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### Use of an Anatomical Scalar to Control for Sex-Based Size Differences in Measures of Hyoid Excursion During Swallowing

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#### Abstract

**Purpose**—Traditional methods for measuring hyoid excursion from dynamic videofluoroscopy recordings involve calculating changes in position in absolute units (mm). This method shows a high degree of variability across studies but agreement that greater hyoid excursion occurs in men than women. Given that men are typically taller than women, we hypothesized that controlling for participant size might neutralize apparent sex-based differences in hyoid excursion.

**Methods**—We measured hyoid excursion in 20 young (<45) healthy volunteers (10 male), stratified by height, in a tightly controlled videofluoroscopic protocol.

**Results**—We identified an anatomical scalar (C2-4 length), visible on the videofluoroscopic image, correlated with participant height. This scalar differed significantly between men and women. By incorporating the anatomical scalar as a continuous covariate in repeated measures mixed model ANOVAs of hyoid excursion, apparent sex-based differences were neutralized. Transforming measures of hyoid excursion into anatomically scaled units, achieved the same result, reducing variation attributable to sex-based differences in participant size.

**Conclusions**—Hyoid excursion during swallowing is dependent on a person's size. If measurements do not control for this source of variation, apparent sex differences in hyoid excursion are seen.

#### Keywords

Swallowing; deglutition; dysphagia; hyoid; variation; normalization; videofluoroscopy

Eating and drinking are not only important life-sustaining functions, but are also central to social activity and quality-of-life. The act of safe swallowing is a complex neuromuscular process involving a sequence of bilateral inhibition and activation of multiple muscles in the lips, tongue, palate, larynx, pharynx and esophagus (Ertekin & Aydogdu, 2003). A disruption in swallowing function is called dysphagia, and can occur secondary to a variety of neurological, structural, and degenerative causes. The assessment of dysphagia frequently

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involves videofluoroscopy (VF), a radiographic imaging technique that allows real-time dynamic visualization of swallowing physiology. Kinematic and temporal measurements can be extracted from VF recordings, and are used to gain insight regarding the underlying reasons for and nature of dysphagia.

During the pharyngeal phase of swallowing, the pharynx must reconfigure to protect the airway while food and/or liquid moves from the mouth to the esophagus. This critical event, which prevents material from being aspirated into the lungs, is achieved in part by biomechanical displacement of the hyoid bone in a superior-anterior trajectory. Muscular connections between the hyoid bone, the larynx and the pharynx enable this movement to contribute to closure of the laryngeal vestibule and down-folding of the epiglottis for airway protection (Logemann et al., 1992), and also to opening of the upper esophageal sphincter (UES) (Jacob, Kahrilas, Logemann, Shah, & Ha, 1989), allowing material to exit the pharynx. This manuscript details a technique for the accurate measurement of hyoid excursion using a dataset of VF swallowing recordings from 20 healthy young volunteers who swallowed a variety of volumes of ultra-thin liquid barium. The technique is applied to a healthy data set to clarify whether or not differences in the extent of hyoid excursion should be expected between men and women.

#### Literature Review

The current standard of clinical practice with respect to interpreting VF recordings involves perceptual judgment of the adequacy of hyoid movement. Perceived reductions in hyoid movement, either in the superior or anterior direction, have been cited as contributing to, or explaining, swallowing dysfunction (see for example, Perlman, Booth, & Grayhack, 1994). However, perceptual judgments of hyoid excursion are known have poor reliability (Perlman, VanDaele, & Otterbacher, 1995). An alternative to perceptual judgment is to take quantitative measures of hyoid excursion using image analysis tools, however this is rarely done in clinical VF analysis. There are several steps involved in image-based hyoid measurement. These include defining:

- **a.** a measurement system including an origin, which will remain stable despite head movement (usually a point on the cervical spine);
- **b.** a Cartesian coordinate system relative to the origin (usually by setting the vertical axis of displacement in relation to the cervical spine and deriving the horizontal axis of displacement perpendicular to the vertical axis); and
- c. a measurement scale (typically absolute distance in millimeters, derived using an externally-placed scalar of known dimensions, such as a coin) (Dantas et al., 1990; Dodds et al., 1988; Kang et al., 2010; Kim, Han, & Kwon, 2010; Logemann, Pauloski, Rademaker, & Kahrilas, 2002; Paik et al., 2008; Perlman et al., 1995; Zu, Yang, & Perlman, 2011).

