Modulation of laryngeal and respiratory pump muscle activities with upper airway pressure and flow

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Stella, M. H., and S. J. England. Modulation of laryngeal and respiratory pump muscle activities with upper airway pressure and flow. J Appl Physiol 91: 897–904, 2001.—The hypothesis that upper airway (UA) pressure and flow modulate respiratory muscle activity in a respiratory phase-specific fashion was assessed in anesthetized, tracheotomized, spontaneously breathing piglets. We generated negative pressure and inspiratory flow in phase with tracheal inspiration or positive pressure and expiratory flow in phase with tracheal expiration in the isolated UA. Stimulation of UA negative pressure receptors with body temperature air resulted in a 10–15% enhancement of phasic moving-time-averaged posterior cricoarytenoid electromyographic (EMG) activity above tonic levels obtained without pressure and flow in the UA (baseline). Stimulation of UA positive pressure receptors increased phasic moving-time-averaged thyroarytenoid EMG activity above tonic levels by 45% from baseline. The same enhancement of posterior cricoarytenoid or thyroarytenoid EMG activity was observed with the addition of flow receptor stimulation with room temperature air. Tidal volume and diaphragmatic and abdominal muscle activity were unaffected by UA flow and/or pressure, whereas respiratory timing was minimally affected. We conclude that laryngeal afferents, mainly from pressure receptors, are important in modulating the respiratory activity of laryngeal muscles.

INTRINSIC LARYNGEAL MUSCLES contract to bring about changes in glottic size, which can affect resistance to airflow into and out of the lungs. Phasic laryngeal abduction during inspiration widens the glottic opening and decreases resistance to airflow (4, 5). Phasic expiratory activity of muscles that adduct or narrow the vocal folds results in increased resistance to airflow out of the lungs (5, 12).

Pressure and/or airflow has been shown to modulate the phasic respiratory activity of intrinsic laryngeal muscles. Phasic activity of the posterior cricoarytenoid muscle (PCA), a laryngeal adductor, increases when airflow is diverted from tracheotomy to upper airway (UA) breathing (25) and in response to negative pressure pulses (38). Insalaco et al. (10) showed increased activity of the thyroarytenoid muscle (TA), a laryngeal adductor, in response to increased resistive loading.

In anesthetized neonatal animal models, constant flow and/or pressure in the isolated UA has been shown to result in a ventilatory inhibition characterized by a reduction in breathing frequency and tidal volume (V̇t) (1, 6, 14, 17, 20). This ventilatory inhibition is abolished after bilateral section of the superior laryngeal nerve (SLN) or topical anesthesia of the laryngeal region (1, 6, 20). However, chronic section of the SLN does not affect the breathing pattern in conscious kittens (20), suggesting that the inhibitory effects of SLN afferents are not a major factor during eupnea in the conscious animal but can be magnified under certain circumstances or that adaptation occurs after ablation of these reflex pathways.

Sant’Ambroigo and co-workers (27) identified and characterized pressure- and flow-sensitive endings in the internal branch of the SLN. “Flow” receptors actually function as thermoreceptors, stimulated by cooling of the UA mucosa from body temperature to room temperature. Pressure-sensitive endings respond to positive or negative transmural pressure applications; they seldom respond to both (27).

To evaluate the contribution of UA receptors sensing pressure and flow to the controlled respiratory phase-specific activation of the PCA, TA, and respiratory pump muscles, we studied the steady-state effects of presumably stimulating specific UA receptors (flow and/or pressure) in anesthetized, tracheotomized, spontaneously breathing piglets with an isolated UA with oscillatory airflows in phase with the animal’s tracheal breathing pattern. The magnitudes of pressure and flow applied to the isolated UA were independent of those in the lower airway and were within the physiological range for the respiratory drive used in our studies. By altering the temperature of the air applied to the isolated UA, we presumably stimulated pressure receptors only (with air at body temperature) or both pressure and flow receptors (with air at room temperature). We hypothesized that UA pressure and flow...
flow would preferentially modulate the phasic activity of laryngeal muscles compared with respiratory pump muscles in a respiratory phase-specific fashion. We also assessed the relative contributions of UA flow and/or pressure to this modulation.

