Monitoring respiratory function and sleep in the obese Vietnamese pot-bellied pig

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Tuck, Stephanie A., Joseph C. Dort, Merle E. Olson, and John E. Remmers. Monitoring respiratory function and sleep in the obese Vietnamese pot-bellied pig. J. Appl. Physiol. 87(1): 444–451, 1999.—Development of drug treatments for obstructive sleep-disordered breathing has been impeded by the lack of animal models. The obese pig may be a suitable animal model, as it has been reported to experience sleep-disordered breathing resembling human obstructive sleep apnea. The purpose of this paper is to describe in detail techniques for chronic instrumentation of the obese Vietnamese pot-bellied pig and to study respiratory function during sleep. Under general anesthesia, four obese pigs were instrumented for long-term recording of intrapleural and tracheal pressures, genioglossal EMG, and bioelectric signals related to sleep. A custom-fitted face mask was used to record respiratory variables including airflow, snoring, and expired CO2. Most chronic instrumentation provided robust signals for up to 6 wk after installation. All pigs displayed sleep-disordered breathing characterized by increased resistance to airflow, snoring, inspiratory flow limitation, and possible sleep disruption. Apneas and hypopneas were not a feature of breathing during sleep in these animals. Nonetheless, this animal preparation may be useful for exploring possible drug treatments for obstructive sleep-disordered breathing.

SLEEP-DISORDERED BREATHING resulting from pharyngeal obstruction is a common phenomenon. In humans, such respiratory disorders comprise a pathophysiologic spectrum ranging from high upper airway resistance during sleep to severe obstructive sleep apnea (OSA). OSA is a common disease having considerable morbidity and mortality (7). Presently, no generally accepted pharmacologic treatment for OSA exists. In vitro and in vivo studies have identified classes of drugs, including serotonin agonists (2) and inhibitory amino acid antagonists (6), that may activate hypoglossal motor output. The development of such potential pharmacological therapies has been impeded by the lack of suitable animal models. A suitable animal would spontaneously exhibit obstructive sleep-disordered breathing, with an underlying cause similar to the human disease, i.e., structural narrowing of the pharynx (10).

The English bulldog has been shown to have obstructive sleep-disordered breathing (8), with apneas and hypopneas occurring primarily during rapid-eye-movement (REM) sleep. In this animal, narrow nares and a thickened soft palate contribute to an abnormally narrow upper airway. Recently, Lonergan et al. (12) reported that obese Yucatan miniature pigs have spontaneous sleep-disordered breathing resembling human OSA. However, these observations were based on minimal instrumentation, including airflow detected by nasal CO2 or thermistors and respiratory effort detected with inductance plethysmography. We sought to develop an obese porcine preparation instrumented to allow measurement of respiratory mechanics. We selected the obese Vietnamese pot-bellied (VPB) pig because of its smaller size and more tractable personality.

The purpose of this paper is to describe in detail our chronically instrumented obese VPB pig preparation and to document the effects of sleep on pulmonary gas exchange, respiratory mechanics, and genioglossus muscle activity in these animals.

METHODS

Animals

Four VPB pigs were used, one female and three castrated males. Castrated males were used because of their more manageable behavior. The pigs were fed a standard pig diet (Hog Grower, United Feed) ad libitum, and two of the four received a high-calorie supplement consisting of canola oil and molasses. On both diets, the body weight doubled in 7–9 mo. At the time of study, the average weight of the pigs was 108 kg (range: 103–118 kg), and the average age was 21 mo (range: 18–24 mo). The pigs were –2.5 times the maximum breed standard weight (43 kg) according to the North American Potbellied Pig Association (3). Given an approximate snout-to-tail length of 1.2 m, the body mass index of these pigs would range from 70 to 80 kg/m2. All protocols were approved by the Animal Care Committee at the University of Calgary.

