Sham per subject, but additional trials were sometimes conducted if
an event occurred. The goal was to administer one trial of TSO and one trial of
Sham per subject, but additional trials were sometimes conducted if
coughing (n = 2 trials) or limb movement (n = 3) occurred, since
both of these cause measurement error. Although not the primary
purpose of this study, during some MSNA trials, TSO was applied
twice to explore whether hemodynamic and neural responses were
attenuated in the second trial (Fig. 1). At the end of the experiment,
subjects performed a maximal voluntary end-expiratory apnea to
ensure that the quality of the MSNA recording was consistent from
beginning to end.

Experiment 2: Effect of Lidocaine Inhalation on HR and BP during
TSO

Ten individuals (six men, four women; 26 ± 1 years, 1.78 ± 0.03 m,
79.0 ± 5.2 kg, 24.8 ± 1.0 kg/m²) participated in Experiment 2. After
baseline measurements of MSNA and BP, TSO was conducted in the
supine posture. Next, the participants assumed the seated, upright
posture, and the TSO procedures were repeated within 2–3 min.

Cold Pressor Test

On a separate day, four subjects underwent additional studies,
which sought to determine whether the aforementioned lidocaine-
inhalation protocol had a systemic effect (10, 26). After instrumenta-
tion and baseline measurements in the supine posture, subjects under-
went the cold pressor test (CPT; hand into 1°C water) for 90 s. This
procedure activates the sympathetic nervous system (23) and increase
RPP, an index of myocardial oxygen demand (33). Immediately after
the CPT, subjects were asked to rate their hand thermal sensation
(where 0 = neutral/no sensation of cold, and 11 = unbearable cold)
(15) and hand pain (where 0 = no pain, and 10 = unbearable pain)
(18). Lidocaine inhalation was then conducted identically to the
procedures listed above, and the CPT was repeated.

Measurements. During Experiment 1, multifiber recordings of
MSNA were obtained, with a tungsten microelectrode (FHC, Bow-
doin, ME) inserted in the peroneal nerve of a leg. A reference
electrode was placed subcutaneously, 2–3 cm from the recording
electrode, which was adjusted until a site was found in which muscle
sympathetic bursts were clearly identified using previously estab-
lished criteria (52). Briefly, MSNA was distinguished from other
nerve signals when there was increased burst activity in response to
maximal voluntary end-expiratory apnea (to activate arterial chemore-
flex) (17) and/or passive muscle stretch but not with skin stroking of
the innervated area, rapid inspiration, or arousal stimuli (52). The
nerve signal was amplified, band-pass filtered with a bandwidth of
500–5,000 Hz, and integrated with a time constant of 0.1 s (Model
662C-3; The University of Iowa Bioengineering, Iowa City, IA). The
nerve signal was also routed to a loudspeaker and a computer for monitoring throughout the study.

During Experiment 1, transabdominal Doppler ultrasound (HDI 5000; ATL Ultrasound, Bothell, WA) was used to measure renal blood flow velocity (RBV), as described previously (28, 40). The artery was scanned with a curved array C5-2 transducer.

Prior to all experiments, resting measures of systolic BP (SBP) and diastolic BP (DBP) were obtained via automated sphygmomanometry of the brachial artery (SureSigns VS3; Philips Healthcare, Andover, MA) in triplicate. Beat-by-beat BP, HR, MSNA, and respiratory movement were sampled at 200 Hz by a data acquisition system (PowerLab; ADInstruments, Colorado Springs, CO).

Data collection and statistical analysis. Beat-by-beat physiological parameters were analyzed offline using LabChart 7 (ADInstruments). The variables of interest included HR, SBP, DBP, mean arterial pressure (MAP), MSNA burst rate, and MSNA total activity. RPP was calculated as HR × SBP and was considered to be the primary outcome measure for two reasons. First, it was collected in all experiments, allowing consistent comparison (i.e., MSNA and RVR were only collected in Experiment 1). Second, it is a primary determinant of myocardial oxygen consumption that is relevant during upper-airway clinical procedures (29, 41). RBV was measured offline using Prosolv 3.0, and RVR was calculated as MAP/RBV; an increase in RVR is considered to be renal vasoconstriction (5, 27, 28, 40).