METHODS

The protocol was approved by the Institutional Animal Care and Use Committee at the Robert Wood Johnson Medical School.

Surgical preparation. Twenty-seven piglets of either gender (15.4 ± 1.9 days of age, range 3–33 days; 3.92 ± 0.28 kg body wt) were premedicated for surgical preparation with acepromazine (1 mg/kg im) and ketamine (45 mg/kg im). The animals were secured, in the supine position with the neck extended, on a surgical table. Rectal temperature was monitored (Yellow Springs Instruments telethermometer probe) and maintained at 37–39°C with a heating pad. A few drops of Xylocaine (2% lidocaine HCl, 20 mg/ml sc) or a topical anesthetic skin refrigerant (ethyl chloride) were used to premedicate areas before incisions. Cannulas were placed in the femoral artery for blood pressure monitoring and blood-gas sampling and in the femoral vein for maintenance of anesthesia by constant infusion of ketamine (0.3 mg.min⁻¹.kg⁻¹ as required to maintain anesthesia). A longitudinal neck incision was made to expose the larynx and trachea. The trachea was sectioned between the third and fourth ring, and an endotracheal tube was inserted in the caudal segment. A differential pressure transducer (Validyne) and a pneumotachograph (Hans Rudolph) connected to a transducer (Validyne) were attached to monitor tracheal flow and pressure, respectively. The rostral segment of the trachea was intubated below the larynx and attached to a pneumotachograph (Fleisch) connected to a differential pressure transducer (Validyne) for the measurement of UA flow and to a differential pressure transducer (Validyne) for the measurement of pressure.

Bipolar fine-wire stainless steel electrodes were inserted into the diaphragm (Dia), PCA, TA, and abdominal muscle (Abd) for recording of electromyographic (EMG) activity. The bare tip of each electrode was hooked onto a 20-gauge needle, which was inserted into the muscle and then retracted, leaving the electrode positioned in the muscle. Electrodes were placed in the PCA, a few millimeters apart, under direct visualization by reflection of the upper tracheal cannula rostrally. The TA was accessed by insertion of needles through the cricothyroid membrane bilaterally. The lower costal margin of the Dia and the inner muscular portion of the transversus abdominis were accessed through the skin. A grounding electrode was connected to the lower limb. Placement of electrodes was confirmed on autopsy. EMG signals were amplified, full-wave rectified, band-pass filtered between 20 and 3,000 Hz (BMA 830, CWE), and electronically integrated with a moving time averager (MTA) with a time constant of 100 ms (MA 821, CWE). A 60-Hz notch filter (model NL126, Neurolog) was used to reduce background noise.

Animals were initially exposed to hyperoxia (50% O₂-balance N₂). Phasic UA muscle activity during hyperoxia was reduced in our preparation in the absence of pressure and flow in the isolated UA after tracheotomy with ketamine anesthesia (9) and neck extension (2). Whenever necessary, to enhance phasic PCA activity above tonic levels, animals were exposed to hypercapnia (4–6% CO₂-50% O₂-balance N₂). To enhance phasic TA activity above tonic levels, animals were exposed to a hypoxic inspired gas mixture (12% O₂-balance N₂). These conditions were maintained throughout baseline (the absence of UA flow and/or pressure) and experimental measurements. In 18 of 19 studies, hypercapnia was required to enhance phasic PCA activity. In 4 of 11 studies, hypoxia was required to obtain phasic TA activity.

Isolated UA breathing circuit. The UA was isolated, and a breathing circuit was constructed for the generation of oscillatory pressure and flow with air at room temperature or warmed to 40°C by passage through coiled tubing submerged in a temperature-controlled bath to achieve body temperature when the air passed through the upper airway.