Chronic Instrumentation

Implanted devices. Wire electrodes were used to record the following bioelectric signals related to sleep: electroencephalogram (EEG), neck electromyogram (EMG), and electrooculogram (EOG). A pair of EEG electrodes and a pair of EOG electrodes were constructed from Teflon-coated, single-strand
stainless steel wire (30 AWG, A-M Systems), with the Teflon removed from a 1-cm segment at the end. A pair of EMG electrodes was constructed from Teflon-coated multistrand stainless steel wire (AS 632, Cooner Wire), with the Teflon removed from a 1-cm segment, 3 cm from the end. All wire electrodes plus a ground wire were soldered to a nine-pin stainless steel connector (DE-9P, AMP). A similar pair of genioglossal (GG) EMG electrodes was prepared but was not attached to the connector.

To measure intrapleural pressure (Pip), two types of balloons were used. Both types were connected to 1 m of silicone tubing (0.312-cm ID, 0.625-cm OD, Baxter). The first type was a cylindrical balloon constructed from a latex finger cot, coated with a thin layer of silicone. The volume of air where balloon compliance was highest was determined from the pressure-volume relationship of the balloon, 7 ml, and was injected into the balloon when pressure measurements were made. The second balloon was a square balloon with a flat profile, identical to that described by Smiseth et al. (19) to measure pericardial pressure in dogs. The balloon was inflated with 0.5–1.5 ml of air, it accurately measured negative pressure in the range from 0 to −40 cmH2O in an artificial system (r = 1.0). The balloon was inflated with 1 ml of air during recording sessions. Both types of balloons were evacuated when not in use.

Tracheal pressure was measured with a modified transtracheal oxygen catheter (Scoop, Transtracheal Systems). The catheter, composed of radiopaque polyurethane, was 24 cm long with an outside diameter of 3 mm. A flange, attached 3 cm from the external end, was used to secure the catheter to subcutaneous tissues.

An implantable catheter system, the Vascular-Access-Port (VAP; Access Technologies) was used for chronic vascular catheterization, as has been previously reported in swine (1, 14). This system consisted of a titanium reservoir with a self-sealing rubber septum (TI-AC) and a silicone catheter (71S), 2.3-mm OD, and 60 cm long. The titanium reservoir is placed subcutaneously and can be accessed through the skin with a needle for infusion or withdrawal.

Surgical procedures. After premedication with acepromazine (0.2 mg/kg iv, Ayerst), ketamine (10 mg/kg im, Ayerst), and atropine (0.05 mg/kg im, Vetcom), halothane was administered by mask and the trachea was intubated. The pigs were then mechanically ventilated (Harvard Respirator) and maintained on isoflurane in 100% O2 for the duration of the surgical procedures. Not all techniques were applied in each pig; the instrumentation used in each pig is shown in Table 1.

The VAP was implanted by using a modification of the techniques of Pinto et al. (15) for piglets. The pig was secured in sternal recumbency, and a midline incision was made from the caudal margin of the ears to the rostral margin of the eyes. The skin and fat were retracted to expose the frontal bone. Subcutaneous fat was excised along the incision bilaterally to help subsequent closing of the incision and healing. The internal plate of the frontal bone was exposed in the midline by drilling through the external plate and underlying sinuses by using a rotary tool with a cutting bit (MultiPro395, Dremel). The hole was enlarged into a cavity ~4 cm wide × 5 cm long to provide adequate exposure. Two holes (2 mm in diameter) were drilled bilaterally, 1 cm lateral to the midline, through the internal plate by using a small, round drill bit (Dremel), with care taken not to puncture the dura. An EEG electrode was inserted through each hole, directed laterally between the dura and the bone, and secured in place with tissue glue (Histocryl blue, Braun). The rostral end of the splenius muscle was exposed at the caudal end of the incision, and the EMG electrodes were tied into the muscle. Each wire was tied so that the uninsulated segment formed a loop that lay within the muscle fibers. EOG electrodes were placed behind each eye by inserting the wires into small holes drilled through the orbital wall. The ground wire was inserted laterally into subcutaneous tissue. The cavity in the skull was filled with methyl methacrylate (LVC Bone Cement, Zimmer). The connector was positioned in the cement at the level of the skin, and the incision was closed around the cement and connector.