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RESULTS

Experiment 1: Effect of TSO on MSNA and RVR

Resting baseline values for MAP (77 ± 1 mmHg), HR (59 ± 3 beats/min), MSNA burst rate (11 ± 2 burst/min), and RBV (69 ± 8 cm/s) were within normal levels for young, healthy subjects. In all subjects, TSO elicited the gag reflex, which was audible and confirmed by the subject and the investigators. Qualitative data (Fig. 1) indicate that TSO increased MSNA total activity as well as HR and MAP. Indeed, the ∆MAP (12 ± 3 vs. 6 ± 1 mmHg), ∆HR (10 ± 3 vs. 3 ± 3 beats/min), and ∆RBV (−15 ± 4 vs. 1 ± 1 cm/s) were greater following TSO compared with Sham (i.e., without eliciting the gag reflex). As shown in Fig. 2, the ∆RPP (P = 0.042), ∆MSNA (P = 0.040), and ∆RVR (P = 0.010) were significantly greater with TSO compared with Sham. Relative to baseline, Sham stimulation caused significant increases in RPP (P = 0.048) and MAP (P = 0.005) but not HR (P = 0.366), RBV (P = 0.691), or RVR (P = 0.304). Additionally, beat-by-beat analysis of MSNA revealed that in the first cardiac cycle following TSO, six of six subjects had a MSNA burst compared with zero of six subjects following Sham. In the second cardiac cycle following TSO, five of six subjects had a MSNA burst compared with two of six subjects following Sham.

Fig. 2. Changes in rate-pressure product (∆RPP; top, n = 13), ∆MSNA (middle, n = 6), and renal vascular resistance index (∆RVR; bottom, n = 8) in response to TSO (black bars) and tactile stimulation of the hard palate (Sham; white bars) during Experiment 1. Data are means ± SE. *P < 0.05 between TSO and Sham; †P < 0.05 vs. respective baseline.
Experiment 2: Effect of Lidocaine Inhalation on HR and BP during TSO

Inhalation of lidocaine did not affect supine resting MAP (pre: 77 ± 2; after (post): 78 ± 2 mmHg) or HR (pre: 58 ± 3; post: 56 ± 3 beats/min). During and after lidocaine inhalation, subjects reported a lack of sensation of the tongue and throat, and some individuals complained of hoarseness. Lidocaine blocked the gag reflex in all subjects (Fig. 3). The RPP response to TSO was reduced drastically following lidocaine (Fig. 4). Opening the mouth and sticking out the tongue elicited a modest increase in RPP, but this was not different between treatments (P = 0.122). Lidocaine reduced the peak RPP response to TSO (P = 0.004), as well as the RPP response within the first five cardiac cycles of recovery (P = 0.014). As expected, the peak SBP (pre: 25 ± 4 vs. post: 7 ± 2 mmHg), peak HR (pre: 12 ± 3 vs. post: 2 ± 2 beats/min), and peak MAP (pre: 16 ± 2 vs. post: 5 ± 2 mmHg) response to TSO were also attenuated following lidocaine inhalation (all P < 0.01). As shown in Fig. 5, there was a strong, positive relationship between TSO-induced RPP responses within the same day (Cronbach’s α = 0.743, P = 0.036).

Experiment 3: Effect of Lidocaine Inhalation on HR and BP during CPT

To determine whether inhaled lidocaine would attenuate hemodynamic and perceptual responses to the CPT, paired t-tests were conducted. The RPP was not different before (2,347 ± 873 mmHg × beats/min) and after (2,559 ± 629 mmHg × beats/min) lidocaine (P = 0.591). Similarly, the MAP (pre: 30 ± 6; post: 26 ± 8 mmHg) and the HR (pre: 4 ± 6; post: 6 ± 4 beats/min) were also not statistically different. Furthermore, hand thermal sensation (pre: 9 ± 1; post: 8 ± 1) and hand pain (pre: 7 ± 1; post: 7 ± 1) were comparable before and after lidocaine, indicating that lidocaine inhalation did not cause systemic sympathoinhibition.
DISCUSSION