When these contextual definitions have been established, pixel measurements of hyoid position or displacements (i.e. the difference between position measures on two selected frames) can be made. Nevertheless, even when this level of methodological rigor in quantitative measurement has been employed, the literature suggests that hyoid movement

in swallowing is a highly variable phenomenon, even in healthy individuals. A recent metaanalysis of 13 studies that report quantitative measures of hyoid excursion in healthy participants, found that reported means for anterior hyoid excursion ranged from 7.6 mm to 18.0 mm, while mean superior excursion ranged from 5.8 mm to 25.0 mm (Molfenter & Steele, 2011). In the context of such large variation across studies, it is important to explore ways to limit any variability that arises from methodological factors rather than true patient variation. In their review, Molfenter and Steele (2011) identified several controllable sources of variation that might account for previous differences across studies, including:

- sample size (small samples are susceptible to display greater variability);
- representativeness of the data (repeated sampling of behavior will reduce variability);
- the method used to capture hyoid excursion (frame-by-frame marking; two-frame comparisons of rest and peak hyoid position; or single measures of peak position (Humbert et al., 2005));
- the definition of rest/minimum position (pre- versus post-swallow) (Ishida, Palmer, & Hiiemae, 2002);
- specification of the coordinate system (origin, axes, units);
- separate reporting of anterior and superior displacements (e.g., Dantas et al., 1990; Dodds et al., 1988; Kim & McCullough, 2008; Perlman et al., 1995) versus reporting of the vector (hypotenuse) of movement (e.g., Leonard, Kendall, McKenzie, Gonçalves, & Walker, 2000);
- measurement error (poor intra- or inter-rater reliability across repeated measurement);
- stimulus characteristics (barium concentration, viscosity, volume);
- protocol decisions (use of cued versus spontaneous swallows); and
- participant factors including sex, age, and anthropometric features (e.g., height, weight or other measures of size).

In this study, we sought to measure hyoid excursion in healthy swallowing, using strict control to constrain methodological sources of variability. Our primary objective was to confirm whether hyoid excursion differs between men and women. It is well accepted that the anatomical position of the hyoid and larynx sits lower at rest in post-pubertal men than in women. It therefore seems reasonable to expect that women might show reduced extent of hyoid movement compared to men, however prior studies of hyoid movement differ on this point. Logemann and colleagues (2002) studied a sample of 16 healthy women who showed significantly reduced vertical hyoid excursion on thin liquid barium swallows compared to data from a previous study in healthy male participants (Logemann et al., 2000). By contrast, Kim and McCullough (2008) failed to find significant sex differences in hyoid movement in a sample of 40 healthy participants, as did Ishida et al. (2002) in a sample of 12 healthy participants. Other studies have studied healthy participants of only one sex (e.g.,

Perlman et al., 1995) or have not analyzed participant sex as a factor (Paik et al., 2008; Bingjie, Tong, Xinting, Jianmin, & Guijun, 2010; Kim et al., 2010).

Our interest in this question was further stimulated by several studies in which it has been suggested that measures of hyoid excursion should be normalized to account for the variation arising from differences in length of the pharynx or neck (which we will call sizeof-the-system). Leonard et al., (2000) reported statistically significant, albeit weak positive correlations between participant height and hyoid excursion (r=0.37 for 20ml boluses). Perlman and colleagues (1995) proposed reporting measures of hyoid displacement in 'cervical units' (a participant-specific measure capturing the length of C1 to C5) and showed that this approach was able to capture reductions in hyoid excursion in a manner similar to millimeter measurement. Others have reported normalized measures of swallowing biomechanics using anatomical scalars (Kahrilas, Lin, Rademaker, & Logemann, 1997; Potratz, Dengel, Robbins, & Brooks, 1993). In a hybrid approach, Kim and McCullough (2008) used the C3 vertebra as an anatomical scalar, but assigned it a fixed assumed height value of 15 mm in their derivation of measures of hyoid movement. Although this is a valid method of controlling for differences in magnification across images within participant, it also has the effect of neutralizing any true differences between participants in C3 length or measures derived using that scalar reference. Logemann and colleagues have adopted the practice of using a measure of cervical spine length as a covariate in their analyses of hyoid displacement (Logemann et al., 2000; Logemann et al., 2002). The differences in method between the Logemann studies (Logemann et al., 2000; Logemann et al., 2002) and the Kim and McCullough (2008) approach may well be substantial enough to lead to different results regarding the presence of significant sex-based differences in hyoid excursion.