Table 1. Instrumentation used in each pig

<table>
<thead>
<tr>
<th>Pig</th>
<th>Pig2</th>
<th>Pig3</th>
<th>Pig4</th>
</tr>
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<tbody>
<tr>
<td>EEG</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Neck EMG</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>EOG</td>
<td>X</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cylindrical intrapleural balloon</td>
<td>X</td>
<td></td>
<td></td>
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<tr>
<td>Flat intrapleural balloon</td>
<td>X</td>
<td>X</td>
<td></td>
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<tr>
<td>Tracheal catheter</td>
<td>X</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Genioglossal EMG</td>
<td>X</td>
<td></td>
<td></td>
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<tr>
<td>Vascular access port</td>
<td>X</td>
<td></td>
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</tbody>
</table>

EEG, electroencephalogram; EMG, electromyogram; EOG, electro-oculogram.

To implant the intrapleural balloon, a 6-cm incision was made at the level of the fifth intercostal space. The parietal pleura was punctured at the level of the fourth intercostal space, and a balloon was inserted into the intrapleural space. The position and function of the balloon in the space were confirmed by digital palpation of the balloon in the intrapleural space while the balloon was inflated and deflated. The tubing from the balloon was tethered to subcutaneous tissue at the skin incision site before being tunneled subcutaneously to exit to the dorsum of the pig between the scapulae. A pouch was sutured over the tubing exit site to contain the tubing when not in use.

The tracheal catheter was implanted through a midline incision on the ventral aspect of the neck, and the trachea was exposed. By using an 18-gauge needle, a small puncture was made in the cricothyroid space. A wire guide was inserted through the needle and advanced distally within the trachea. The needle was removed, and the tracheal catheter was inserted over the wire guide. The catheter was passed distally into the trachea for a distance of ~18 cm and was secured by sewing the flange into the subcutaneous tissues. The external end of the catheter exited ventrally through the skin incision and protruded ~2 cm beyond the skin.

The VAP was implanted by first exposing the external jugular vein. The sphenoid catheter was placed into the vessel through a small puncture hole and secured with an occluding tie around the vessel. The reservoir was placed in a subcutaneous “pocket” in the dorsolateral neck. The free catheter end was tunneled subcutaneously to the pocket and attached to the reservoir. The reservoir was then sutured within the pocket and flushed with heparinized saline.

Postoperative care. After the surgery was completed, an antibiotic (gentamicin, 100 mg iv, Schering-Plough) and an analgesic (morphine, 20 mg im, Sabex) were administered. Prophylactic oral antibiotics (amoxicillin, 10 mg/kg, Apotex) were given for 5 days. All incision sites and the tubing exit site were flushed daily with an iodine solution and sprayed.
with a topical antibiotic (Gentocin, Schering-Plough). The
VAP reservoir was penetrated with a noncoring Huber point
needle (Access Technologies) and flushed once per week with
a heparinized saline solution. A stylet in the tracheal catheter
minimized accumulation of secretions within the lumen when
the catheter was not in use. The pigs recovered for 1 wk before
recordings were made. Six weeks after surgery, the pigs were
euthanized by barbiturate injection.

Noninvasive Instrumentation

Airflow measurements. Respiratory airflow was measured
by using a face mask-pneumotachograph system (Fig. 1). A
custom face mask was constructed for each pig, using a
modification of the techniques of Stavert et al. (21) for dogs.
The mask was adapted to a pneumotachograph (3700, Hans
Rudolph) by gluing the top portion of a plastic water bottle
(750 ml, Evian) over the snout end of the mask. The combined
volume of the adapter and pneumotachograph was \( \approx 130 \) ml.
Ports in the adaptor allowed for sampling of expired gases,
recording of mask pressure, and insertion of a small micro-
phone to record snoring. The mask was secured to the face via
four Velcro straps that attached to a vest worn by the pig
(Lomir Biomedical). After six to eight training sessions, each
pig slept in the lab while wearing the face mask-pneumotacho-
graph. A progressive approach to training was most effective,
starting with the body of the mask then adding the adaptor
and pneumotachograph.

To examine the effects of the face mask-pneumotachograph
on respiratory mechanics, PIP was recorded in 1 pig during
non-REM (NREM) sleep for 10 consecutive breaths while the
animal was wearing the face mask-pneumotachograph and
for 10 breaths without the face mask.