Previous research has demonstrated that laryngoscopy and upper-airway intubation cause rapid increases in HR, BP, MSNA, and plasma catecholamines (1, 8, 16, 21, 38, 53). These changes are likely to be reflex mediated, because they can be blunted or abolished by using different pharmacotherapies (i.e., ganglionic blockade, beta blockade) (46, 51). However, these cited studies were performed in anesthetized patients, and it is known that general anesthesia can affect reflex pathways (44). For this reason, we sought to document how conscious, unmedicated humans would respond to TSO. The primary, novel findings are: 1) TSO elicited reflex increases in MSNA, MAP, HR, and RVR compared with Sham; 2) inhalation of 4% topical lidocaine prior to TSO blocked the gag reflex and attenuated increases in HR and BP; and 3) inhalation of 4% topical lidocaine did not affect the hemodynamic or perceptual responses to the CPT. To our knowledge, this is the first report of TSO eliciting sympathoexcitation in conscious, unmedicated humans.

The afferent arm of the gag reflex is comprised of the glossopharyngeal nerve (cranial nerve IX) and the laryngeal branch of the vagus nerve (cranial nerve X). These nerves relay sensory information from the pharynx, tonsils, epiglottis, and base of the tongue to the medulla (31, 57). The efferent arm of the gag reflex includes the vagus nerve (cranial nerve X) and results in contraction of the posterior oral and pharyngeal musculature, thus preventing foreign bodies from entering the trachea (25). On the other hand, the hard palate is innervated by the maxillary branch of the trigeminal nerve, also called the nasopalatine nerve. The application of pressure to this nerve does not typically elicit the gag reflex. Therefore, Sham was considered to be the control in Experiment 1.

As noted in Fig. 1, TSO elicited rapid increases in HR and MAP, as detected by the Finometer device. Previous experiments using an arterial catheter have shown that upper-airway stimulation increases SBP by 30–60 mmHg and DBP by 10–30 mmHg, while causing a modest tachycardia (ΔHR 15–30 beats/min) (1, 8, 16, 21, 53). Our data are comparable, considering that TSO was of shorter duration than laryngoscopy and intubation (but elicited the same airway defense reflex). The nucleus tractus solitarius (NTS) receives inputs from cranial nerves IX and X, as well as the carotid chemoreceptors and baroreceptors that are well recognized to participate in circulatory homeostasis (34). We postulate that in response to TSO, the NTS activates both pharyngeal muscle contraction and sympathetic outflow to skeletal muscle and the kidney in a parallel manner.

Ebert et al. (9) were the first to demonstrate that MSNA increases during laryngoscopy and intubation in thiopental-anesthetized humans. This finding was corroborated in subsequent experiments that employed a pharyngeal suction stimulus (43). MSNA is a direct measure of sympathetic outflow to blood vessels within skeletal muscle and participates in reflex adjustments in BP. In the current study using conscious, unmedicated humans, we demonstrate that MSNA increased, and RVR also increased within one to two cardiac cycles of TSO (Fig. 2). Based on the temporal response, we hypothesize that TSO increased renal sympathetic nerve activity, thereby evoking renal vasoconstriction. A study in dogs documented that renal sympathetic nerve activity increased during both intubation and extubation (42). In our experiment, the magnitude of increase in RVR following TSO within one to two cardiac cycles was comparable with that seen after ~60 s of isometric handgrip (40% maximal contraction) (27). During Sham stimulation, there was no increase in MSNA and no renal vasoconstriction, but we did document a small increase in RPP and MAP, attributable to participant anxiety and/or opening the mouth and protruding the tongue.

Lidocaine inhalation has been used in clinical anesthesia to lessen the circulatory responses resulting from intubation and laryngoscopy (4, 22, 54). Fundamentally, lidocaine blocks sodium channels and prevents afferent nerves from generating an action potential. Because of this, lidocaine has also been used in research studies to understand how peripheral afferents (e.g., within muscle or blood vessel) affect BP homeostasis (6, 10, 36). With the use of lidocaine inhalation to block sensory afferents in the oropharynx (presumably cranial nerves IX and X), we have demonstrated that TSO elicits a sympathoexcitatory reflex (in addition to the well-characterized gag reflex) in conscious, healthy humans. It is important to note that our lidocaine-inhalation protocol did not affect the physiological or perceptual responses to the CPT (Experiment 3). Regional administration of lidocaine into a limb is known to block the presensor and pain response to the CPT (14). Taken together, these data provide strong experimental evidence that inhalation of lidocaine is exerting a local, not systemic, anesthetic effect. Lidocaine inhalation is simple, well tolerated, and does not affect resting hemodynamics; these factors make it ideal to use in future research studies.