Tracheal flow during inspiration was used to command a servo-controlled valve (3) connected to the isolated UA. Tracheal inspiration resulted in concomitant opening of the valve, which exposed the larynx to a negative pressure source, pulling bias air available at the snout in the inspiratory direction, concomitant with tracheal inspiration. Closure of the valve (triggered by the lack of inspiratory tracheal flow) resulted in the absence of UA pressure and flow (Fig. 1A). Alternatively, tracheal flow during expiration was used to command opening of the servo-controlled valve, exposing the larynx to a positive pressure source, which pushed air in the expiratory direction, concomitant with tracheal expiration (Fig. 1B). The shape of the resultant UA pressure and flow waveforms is illustrated in Fig. 1. The chosen ranges of UA pressure were within physiological ranges for the respiratory drive used in these studies. The maximum negative pressure applied to the UA was −9.5 ± 0.7 cmH₂O, whereas the maximum positive pressure was 9.5 ± 0.5 cmH₂O, resulting in UA flows of ≥0.5 l/min.
Static respiratory mechanics measurements. Pressure-volume relationships were constructed for the total respiratory system and lung only, postmortem, before and after thoracotomy in 10 piglets. Animals were euthanized, and the tracheotomy tube was fitted with a three-way connector attached to a 60-ml syringe and a pressure transducer. To determine the pressure-volume relationship for the total respiratory system, lung inflations (5, 10, 15, 20, 25, 30, 35, and 40 ml) and deflations (5, 10, 15, and 20 ml), relative to resting volume, were applied at a constant rate. Once each target inflation or deflation volume was reached, it was maintained to obtain a plateau on the pressure recording, and then the volume was returned to resting levels. Subsequently, once the pressure returned to resting levels, the procedure was repeated until all pressure measurements were obtained for all target volumes. A thoracotomy was performed via a longitudinal incision alongside the sternum to expose the lungs, the chest wall was widely retracted, and the pressure-volume relationship for the lung only was determined by repeating the protocol.

Data collection and analysis. Data were recorded on chart paper (model TA 4000, Gould) and digital tape (model 4000A, Vetter). Pressure, flow, and EMG MTA data were digitized at a sampling rate of 25 Hz for computer-assisted analysis on-line on a desktop microcomputer fitted with data-acquisition hardware and storage (CODAS, DATAQ). EMG activities were quantitated as the peak height of the phasic MTA activity above tonic levels during inspiration (PCA and Dia) or expiration (TA and Abd). Ventilatory parameters [inspiratory (Ti), expiratory (Te), and total respiratory time (Tr)] were measured or calculated breath by breath using data-acquisition software (Advanced CODAS, DATAQ). Ti was defined as the time from the onset to maximum phasic activity of the MTA Dia EMG above tonic levels for each breath. Te was defined as the time from the maximum level of phasic MTA Dia EMG above tonic values to the onset of the subsequent breath. Tr was defined as the sum of Ti and Te. Vt was calculated from digital integration of tracheal flow signals. Additional calculations were performed with spreadsheet software (Quattro Pro for Windows, Borland).

Responses obtained during control or baseline conditions were calculated as an average of values obtained in 20 breaths before the application of each pressure and flow stimulus to the isolated UA. Stimulus values were computed as an average of values corresponding to 20 consecutive breaths. The first 10 breaths after the application of the stimulus were excluded to avoid pressure and flow artifacts due to opening and closure of the servo-respirator valve and to allow for attainment of a steady-state response. By assessment of the coefficient of variation, we determined that, during the first 10 breaths after stimulus application, there was a gradual change in the respiratory parameters to the steady-state response measurable in the subsequent 20 breaths. Changes in parameters were quantified as percent changes from control parameters.

Total respiratory system (Cr) and lung compliance (Ct) were calculated from the inverse of the slope of the linear portion of the pressure-volume relationship, within the resting volume range, before and after thoracotomy, respectively. Pressure-volume curves were fitted with a third-order polynomial regression (SigmaPlot, Jandel Scientific), and the coefficient of the first term was used as the slope of the linear portion of the curve. Chest wall compliance (Cw) was calculated from the following relationship: 1/Cw = 1/Cr - 1/Ct. The relationship between Cr, Cw, and Cw/Ct with age was determined, and the correlation between Cw/Ct and age was calculated.