Nose-twitch transducer. Episodes of nose twitching were a
prominent feature of REM sleep. To record these, a 0.7 \( \times \)
2.5-cm piezoelectric strip (Night Watch eye sensor, Health-
dyne Technologies) was taped on the upper lip, and the face
mask was placed over the strip. Deformation of the piezoelec-
tric strip by movement of the nose created a voltage that was
amplified and recorded.

Oxygen saturation measurement. Arterial oxygen saturat-
ion (\( S_aO_2 \)) was measured with an oximeter (4700 OxiCap,
Ohmeda) attached to the tail with a flexible sensor attach-
ment. The tail was shaved and rubbed with mentholatum to
promote vasodilation, and the sensor was placed midway
along the tail and secured with tape.

The accuracy of the oximeter in pigs has not been previ-
ously reported. In a separate study using an anesthetized,
nonobese pig, we compared oximeter \( S_aO_2 \) values to those
calculated from arterial blood-gas analysis over a range of
saturation from 60 to 100%. For blood-gas analysis, tempera-
ture corrections were made by using a coefficient for pigs (23),
and \( S_aO_2 \) was calculated by using a pig blood \( O_2 \) affinity curve
(22). Linear regression analysis and a Pearson correlation
coefficient between the calculated and estimated \( S_aO_2 \) mea-
surements for all points were determined.

Experimental Procedures

All recording sessions took place in a normally lit labora-
tory, usually during the day, with no effort made to suppress
normal ambient noise. The pig was placed in a raised box with
Plexiglas sides measuring 1.5 \( \times \) 0.6 m. After the mask and
vest were placed on the pig, the animal was allowed to
accommodate. The pneumotachograph was connected to a
differential pressure transducer (MP-45–15, Validyne), ex-
pired \( CO_2 \) was measured by a capnometer (4700 OxiCap,
Ohmeda), and snoring was recorded with a small microphone
(33–1063, Realistic). EMG potentials from the GG electrodes
were recorded by using single-point test clips.

The intrapleural balloon was inflated and connected to a
differential pressure transducer (MP-45–32, Validyne). The
tracheal catheter was connected by tubing (3-mm ID, 6-mm

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**Fig. 1.** Custom respiratory face mask

for pigs. Body of mask was constructed

from layers of liquid latex. Inner dia-

phragm of rubber fits behind snout and

mouth to provide a competent seal.

Velcro straps attach to a vest worn by

pig. A 16-gauge needle attached to sam-

ing line was inserted between plastic

adaptor and pneumotachograph to mea-

sure expired \( CO_2 \).
OD) to a differential pressure transducer (PM5, Statham). Both \( P_{ip} \) and tracheal pressures were referred to mask pressure. During data collection, all signals were amplified, filtered, and recorded on paper by a polygraph (model 7D, Grass Instruments). All signals were also collected on FM tape (7DS, Racal) for off-line analysis.

To ensure accurate measurements of resistance, the airflow and pressure signals must be in phase. Spectral analysis of typical airflow and transpulmonary pressure signals indicated that the frequency content of these signals was below 5 Hz. The relative phase of the pressure and airflow signals was evaluated as described by Jackson and Vinegar (11). At a frequency of 12 Hz, the pressure signal lagged the flow signal by 12 degrees. Therefore, phase lag at frequencies below 5 Hz, if any, was considered negligible.

Data Analysis

Wakefulness, NREM sleep, and REM sleep were identified by manual scoring, according to published criteria for pigs (18). A single recording session was analyzed for pig 1, recording sessions from two different days were analyzed for pigs 2 and 3, and from seven different days for pig 4. From each recording session, one or two 60- to 70-s epochs of data each were analyzed for wakefulness, NREM sleep, and REM sleep. If two epochs of the same state were analyzed within a recording session, they were separated in time by at least one change in sleep state. An average of 68 breaths/pig (range: 22–148) was analyzed in total for each condition (wakefulness, NREM, REM).