In this study, we sought to clarify the influence of participant size-of-the-system on measures of hyoid excursion and on sex differences in hyoid excursion in healthy swallowing. The general trend in the population is that men are taller than women. As mentioned above, measures of full body height are known to be correlated with hyoid movement (Leonard et al., 2000), however we believe that a size measure taken from the region of the head and neck is likely to be more appropriate to use in the context of questions about swallowing, given that it is theoretically possible for two people to have the same height but different leg, trunk and neck lengths. Furthermore, if size-of-the-system is confirmed to be relevant for future measures of hyoid movement in swallowing assessment, a measure of size that can be taken directly from a fluoroscopic image would lend itself more easily to clinical uptake. We hypothesized that sex differences in hyoid movement would pattern according to the sex distribution of size-of-the-system. Conversely, we predicted that measures of hyoid movement that are normalized using an anatomical scalar measure to capture size-of-the-system would not differ significantly between male and female participants. If confirmed, these results would support the conclusion that measures of the adequacy of hyoid excursion in swallowing should take size-of-the-system into account. To explore these hypotheses, we developed a list of 6 research questions:

Q1 Of a set of 13 internal anatomical scalar candidates, visible in the VF image, which one has the best correlation with participant height (our criterion for being selected as a scalar measure to represent the size-of-the-system)?

- Q2 Does the selected size-of-the-system scalar vary significantly by sex?
- Q3 Does hyoid excursion (measured in millimeters) vary with the size-of-thesystem?
- **Q4** When tested in a mixed model repeated measures ANOVA, do millimeter measures of hyoid excursion vary significantly by sex and/or bolus volume?
- **Q5** Does adding a size-of-the-system covariate (from Q1) to the model in Q4 alter the result regarding sex differences in hyoid excursion?
- **Q6** Does changing the metric in Q4 from absolute units to scaled units (via use of the internal anatomical scalar identified in Q1) agree with the results of Q5?

#### Methods

The methodological procedures pertaining to this dataset were carefully chosen to control for potential sources of variability, as identified in Molfenter and Steele (2011) and have been reported in detail elsewhere (Molfenter & Steele, 2012). This study was reviewed and approved by the institution's research ethics board.

#### **Participants**

Twenty (10 male) healthy young volunteers (under 45, mean: 31.5 years, standard deviation (SD): 5.7 years) were recruited to participate in a VF protocol. Participant recruitment was stratified by height to span a range between the national reported mean lower and upper height quartiles for adults by sex (Shields, Gorber, & Tremblay, 2009). Height distribution by sex for the study sample is displayed in Figure 1.

#### VF Stimuli

Each participant performed a series of 15 swallowing tasks, 9 of which were included in this analysis: 3 swallows each of 5ml, 10ml and 20ml liquid barium 22% w/v suspension (Bracco Polibar suspension diluted with water). Suspensions with this concentration of barium are called 'ultra-thin' in the dysphagia literature (Fink & Ross, 2009). A protocol of three repetitions per volume condition was used to ensure representative sampling of intraparticipant variability, while keeping radiation exposure time to a minimum (Lof & Robbins, 1990). Sample volumes were measured using a pipette and placed in 30ml capacity plastic cups. Based on pilot testing, a sample 1ml greater than the target volume for swallowing was pipetted into each cup to allow a margin for residual material likely to remain in the cup after each sip. Cups were weighed before and after drinking so that the exact volume consumed could be determined. In the event that a participant used piecemeal deglutition for a particular bolus (i.e., division of a sip into two or more swallows by partitioning the bolus in the mouth), the data for that bolus were excluded from the analysis, due to our inability to accurately measure the volume of the bolus ingested in each subswallow (1 instance at 5ml and 6 instances at 20ml). Table 1 lists means and 95% confidence intervals for the average volume swallowed by target volume. These results confirm that despite adding extra barium to each cup, there was a tendency for a portion of each administered bolus to remain behind as residual in the cup. All statistical analyses

#### **VF Procedure**

All VFs were conducted using a Toshiba Ultimax (Toshiba America Medical Systems, Inc., Tustin, CA) in lateral view at full resolution (30 pulses per second) and captured and recorded on a Digital Swallowing Workstation (KayPentax, Lincoln Park, NJ) at 30 frames per second. A coin of known diameter (19.05 mm) was placed over the left mastoid process of each participant using medical tape to facilitate the later conversion of pixel-based measures of structural movement into millimeters. Boluses were arranged and presented in blocks of three cups of the same volume on a table within easy reach of the participant. The order of bolus volume block was randomized. Once the VF was turned on, the participant was instructed to self-feed and swallow the liquid from each block, one cup at a time, at a spontaneous, comfortable pace. Self-feeding and spontaneous swallows were used to avoid changes in swallow timing associated with cued swallowing (Daniels, Schroeder, DeGeorge, Corey, & Rosenbek, 2007; Nagy et al., 2013). The VF was turned off after the hyoid returned to rest following the final bolus of each three-cup sequence. The average total VF exposure time (for the entire 15 task sequence) was 1.75 minutes (SD: +/-0.31 minutes).