Statistical differences in parameters obtained under stimulus conditions were assessed by t-tests. Whenever normality and equal variance tests failed, nonparametric statistics (sign tests or rank-sum tests) were used instead to interpret results. The chosen level of significance was $P < 0.05$. Maturation changes were assessed by computing the Pearson correlation coefficient for the measured respiratory variables vs. age, and significance was assessed by the calculated probability value. Values are means ± SE. Tests were conducted using computer software for statistical analysis (SigmaStat, Jandel Scientific).

RESULTS

Effect of UA negative pressure and inspiratory flow applied during inspiration only. Experiments to assess the effect of UA negative pressure and inspiratory flow, applied during inspiration only, on respiratory activity were conducted under exposure to hypercapnia or, when phasic PCA activity was present, under hyperoxia. Figure 2 illustrates the typical pattern of responses observed, which consists of UA airflows at body and room temperature, resulting in enhancement of phasic PCA EMG while Dia and Abd remained unaffected compared with baseline values (absence of pressure and flow in the isolated UA).

The observations in a single animal were supported by the mean data assessed in 19 piglets (15.4 ± 2.5 days of age, range 3–33 days; 3.81 ± 0.33 kg body wt; Table 1) of the response of the PCA EMG to UA body or room temperature oscillatory flow and negative pressure during inspiration only. Both air temperatures resulted in significant enhancement of PCA EMG. The degree of enhancement brought about by stimulation of pressure receptors only with body temperature air (11.1 ± 3.1%) or by stimulation of pressure and flow...
Table 1. Respiratory effects of negative pressure and inspiratory flow applied to the isolated UA

<table>
<thead>
<tr>
<th>UA Air</th>
<th>PCA</th>
<th>Dia</th>
<th>Abd</th>
<th>Tr</th>
<th>Tx</th>
<th>Vt</th>
</tr>
</thead>
<tbody>
<tr>
<td>Room temperature</td>
<td>15.7 ± 3.7*</td>
<td>-2.8 ± 0.8</td>
<td>6.7 ± 4.7</td>
<td>4.2 ± 1.1*</td>
<td>2.7 ± 0.9*</td>
<td>0.8 ± 1.0</td>
</tr>
<tr>
<td>Body temperature</td>
<td>11.1 ± 3.1*</td>
<td>-2.7 ± 2.3</td>
<td>-3.5 ± 4.4</td>
<td>4.2 ± 1.0*</td>
<td>2.4 ± 1.9*</td>
<td>0.3 ± 1.4</td>
</tr>
</tbody>
</table>

Values (means ± SE) represent percent change from baseline [absence of pressure and flow in the isolated upper airway (UA)]; n, number of piglets. Room temperature air in the UA stimulates negative pressure and flow receptors; body temperature air in the UA stimulates negative pressure receptors only. PCA, Dia, and Abd, posterior cricoarytenoid, diaphragm, and abdominal electromyogram activity, respectively; Tr, total respiratory time; Te, expiratory time; Vt, tidal volume. *Significantly different from baseline (P < 0.05).

receptors with room temperature air (15.7 ± 3.7%) was not significantly different. The accompanying responses of the Dia, as well as of Abd, Tr, and Te, were assessed in 17, 11, 17, 17, and 19 piglets (Dia EMG from 2 piglets was technically unacceptable, tracheal flow records for 1 piglet were unavailable, phasic Abd EMG was present in 11 piglets). Dia and Vt were not significantly different from baseline values. Tr and Te were significantly increased from baseline with airflow at body temperature (4.2 ± 1.0 and 1.5 ± 2.0%, respectively) and room temperature (4.2 ± 1.1 and 2.2 ± 0.9%, respectively) applied to the isolated UA. These changes were not significantly different from each other. Our results did not reveal significant age differences in the respiratory response to UA negative pressure and inspiratory flow applied during inspiration only.