Airflow was integrated to obtain inspired and expired tidal volume, and minute ventilation (\( V_E \)) was calculated. Average \( S_aO_2 \), was calculated for each epoch of data, incorporating a lag of 7 s to correct for the time delay associated with measuring \( S_aO_2 \). End-tidal \( PCO_2 \) (\( PETCO_2 \)) was measured as the peak value of \( CO_2 \) at the end of expiration. Snoring was assessed visually, with an increase in snoring defined as an increase in the frequency (i.e., the number of breaths associated with a snore) and/or amplitude of the snores.

The transpulmonary or tracheal pressure and airflow signals were digitized at a sampling frequency of 100 Hz by using a personal computer, analog-to-digital board (CIO-AD16, Computer Boards) and commercial data-analysis software (Datapac, Run Technologies). Resistive pressure was computed at each 10-ms interval during inspiration by subtracting lung elastic pressure from transpulmonary pressure, as described by Mead and Whittenberger (13). Total pulmonary resistance (\( RL \)) was then calculated by dividing resistive pressure by flow and averaged throughout inspiration for each breath (\( RL_b \)), excluding the fast transition between the end of inspiration and the beginning of expiration. To calculate upper airway resistance, tracheal pressure was divided by airflow at each sampling point throughout inspiration, and an average resistance throughout inspiration (\( R_{aw} \)) was calculated.

The GG EMG signal was amplitude demodulated by a modified Bessel filter with a 50-ms time constant (16), then digitized at a sampling frequency of 100 Hz, and smoothed with a 300-ms window. For each breath, peak inspiratory GG EMG (GG EMGpeak) and average expiratory GG EMG (GG EMGE) were calculated. Both GG EMGpeak and GG EMGE were expressed as a percentage of the average value obtained during wakefulness.

Mean values of the dependent variables (\( V_E \), \( PETCO_2 \), \( S_aO_2 \), \( RL_b \), \( R_{aw} \), GG EMGpeak, and GG EMGE) for each pig during wakefulness, NREM sleep, and REM sleep were calculated by averaging across epochs from the same recording session as well as across recording sessions from different days. The mean values were compared during wakefulness, NREM sleep, and REM sleep by using one-way ANOVA. To satisfy the assumptions of normality of distribution and homogeneity of variance, rank transformations of the data were done before analysis of each variable. Dunn’s method was used for post hoc analysis.

RESULTS

Performance of Instrumentation

Adequate EEG, neck EMG, and GG EMG signals were maintained in all pigs throughout the instrumentation period. All pigs developed minor local infections ~1 mo postoperatively at the electrode connector site. However, wound healing and general health of the animal were not affected. The EOG signals failed to exhibit phasic deflections during REM sleep. The piezoelectric transducer proved to reliably detect episodes of nose twitching, which occurred only during REM sleep and were seen in all pigs.

The performance of the flat intrapleural balloon was superior to that of the cylindrical intrapleural balloon. Pressure measurements from the latter deteriorated after ~4 wk, as indicated by a reduction in the magnitude of pressure swings during respiration and an increasingly positive mean value of \( P_{ip} \). A fibrous capsule was noted postmortem. The flat balloon provided a stable signal throughout the study period and, postmortem, no local pleural fibrosis was evident. An infection at the site of the intrapleural balloon and/or tubing occurred in one pig.

Airway secretions occasionally occluded the tracheal catheter, as indicated by a damping or complete absence of tracheal pressure swings. Flushing with saline and/or the insertion of a metal guide wire into the catheter reversed the occlusion. Otherwise, the tracheal pressure measurements were stable for the study period.

The VAP remained patent for flushing during the study period. The primary purpose of the VAP in this preparation was drug administration; therefore, we did not assess the ability to withdraw blood samples from the VAP over the study period.

The wearing of the face mask had little effect on the maximal negative pulmonary pressure observed during NREM sleep compared with when the pig was not wearing the face mask. The average maximal negative pulmonary pressure for 10 breaths without the face mask was 26.4 ± 1.2 cmH2O and was 27.8 ± 1.2 cmH2O with the face mask on.