#### VF Post-Processing and Analysis

The positions of the following structures were marked in every frame of each swallow: the anterior inferior corner of the C4 vertebra (origin); the anterior inferior corner of the C2 vertebra (Y vector); and the anterior inferior corner of the hyoid. The position of the hyoid in each frame was calculated based on its XY position relative to the origin (C4) with the Yaxis defined parallel to the spine. The positional data were then scaled using two methods: 1) in absolute units (mm) using the external coin scalar (19.05mm) and 2) in units scaled using an internal anatomical scalar (%C2-4 spine length). Justification for this choice of particular scalar comes from the results of Q1 below. Both sets of positional data were exported to an Excel file (Microsoft) with an embedded macro that was devised to find the maximum and minimum values (in both the X and Y planes) between two user-defined frames of interest. The 'start' frame was designated as 10 frames prior to the sudden upward/forward burst of hyoid movement associated with a swallow and the 'end' frame was defined as 10 frames after the epiglottis returned to a vertical position. Each swallow yielded four data points for analysis (minimum X position, maximum X position, minimum Y position, and maximum Y position). These data points were then used to derive four parameters capturing hyoid excursion. Three of these parameters were displacement measures (i.e., the difference between minimum and maximum position):

- anterior displacement (i.e., maximum X position minus minimum X position);
- superior displacement (i.e., maximum Y position minus minimum Y position); and
- hypotenuse displacement (calculated as the square root of the sum of the squared anterior and superior displacement measures).

We also derived a single point measure of maximum XY hyoid position relative to the C4 origin (calculated as the square root of the sum of the squared maximum X and Y position data point measures). These parameters are illustrated in Figure 2.

#### **Reliability Measures**

Inter- and intra-rater reliability were measured for each hyoid excursion parameter using a random selection of 10% of the swallows in the dataset, using two-way mixed intra-class correlation coefficients (ICC) for consistency. Results (Table 2) for superior displacement, hypotenuse displacement and maximum XY position all demonstrated 'excellent' reliability, while scores for anterior displacement achieved only 'fair to good' reliability (i.e., 0.40– 0.75) (Fleiss, 1986). One possible explanation for the lower reliability observed for anterior displacement is that head movement causes anterior-posterior movement of both the hyoid and the origin of the measurement system, but has minimal effect on the superior-inferior position of the origin. Variation in anterior-posterior position of the origin increases the opportunity for measurement error in the anterior-posterior plane between and across raters.

#### **Statistical Analysis**

All statistical analyses were conducted using IBM SPSS Statistics Version 20. Two-tailed *p*-values < 0.05 were considered statistically significant. For the mixed model analyses of variance, a compound symmetry model structure was determined to have the best fit with the data. When main effects were significant, post hoc pairwise comparisons were conducted with Sidak adjustment for multiple comparisons. Effect sizes for pairwise comparisons were calculated using Cohen's *d* and values of 0.2–0.5 were considered to show small effects, 0.5–0.8 to show medium effects and values > 0.8 to show large effects (Kotrlik & Williams, 2003).

**Q1.** To determine which internal anatomical scalar is the best candidate to represent the size-of-the-system, 13 potential internal scalars (details in Figure 4) were measured in pixels using ImageJ software (National Institutes of Health, Bethesda, MD) on a single pre-swallow frame. The length of each scalar candidate was transformed from pixels to millimeters using the externally placed coin scalar as a reference. Pearson's correlations were used to examine the relationship between the various internal anatomical scalars (mm) and participant height (cm). It was decided a priori that the scalar displaying the highest correlation with height would be used to represent 'size-of-the-system' in the subsequent research questions.

**Q2.** A one-way analysis of variance (ANOVA) was run to explore the influence of sex on the size-of-the-system.

**Q3.** To determine whether hyoid excursion (in mm) varies significantly according to the size-of-the system, mixed model repeated measures ANOVAs for each hyoid parameter were run with a continuous predictor of size-of-the-system, a within participant factor of bolus volume, and a repeated factor of trial-within-bolus-volume. Pearson's correlation coefficients between size-of-the-system and hyoid excursion measures were also calculated.

**Q4.** To test the association between sex, bolus volume and hyoid excursion (in mm), mixed model repeated measures ANOVAs for each hyoid measurement parameter were run with a between participants factor of sex, a within participant factor of bolus volume, and a repeated factor of trial-within-bolus-volume.

**Q5.** In order to test whether size-of-the-system influences alters the significance of differences in hyoid excursion (in mm) by sex, the size-of-the-system scalar (from Q1) was added as a covariate to the Q4 models.