Effect of UA positive pressure and expiratory flow applied during expiration only. Experiments to assess the effect of UA positive pressure and expiratory flow, applied during expiration only, on respiratory activity were conducted under exposure to hyperoxia when phasic TA activity was present. Animals were exposed to hypoxia if phasic TA activity was absent under hyperoxic inspired gas mixtures. Phasic PCA and Abd activity was rarely present. The typical pattern of responses, which consists of UA airflows at body and room temperature, resulting in enhancement of phasic TA EMG compared with baseline values. Figure 3 also shows a reduction of respiratory frequency with room or body temperature oscillatory pressure and flow.

The observations in a single animal were supported by the mean data assessed in 11 piglets (17.0 ± 2.6 days of age, range 6–31 days; 3.98 ± 0.37 kg body wt; Table 2) of the response of the TA EMG to body or room temperature oscillatory flow and positive pressure applied to the UA during expiration only. Both air temperatures resulted in significant enhancement of TA EMG. The degree of enhancement brought about by stimulation of pressure receptors only with body temperature air (45.3 ± 9.2%) was not significantly different from that brought about by stimulation of both pressure and flow receptors with room temperature air (49.6 ± 20.7%). The accompanying responses of Dia EMG activity, Vt, Tr, and Te were also assessed. Vt was not significantly different from baseline values. Dia EMG activity increased with room temperature air only (4.5 ± 3.6%), Te increased with body temperature air only (10.1 ± 3.4%), and Tr increased with room and body temperature air (5.2 ± 2.5 and 10.3 ± 2.4%, respectively) compared with baseline values. When the effects of perfusing the isolated UA with body and room temperature air were compared, changes in Tr were not significantly different from each other. Our results did not reveal significant age differences in the respiratory response to UA positive pressure and expiratory flow applied during expiration only.

Static respiratory compliance measurements. Maturation changes in Ctr, Cl, Cw, and Cw/Cl were assessed in 10 piglets (16.5 ± 3.1 days of age, range 5–33 days; 4.17 ± 0.49 kg body wt; Fig. 4). Ctr and Cw diminish with age, with the majority of the changes occurring within the 1st wk and stabilizing by the 2nd and 3rd wk of life. The correlation between Cw/Cl and age (Pearson coefficient = −0.605, r² = 0.366, P = 0.064), although not statistically significant, suggests an inversely proportional trend between the two variables. Cw/Cl remains above unity during the 1st mo of life of the piglet.

DISCUSSION

UA flow-sensitive endings can be inactivated by perfusion of the isolated UA with body temperature air, whereas room temperature air can be utilized to stim-
ulate pressure and flow receptors (29). Cooling of the laryngeal mucosa, while stimulating flow-sensitive endings, diminishes but does not inactivate pressure receptor discharge compared with air at body temperature (26). When the response of the PCA to oscillatory negative pressure and flow is compared, the degree of enhancement was not statistically significantly different between air temperature treatments. The response of the PCA suggests that, despite pressure receptor inhibition due to cooling, feedback from flow receptors is sufficient to result in the same degree of enhancement under both temperature conditions.

Studies of the TA response to receptor stimulation showed that activation of UA positive pressure receptors only or both pressure and flow receptors favors adduction. The pressure receptor population stimulated in these experiments differs from that stimulated with negative pressure, inasmuch as laryngeal pressure receptors have been described as being sensitive to positive or negative transmural pressure applications, but seldom to both (27). Because there were no statistically significant differences between afferent stimulation due to oscillatory room temperature and body temperature air in the isolated UA, feedback from flow receptors is sufficient to counteract the inhibitory effect of cooling on pressure receptors and result in the same degree of enhancement under both temperature conditions.