Experiment 2 required that each subject experience the gag reflex twice within the same day (i.e., before lidocaine was given and after it had worn off). Although not the intended purpose of our study, this experimental design allowed for test-retest reliability to be determined. As displayed in Fig. 5, the ΔRPP in response to TSO was similar within the same day, such that higher RPP responses to the first TSO were related to higher responses to the second TSO. This supports the previously established concept that individual differences in cardiovascular reactivity to upper-airway stimulation exist (19, 35).

On a physiological level, the current data indicate that short-duration tactile stimulation of the upper airway elevates sympathetic outflow to the kidney and skeletal muscle. These data in conscious human subjects extend upon prior publications, suggesting that the oropharynx is a sensory organ capable of initiating sympathetic reflexes (51, 56). On a clinical level, thousands of intubations are performed each day throughout the world, and many of these patients have underlying cardiovascular disease (29, 35). The sympathetic nervous system likely contributes to cardiac and renal complications observed in these patients. Specifically, longer intubation durations are linked with increased risk of myocardial infarction (2), and electrocardiographic abnormalities are most common during intubation (20, 37, 53). Clinical observations have also shown that postoperative renal failure (seemingly due to reductions in blood flow) is linked to increased mortality (49). Furthermore, ventricular tachycardia and cerebrovascular accident have been documented in one kidney-transplant patient following endotracheal intubation (12). During upper-airway procedures, it is desirable to prevent a large increase in RPP (i.e., with afferent blockade) rather than to give vasoactive medication or additional inhalation anesthetics (i.e., with eff-
different blockade, which would not normally be used) to combat an increased RPP. Whether oropharyngeal afferents also contribute to adverse outcomes resulting from other airway stimuli (e.g., cold-air inhalation, cigarette smoking, prolonged ventilator use) is yet to be determined.

The current experiments used a physiological approach to understand how TSO impacts cardiovascular homeostasis in young, healthy humans. As such, extrapolation to healthy, older adults or patient populations must be done with caution. It should be noted that TSO was of short duration in the current study, leading to similar yet smaller responses compared with previous studies (1, 8, 16, 21, 53). For ethical reasons, we chose not to intubate our conscious, healthy subjects and instead, focused on the physiological mechanisms underlying TSO. It is also possible that stimulation of the larynx or lower respiratory tract may elicit a different response, and this response may be modulated by the type of stimulus (e.g., pressure, temperature, irritation) (32, 33, 39). Additional studies are warranted to unravel how TSO impacts human physiology in both healthy and diseased states.

Clinical Implications

In the current study, TSO elicited acute increases in MSNA, MAP, HR, and RVR in conscious humans, and this effect could be blocked with local anesthesia of the upper airway. These data provide evidence that airway defense mechanisms (e.g., gag reflex) engage the sympathetic nervous system and elevate HR and BP. We speculate that a sensitized upper airway (e.g., gag reflex) engage the sympathetic nervous system and elevate HR and BP. We speculate that a sensitized upper airway due to allergies, cigarette smoking, or gingivitis, places these data provide evidence that airway defense mechanisms (e.g., gag reflex) engage the sympathetic nervous system and elevate HR and BP. We speculate that a sensitized upper airway due to allergies, cigarette smoking, or gingivitis, places as such, extrapolation to healthy, older adults or patient populations must be done with caution. It should be noted that TSO was of short duration in the current study, leading to similar yet smaller responses compared with previous studies (1, 8, 16, 21, 53). For ethical reasons, we chose not to intubate our conscious, healthy subjects and instead, focused on the physiological mechanisms underlying TSO. It is also possible that stimulation of the larynx or lower respiratory tract may elicit a different response, and this response may be modulated by the type of stimulus (e.g., pressure, temperature, irritation) (32, 33, 39). Additional studies are warranted to unravel how TSO impacts human physiology in both healthy and diseased states.

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author.

AUTHOR CONTRIBUTIONS


REFERENCES