**Q6.** In order to test whether scaling hyoid movement using an internal anatomical scalar (Q1) can account for the variation attributable to size-of-the-system, the analyses from Q4 were repeated with the metric of hyoid excursion transformed from absolute units to scaled units.

#### Results

**Q1.** Correlations with height were explored for eleven spine-based and two non-spine-based scalars (see Figure 4). Pearson correlations ranged from 0.46 to 0.83 and are listed in Figure 4. The length of the C2-4 unit (measured from the anterior inferior corner of C2 to the anterior inferior corner of C4) displayed the strongest correlation with true participant height (r=0.83, p<0.001) and was therefore chosen to represent size-of-the-system in subsequent analyses. Agreement for measurement of the C2-4 scalar was measured using a two-way mixed ICC for consistency on 20% of participants, selected at random, and revealed excellent consistency with scores of 1.00 (95% CI: 0.98, 1.00) for inter-rater and 0.97 (95% CI: 0.55, 1.00) for intra-rater agreement.

**Q2.** Results confirmed that size-of-the-system (C24 length in mm) was significantly greater in men (mean: 41.8 mm, 95% CI: 40.9–42.7) than in women (mean: 34.6 mm, 95% CI: 33.5–35.6) [F(1, 18)=34.12, p<0.001). This result was associated with a large effect size (Cohen's d=1.58).

**Q3.** The associations between size-of-the-system (as measured by C2-4 length) and hyoid excursion (in mm) are summarized in Table 3. Three of the four hyoid excursion parameters were significantly influenced by and positively correlated with the size-of-the-system: superior displacement, hypotenuse displacement and maximum XY position. Anterior displacement measures did not show any dependence on size-of-the-system. Maximum XY position measures displayed the highest correlation with the size-of-the-system at r = 0.63.

**Q4:** Descriptive statistics for each hyoid excursion parameter by sex and bolus volume in absolute units (mm) are presented in Table 4. A mixed model repeated measures ANOVA with factors of bolus volume and sex found significant sex differences for all measures except anterior hyoid displacement. On average, male participants consistently demonstrated greater hyoid excursion than female participants. Only the maximum XY position measure demonstrated a significant main effect of volume. Maximal XY position of the hyoid was significantly further from the C4 origin in the 20ml condition (mean: 64.9mm, 95% CI: 62.8–67.2) compared with both of the smaller bolus conditions (5ml mean: 61.9mm, 95% CI: 59.7–64.0 vs. 10ml mean: 62.7mm, 95% CI: 60.6–64.8); however, this result achieved

only a small effect size (Cohen's d=0.34). Results are summarized in Table 5. Given that anterior displacement did not demonstrate sensitivity to size-of-the-system (Q3) or sex (Q4), it was not included in the remaining analyses.

**Q5.** When the size-of-the-system covariate was added to the statistical model from Q4, all main effects of sex were neutralized (see Table 6). This was true for all three measures of hyoid excursion tested (superior displacement, hypotenuse displacement and maximum XY position, all measured in mm). In other words, no significant main effects of sex on hyoid excursion were found when the model accounted for size-of-the-system. Consistent with the analysis from Q4, a significant main effect of volume was observed when hyoid excursion was captured using maximal XY position, whereby 20ml boluses demonstrated significantly greater distance from C4 (mean: 66.34mm, 95% CI: 62.6–70.1) than the 5ml boluses (mean: 62.2mm, 95% CI: 58.4–65.8) or 10ml boluses (mean: 61.7, 95% CI: 58.0–65.4) with a medium effect size.

**Q6.** Our final statistical analysis involved transforming the metric of hyoid excursion from absolute units (mm) to internally anatomically scaled units (i.e., %C2-4 length). Descriptive statistics for the hyoid excursion parameters by sex and bolus volume are shown in scaled units in Table 7. The model explored in Q4 was repeated using these scaled units. The findings from Q5 were replicated (Table 8): no significant differences by sex were found, but a significant main effect of volume was observed when hyoid excursion was captured using maximal XY position whereby greater distance from the C4 origin for the 20ml bolus volume was observed (mean: 58.8 %C2-4, 95% CI: 53.9–63.7) compared to the 5ml (mean: 56.5 %C2-4, 95% CI: 51.6–61.3) and the 10ml boluses (mean: 55.3 %C2-4, 95% CI: 50.5–60.1) with a small effect. This finding demonstrates that the use of the internal anatomical scalar controlled for sex-related differences in participant size and hyoid position.