Although our results showed statistically significant changes in Dia EMG activity and respiratory timing with UA flow and/or pressure, these small changes are unlikely to be physiologically significant. However, our results suggest that laryngeal endings do modulate the phasic respiratory activity of laryngeal muscles. Does removal of this sensory feedback modify eupneic breathing pattern? Stockwell et al. (33) compared ventilatory parameters in humans before and after lidocaine-induced anesthesia of the SLN. They did not observe any differences in parameters attributable to SLN conduction blockade and concluded that SLN afferents do not play a significant role in resting breathing pattern in humans. Mortola and Rezzoionico (20) addressed the same question by studying anesthetized and unanesthetized kittens. Although acute section of the SLN abolished inhibitory ventilatory effects of steady airflows applied to isolated UA, ventilatory inhibition is still present with chronic SLN section. The authors allude to other non-SLN UA receptors developing compensatory mechanisms. One must consider the possible role of sensory innervation of the nasopharynx supplied by the glossopharyngeal nerves investigated by Widdicombe et al. (37) and Mathew and Sant’Ambrogio (15). Another strong candidate is innervation of the nasal mucosa supplied by the ophthalmic and maxillary divisions of the trigeminal nerve (29). Tsubone (34) found cold- and pressure-sensitive endings in the nasal cavity innervated by the ethmoidal nerve, a branch of the ophthalmic division of the trigeminal nerve. Unlike studies from the laboratories of Sant’Ambrogio and Mortola, which largely bypassed nonlaryngeal UA receptors by placing a cannula immediately above the larynx, the isolated UA route of breathing in our experimental design spanned nasal-to-subglottic areas, thus stimulating nasal, pharyngeal, and laryngeal pressure and flow receptors.

Mathew et al. (13), as well as Al-Shway and Mortola (1), were able to eliminate reflex responses to change in UA pressure and/or flow by topical anesthesia of the laryngeal region, suggesting a superficial location for these receptors. Although the majority of the UA pressure- and flow-sensitive endings alluded to in this study do travel via the internal branch of the SLN, because additional receptors are located in the nasal and pharyngeal UA, bilateral sectioning of the SLN in our preparation would not necessarily eliminate the reflexes described here.

A recent review by Sant’Ambrogio et al. (29) comments that, under eupneic breathing conditions, laryngeal afferents have not been shown to substantially modify breathing patterns. However, the authors suggest a possible role in maintenance of respiratory homeostasis during hyperventilation or “special conditions” that challenge the respiratory system. In our experimental design, animals were challenged with hypercapnia or hypoxia. Although these conditions were present throughout control and UA receptor-stimulated trials, they may have been sufficient to provide a different set point from eupnea for the respiratory system to bring about the modulating effects of UA pressure and flow receptor stimulation. This may play a role in the fact that UA positive pressure and expiratory flow were shown to affect TA activity more than negative pressure and inspiratory flow affected PCA activity (10–15% vs. 45–50%). Furthermore, a stronger reflex activation of TA than of PCA activity from UA receptor stimulation can be speculated when the maturational age of the animal model used in our studies is considered.

Increased phasic TA activity or adduction of the vocal folds during expiration lengthens the expiratory time constant for airflow, resulting in “braking” of...
experatory airflow. Experatory airflow braking is a mechanism by which end-expiratory lung volumes can be increased above passive levels (24). Breathing from elevated end-expiratory lung volume is a characteristic of neonates (11, 19, 21), who must do so to compensate for highly compliant chest walls (8, 16, 23). Our static respiratory mechanics measurements in piglets showed that, although most of the changes in Cw occur by the 1st wk of life (Fig. 4A), Cw/Cl. is still above unity by the 3rd wk of life (Fig. 4B). Thus critical control of laryngeal adduction can be instrumental in these animals to compensate for their mechanical disadvantage. Although our results did not reveal significant age differences in the respiratory response to UA pressure and flow for piglets within the 1st mo of life, we speculate that restricting the study to piglets within the 1st wk of life might reveal such differences. Our static respiratory mechanics measurements are in agreement with those reported by Fisher and Mortola (7) for piglets at 3 ± 2 days of age. We are unaware of any other reports of static compliance measurements in piglets within the 1st mo of life.