\( S_aO_2 \) measured by the oximeter was linearly related to \( S_aO_2 \) calculated from arterial blood samples over the range of 99–65% (\( r = 0.996 \) for all data from the 2 trials). Corrected \( S_aO_2 \) was calculated from observed oximeter \( S_aO_2 \) as follows

\[
\text{Corrected } S_aO_2 = -50.64 + (1.59 \times \text{observed } S_aO_2)
\]

In darkly pigmented pigs (pigs 1, 2, and 4), an adequate signal was difficult to maintain, and the \( S_aO_2 \) data for pigs 2 and 4 were not analyzed because of the instability. In the one lightly pigmented pig (pig 3), an adequate signal was maintained for several hours without adjustment of the probe.
Discrepancies were apparent between changes in \( \dot{V}E \) and changes in \( P_{ETCO_2} \), for example, \( \dot{V}E \) decreased during sleep in pigs 2 and 3 with either no change or a fall in \( P_{ETCO_2} \). We suspect that our measurements of \( P_{ETCO_2} \) do not accurately estimate arterial \( PCO_2 \), as the expired waveform lacks the required criterion of a plateau. Therefore, changes in the difference between the measured \( P_{ETCO_2} \) and the true arterial \( PCO_2 \) may explain the discrepancies between \( \dot{V}E \) and \( P_{ETCO_2} \), although we cannot rule out a change in metabolic rate or in flow-perfusion relationships.

**Fig. 2.** Typical signals during wakefulness (A), non-rapid-eye-movement (NREM) sleep (B), and rapid-eye-movement (REM) sleep (C). Airflow (\( \dot{V} \)), intrapleural pressure (\( P_{Pip} - P_{MASK} \)) cmH\(_2\)O, and end-tidal fraction expired CO\(_2\) (\( FETCO_2 \)) were reproduced from digitized data; all other signals were from a curvilinear polygraph. For \( \dot{V} \), inspiration is positive. \( SaO_2 \), Arterial oxygen saturation; MIC, microphone; EEG, electroencephalogram; EMG, neck electromyogram.
Typical signals during wakefulness and sleep are shown in Fig. 2. Typical recordings of tracheal pressure and GG EMG are shown in Fig. 3. The general characteristics of breathing during NREM sleep appeared similar to those of wakefulness, except for an increase in inspiratory snoring. No apneas or hypopneas were observed during NREM sleep.

Fig. 3. Typical recordings of airflow, tracheal pressure, and genioglossal (GG) EMG during wakefulness, NREM sleep, and REM sleep. Note marked suppression of both phasic inspiratory activity and tonic expiratory activity with sleep. Arb., arbitrary.

Fig. 4. Example of arousal from REM sleep associated with breathing events. Arrow, occurrence of arousal. Before arousal, hypopnea associated with phasic REM episode (indicated by nose twitching) is observed with a related fall in SaO2. Breaths immediately preceding arousal are associated with a decrease in peak inspiratory flow, flat or decrementing inspiratory flow profiles, increasingly negative pulmonary pressure, and snoring. After arousal, a large increase in peak inspiratory flow is observed.
During REM sleep, breathing became irregular, peak inspiratory flow decreased, and the inspiratory flow profile became flat or decrementing. During some breaths with a flat or decrementing inspiratory flow profile, transpulmonary or tracheal pressure became increasingly negative, suggestive of inspiratory flow limitation. No prolonged apneas were observed during REM sleep. During phasic REM, as indicated by episodes of intense nose twitching, transient drops in $S_{a}O_{2}$ occurred, associated with a highly irregular respiratory pattern. Respiratory fluctuations in transpulmonary or tracheal pressure decreased during these episodes, indicating a decrease in inspiratory motor output during these phasic REM events. REM sleep was always terminated by arousal and an associated increase in inspiratory airflow, often preceded by a hypopnea and/or inspiratory flow limitation (Fig. 4).

$V_{E}$ tended to decrease during sleep, the decrease being significant during NREM and REM sleep in pigs 2–4 (Table 2). Pigs 1, 2, and 4 were hypercapnic during wakefulness and sleep. $PETCO_{2}$ did not change during sleep in pigs 1 and 3 but fell during NREM and REM sleep in pigs 2 and 4. $S_{a}O_{2}$ in pig 1 decreased during NREM and REM sleep, whereas $S_{a}O_{2}$ in pig 3 decreased only during REM sleep.