#### Discussion

In this study, we sought to accurately measure the physiological phenomenon of hyoid excursion during swallowing. We examined the relationship between size-of-the-system, sex and bolus volume and their impact on four different hyoid excursion parameters in a dataset of healthy individuals stratified by height, swallowing controlled volumes of ultra-thin liquid barium. When previous work has reported a significant sex difference in hyoid excursion, it has typically been the case that a greater extent of hyoid excursion was seen in men compared with women (Ishida et al., 2002, Logemann et al., 2002, Leonard et al., 2000). We hypothesized that sex effects in hyoid excursion may arise due to sex-based differences in the size-of-the-system and identified the length of the C2-4 segment as the best of 13 possible internal anatomical scalars, based on its correlation with participant height. This size-of-the-system parameter (C2-4 length) did indeed vary significantly by participant sex, with greater size-of-the-system in men. It is important to note that previous work has suggested that a fixed 15mm reference value can be assigned to the C3 vertebrae (Kim & McCullough, 2008; Sia, Carvajal, Carnaby-Mann, & Crary, 2011). While our data revealed that the mean size of C3 in our sample was remarkably close to this suggested referent (14.98mm), these values demonstrated a SD of +/- 2.49mm. Further, a post-hoc unpaired t-test demonstrated a significant difference between C3 size in men (mean:

16.7mm SD: 1.9) and women (mean: 13.3mm SD: 1.7) [t(18)=4.25, p=0.005]. Thus, we caution against the use of a fixed millimeter measure to represent the length of a single spinal vertebrae such as C3, and against the use of such fixed measures as a constant referent across participants, particularly if both male and female participants are being studied.

Our results confirm that hyoid excursion is greater in individuals with longer C2-4 cervical spine length, as measured by superior displacement, hypotenuse displacement, and maximum XY position parameters. Interestingly, the correlation observed between size-of-the-system and hypotenuse displacement in this study (r = 0.35, averaged across bolus volumes) is comparable to correlations reported previously by Leonard and colleagues (2000) between height and the hyoid hypotenuse displacement parameter (r = 0.37 for 20 ml).

Our results also replicate previous findings of significant sex differences in millimeter measures of hyoid excursion when size-of-the-system is not taken into account. Using this approach, it is clear that men have larger extent of hyoid movement than women. However, when the size-of-the-system is incorporated into the analysis, our results show that sex differences are no longer significant. Thus, we were able to demonstrate that apparent sex differences in hyoid excursion are actually explained by differences in participant size. Predictably, when hyoid excursion was scaled to the size-of-the-system (by expressing measures in %C2-4 units as opposed to mm), the same finding was replicated: sex differences in hyoid excursion were not found. Figure 5 illustrates the overall result of these analyses by participant and ordered by C2-4 length (in mm), showing confidence intervals for the maximum XY hyoid position parameter, averaged across the 5ml and 10 ml volume conditions. The male participants, shown with the black square data points, have longer cervical spine length measures and are consequently shown on the right hand side of the graph. As shown in the Figure, there is a clear overall trend towards greater maximum XY hyoid position with longer C2-4 length, with size-of-the-system explaining 58% of the observed variance. Of particular interest are the data for the participants in the middle of the C2-4 distribution, where we had participants of both sexes who were closely matched for size-of-the-system. For these participants, the dashed ellipse shows a close clustering of maximum XY hyoid position measures no clear separation by sex. Obviously this is a very small sample upon which to draw definitive conclusions about the presence or absence of sex differences.

The C2-4 scalar is a readily available and reliably selectable scalar in the radiographic view captured in a standard VF exam. We advocate for clinicians and researchers to adopt the practice of scaling hyoid excursion measures using the C2-4 length to control for the influence of differences in participant size. When this is done, there should be no reason to expect male patients to differ from female patients in the extent of hyoid excursion. The values in Table 7 can be used as reference values for young healthy participants swallowing ultra-thin liquid barium (22% w/v). We caution against the extrapolation of these reference values to other participant age-groups, bolus textures, barium densities, or swallowing conditions (such as continuous drinking) until future research has confirmed their applicability across contexts. Given known age-related changes in inter-vertebral disc space (Buckwalter, 1995; Logemann et al., 2000; Logemann, Pauloski, Rademaker, & Kahrilas,

2002), future research to confirm the utility of the C2-C4 internal scalar in older adults is particularly warranted. Once scaled reference values for hyoid excursion in healthy aging adults are obtained, they can be used to accurately identify reduced hyoid excursion and to set treatment targets for rehabilitation.