The study of single-unit SLN afferent fiber recordings by Sant'Ambrogio et al. (27) showed that UA pressure and flow receptors can be stimulated for short or long periods of time, suggesting that the reflex response to laryngeal receptor stimulation could be affected by stimulus modality. To ensure activation of UA receptors similar to that stimulated during eupneic breathing, we applied pressure and/or flow waveforms to the isolated UA in phase with those generated by the animal. Although transmural pressure or flow applications to the UA have been previously shown to affect laryngeal muscle activity, the studies presented are unique, in that the modality of pressure and/or flow application is a more physiological-like stimulus than constant, square-wave, or intermittent pressure applications used by other investigators. Furthermore, the UA pressure levels utilized in the studies presented here are closer to physiological values than the levels used by other investigators. For example, Fisher et al. (6) used positive and negative pressures from 2 to 20 cmH2O in puppies; in rabbit studies, Woodall and Mathew (39) used up to 15 cmH 2O positive and negative pressure pulses of 200-ms duration. Although high pressures can result in considerable laryngeal distortion and thus activate other endings such as joint receptors, low pressures can lead to the lack of reflex responses (15). However, the range of receptor discharge responses to pressure suggests that most of the response would be activated in the low pressure range. The chosen ranges of UA pressures used in our studies allowed for minimal effects on respiratory timing (≤10% reduction in Tr or Te) while still enhancing laryngeal muscle activity. This can account for differences between studies in which a more drastic reduction in breathing frequency was observed in response to constant negative pressures, such as the rabbit study by Mathew et al. (13). Finally, the focus of our study was on a steady-state response, present after the first 10 breaths following stimulus application, which differs from previous studies that measured the immediate effects on stimulus application (1, 6).

Our choice of animal model may influence our results, inasmuch as species differences have been found in the relative distribution of pressure and flow receptors in the UA, in their location within the UA, and in

Fig. 4. Maturational changes in total respiratory system (Ct), lung (Cl), and chest wall (Cw) compliance (A) and in ratio Cw/Cl. (B) in 10 piglets. Ct and Cw diminish with age, with the majority of the changes occurring within the 1st wk of life and stabilizing by the 2nd and 3rd wk. Correlation between Cw/Cl. and age (Pearson coefficient = −0.605, r² = 0.366, P = 0.064), although not statistically significant, suggests an inversely proportional trend between the 2 variables. Cw/Cl. remains above unity during the 1st mo of life of the piglet.
the response of these receptors to respiratory stimuli. For example, although flow receptors have been identified in the SLN of dogs (28) and rabbits (18), they were not found in guinea pigs (35). However, cooling of the guinea pig larynx with l-menthol, a selective stimulant of laryngeal cold receptors, results in apnea (22), suggesting the possibility that these endings exist but are difficult to isolate. In the nose, cooling receptors have been identified in cats (36), rats (34), and guinea pigs (32).

Mechanoreceptors with a respiratory modulation have been found in the SLN of dogs (27), rats (31), guinea pigs (35), and rabbits (18). However, unlike laryngeal mechanoreceptors in dogs, the majority of pressure receptors in these species are stimulated by positive pressure and inhibited by collapsing pressure, possibly because of their location on the epiglottis and its movement during respiration (29).

Although the prevalence of laryngeal negative pressure receptors may differ between species, this does not necessarily imply a species difference in the reflex response to UA pressure and flow. UA pressure and flow receptors, although in different numbers and locations, exist in all of these species. For example, the low percentage of laryngeal negative pressure receptors in some species may be compensated by receptors found elsewhere in the UA, inasmuch as nasal pressure receptors have been identified in cats, rats, and guinea pigs (29, 30, 36). We are unaware of any studies that identified pressure and flow receptors in the UA of piglets. However, our study of the respiratory modulation of laryngeal muscle activity with UA pressure and flow suggests the potential existence of these receptors in piglets.

Because UA pressure and flow during inspiration did not affect Dia or Abd activity while favoring enhanced laryngeal abduction, we conclude that the resultant decrease in inspiratory UA resistance should lessen the load imposed on respiratory pump muscles and promote UA patency. Similarly, inasmuch as laryngeal adduction is favored during expiration, increased expiratory UA resistance assists in braking of expiratory airflow, lessening the load imposed on chest wall muscles to achieve the same level of braking. We conclude that UA afferent information plays an important role in modulating phasic respiratory activity of intrinsic laryngeal muscles on a breath-by-breath basis.

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REFERENCES

25. Sant’Ambrogio FB, Mathew OP, Clark WD, and Sant’Ambrogio G. Laryngeal influences on breathing pattern