Of the four hyoid parameters studied in this experiment, anterior displacement was the only one to show no significant pattern of variation according to size-of-the-system, sex or bolus volume effects. One possible reason for this result may lie in the fact that this parameter also demonstrated the poorest inter- and intra-rater reliability scores, thereby suggesting that it is more prone to measurement error. Maximum XY hyoid position performed similarly to superior and hypotenuse displacement measures, showing variation according to sex (without consideration of size-of-the-system) and to size-of-the-system; however, it was the only measurement method that revealed significant variation according to bolus volume. Interestingly, this parameter also had the highest correlation with participant height. We conclude that this measure provides the most accurate method for measuring hyoid excursion, because it captures both planes of movement while also excluding difficulties and variability associated with selecting an appropriate rest frame.

#### Conclusion

Hyoid excursion during swallowing is dependent on a person's size (size-of-the-system). Taller individuals have longer cervical spine length and demonstrate greater superior displacement, hypotenuse displacement and maximal XY position of the hyoid. When measurements do not control for the influence of size-of-the-system, sex differences in hyoid excursion are observed. Using the C2-4 length as an internal anatomical scalar neutralizes these apparent sex differences. Capturing hyoid excursion using the parameter of maximal XY position limits measurement error attributable to difficulties with rest frame selection. This parameter is sensitive to variations in hyoid excursion across bolus volume. Further research in healthy aging is required before applying reference values to patient populations.

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**Figure 1.** Distribution of height by sex.



#### Figure 2.

Illustration of marking points and hyoid parameters, measured relative to the cervical spine (origin at C4).



**Figure 3.** Schematic of research questions 1 through 5.

	C1	Internal Anatomical Scalar	Operational Definition	Pearson Correlation (r) with Height
		C2-C4 Length	h-a	0.83
	C2	C2-C5 Length	j-a	0.82
\ ///	a	C3-C5 Length	j-b	0.78
\ ///	b	Trachea Width	n-m	0.77
	C3	C4 Anterior Height	h-f	0.75
k	c e	Anterior Hyoid Height	l-k	0.72
	fg	C3 Anterior Height	c-b	0.69
	C4	C4 Inferior Length	i-h	0.65
$\setminus$	h i	C3 Posterior Height	e-d	0.64
		C3 Inferior Length	e-c	0.63
() rep	65	C4 Posterior Height	i-g	0.63
		C3 Superior Length	d-b	0.61
// /		C4 Superior Length	g-f	0.46





#### Figure 5.

Means and confidence intervals for maximum XY hyoid position shown by participant in rank order of C2-4 cervical spine length. Male participants are shown by the black square data points and females by the light grey diamonds. The dashed ellipse highlights participants with similar cervical spine length of both sexes.

#### Table 1

Targeted volumes, pipetted volumes and actual volumes (mean and 95% CI)

Towast volume (ml)	Volume ninetted into our (ml)	Maan malance anallemed (ml)	95% Confide	ence Interval
Target volume (m)	volume pipetted into cup (mi)	Mean volume swallowed (mi)	Lower Bound	Upper Bound
5	6	3.54	3.42	3.67
10	11	8.03	7.78	8.28
20	21	17.34	16.84	17.85

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#### Table 2

Intra- and Inter-rater reliability measures for four hyoid excursion parameters. ICC = Intraclass Correlation Coefficient, CI = Confidence Interval

	Intra-ra	ter Reliability	Inter-ra	ter Reliability
	ICC	95% CI	ICC	95% CI
Anterior Displacement	0.61	-0.35-0.88	0.59	-0.43, 0.88
Superior Displacement	0.94	0.79–0.98	0.88	0.59–0.97
Hypotenuse Displacement	0.81	0.35-0.94	0.90	0.65–0.97
Maximum XY Position	0.85	0.49-0.96	0.79	0.26-0.94

#### Table 3

Associations between four methods for capturing hyoid excursion and size of-the-system (as measured by C2-4 length).

	Association between hy	oid measurer	nent method and size-of-	the-system
	Degrees of Freedom (df)	F Statistic	Significance (p value)	Correlation $(r)$ with Size-of-the-System
Anterior Displacement	(1, 18.2)	0.7	0.425	0.12
Superior Displacement	(1, 18.0)	5.2	0.035	0.37
Hypotenuse Displacement	(1, 18.1)	5.1	0.036	0.35
Maximum XY Position	(1, 18.1)	26.4	<0.001	0.63

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# Table 4

Descriptive statistics for hyoid excursion (measured four ways in mm) by sex and bolus volume. CI = Confidence Interval

			MEN			WOMEN	
		5ml	10ml	20ml	5ml	10ml	20ml
	Mean (mm)	15.6	14.7	16.1	14.5	13.9	14.4
Anterior Displacement	95% CI	13.9–17.4	12.9–16.5	14.3–17.8	12.7–16.3	12.1–15.6	12.6–16.3
	Mean (mm)	19.3	20.3	21.3	15.8	15.3	16.4
Superior Displacement	95% CI	16.1–22.5	17.1–23.5	18.1–24.5	12.9–19.0	12.1–18.5	13.1–19.6
	Mean (mm)	25.1	25.3	27.1	21.9	21.0	22.1
Hypotenuse Displacement	95% CI	22.2-28.0	22.4–28.2	24.1–30.0	19.0–24.8	18.1–23.9	19.1–25.1
	Mean (mm)	67.3	69.0	71.8	56.5	56.4	58.2
	95% CI	64.2-70.3	65.9–72.0	68.7–74.8	53.5-59.6	53.4-59.5	55.0-61.3

## Table 5

Results of Question 4 exploring contributions of bolus volume and sex to hyoid excursion measures (in mm) without controlling for the size-of-thesystem

		Association between hyoid	measurement	method and sex	
	Main effect of sex?	Degrees of Freedom (df)	F Statistic	Significance (p value)	Effect size (d)
Anterior Displacement	ON	(1, 18.2)	1.3	0.266	
Superior Displacement	YES	(1, 18.1)	4.8	0.042	0.74
Hypotenuse Displacement	YES	(1, 18.1)	5.2	0.034	0.72
Maximum XY Position	YES	(1, 18.2)	47.1	<0.001	1.34
		Association between hyoid	measurement	method and volume	
	Main effect of volum	e? Degrees of Freedom (d	lf) F Statistic	Significance (p valu	ie) Effect size (
Anterior Displacement	ON	(2, 148.2)	1.3	0.209	ı
Superior Displacement	ON	(2, 147.5)	1.8	0.163	
Hypotenuse Displacement	ON	(2, 147.7)	1.9	0.155	
Maximum XY Position	YES	(2, 148.2)	4.9	0.00	0.34

## Table 6

Results of Question 5 exploring contributions of bolus volume and sex to hyoid excursion measures (in mm) while controlling for the size-of-the-system

		Association between hyoid	l measurement	t method and sex	
	Main effect of sex?	Degrees of Freedom (df)	F Statistic	Significance (p value)	Effect size (d)
Superior Displacement	ON	(1, 16.0)	0.22	0.649	ŗ
Hypotenuse Displacement	NO	(1, 16.0)	0.24	0.629	,
Maximum XY Position	ON	(1, 15.9)	0.21	0.651	ı
		Association between hyoid	l measurement	t method and volume	
	Main effect of volum	e? Degrees of Freedom (G	df) F Statist	ic Significance (p valu	e) Effect size (d)
Superior Displacement	ON	(2, 143.1)	2.1	0.126	
Hypotenuse Displacement	ON	(2, 147.7)	0.8	0.443	,
Maximum XV Position	VFS	() 143 3)	4.0	0.020	0.53

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Descriptive statistics for scaled hyoid excursion (measured in %)	

			MEN			WOMEN	
		5ml	10ml	20ml	5ml	10ml	20ml
Line Design	Mean (%C2-4)	42.7	43.8	46.7	41.4	41.3	43.2
Superior Displacement	95% CI	35.2–50.2	36.3–51.3	39.1–54.2	33.9–48.9	33.8-48.8	35.5–50.9
	Mean (%C2-4)	55.6	54.7	59.4	57.3	55.9	58.2
Hypotenuse Displacement	95% CI	48.8–62.4	47.9–61.5	52.5-66.2	50.5-64.2	49.1–62.7	51.2-65.3
	Mean (%C2-4)	148.2	149.9	157.9	147.6	147.7	153.0
MAXIMUM AT POSITION	95% CI	141.9–156.4	142.6–157.1	150.6-165.2	140.3-154.9	140.5-155.0	145.4-160.6

## Table 8

Results of Question 5 exploring contributions of bolus volume and sex to hyoid excursion measures (in scaled units, %C2-4) to control for the size-of-thesystem

		Association Detween iny VIU		THETHOM WIN SCY	
	Main effect of sex?	Degrees of Freedom (df)	F Statistic	Significance (p value)	Effect size (d)
Superior Displacement	ON	(1, 18.1)	0.26	0.617	1
Hypotenuse Displacement	ON	(1, 18.1)	0.02	0.891	ı
Maximum XY Position	ON	(1, 18.0)	0.44	0.515	ı
		Association between hyoid	measurement	t method and volume	
	Main effect of volum	e? Degrees of Freedom (c	lf) F Statist	ic Significance (p valu	ıe) Effect size (
Superior Displacement	NO	(2, 143.5)	1.5	0.226	
Hypotenuse Displacement	ON	(2, 143.7)	1.8	0.166	ı
Maximum XY Position	YES	(2, 143.9)	5.5	0.005	0.47