$R_{L}$ or $R_{UAW}$ increased during NREM sleep compared with wakefulness in all pigs (Fig. 5). In pigs 3 and 4, an additional increase in resistance occurred during REM sleep. In pigs 1 and 2, resistance tended to fall during REM sleep compared with NREM sleep.

Large phasic inspiratory bursts in GG EMG, as well as substantial tonic expiratory activity, were observed during wakefulness in pig 4 (Fig. 3). Both decreased during NREM sleep and reached low levels during REM sleep. For all the breaths analyzed in pig 4, GG EMG peak and GG EMG E during NREM sleep were significantly less than during wakefulness (50 ± 23% of wakefulness values for GG EMG peak and 43 ± 30% for GG EMG E) and during REM sleep (21 ± 25% of wakefulness values for GG EMG peak and 12 ± 26% for GG EMG E).

### DISCUSSION

We describe techniques for chronic and noninvasive instrumentation of obese pigs to study ventilation during wakefulness and sleep. The obese VPB pigs exhibited sleep-disordered breathing characterized by high inspiratory resistance, snoring, inspiratory flow limitation, and possible sleep disruption.

Methodological Considerations

The instrumentation described in this study performed satisfactorily throughout the instrumentation period with three exceptions: 1) the cylindrical balloon caused a pleural reaction and did not provide stable values of $P_{ip}$; 2) the EOG electrodes failed to reflect REM episodes for unclear reasons; and 3) $S_{a}O_{2}$ was not measurable from the tails of darkly pigmented pigs. We circumvented the first two exceptions by using a flat intrapleural balloon and by recording nose twitch, respectively.

Although use of a face mask could have affected respiratory mechanics by compression of soft tissues, these potential effects were minimal, as the face mask caused no detectable increase in respiratory fluctuations in transpulmonary pressure, snoring, or change in rib cage-abdomen movement. Values of inspiratory resistance calculated from transpulmonary pressure were similar to those calculated from tracheal pressure, which suggests that resistance of the upper airways is the dominant contributor to total $R_{L}$. Airflow resistance of the lower airways in the pig has been reported to be 2.7 ± 0.3 cmH$_{2}$O·l$^{-1}$·s$^{-1}$ (20), which indeed is a small proportion of total $R_{L}$ calculated in the three pigs ($\approx$ 20–30 cmH$_{2}$O·l$^{-1}$·s$^{-1}$ during wakefulness). The sleep-related changes in $R_{L}$ and $R_{UAW}$ were also of the same magnitude, suggesting that these changes are due

### Table 2. Respiratory variables during wakefulness, NREM sleep, and REM sleep in each pig

<table>
<thead>
<tr>
<th>Pig</th>
<th>Wakefulness</th>
<th>NREM Sleep</th>
<th>REM Sleep</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pig 1</td>
<td>$V_{E}$, l/s</td>
<td>5.0 ± 1.0</td>
<td>4.8 ± 0.5</td>
</tr>
<tr>
<td>Pig 1</td>
<td>$PETCO_{2}$, Torr</td>
<td>47.7 ± 12</td>
<td>46.7 ± 0.9</td>
</tr>
<tr>
<td>Pig 1</td>
<td>$S_{a}O_{2}$, %</td>
<td>94.9 ± 1.5</td>
<td>90.1 ± 0.4*</td>
</tr>
</tbody>
</table>

Values are means ± SD. NREM, non-rapid-eye-movement; $V_{E}$, minute ventilation; $PETCO_{2}$, end-tidal PCO$_{2}$; $S_{a}O_{2}$, arterial O$_{2}$ saturation. *Different from wakefulness, P < 0.05. †Different from NREM sleep, P < 0.05.
primarily to increased resistance of the upper airways. Similarly, in humans, Hudgel et al. (9) reported a negligible contribution of lower airways and lung tissue resistance to changes in total respiratory system resistance during sleep; the increase in resistance with sleep was due almost entirely to changes in resistance above the larynx. In conclusion, RL appears to be a good estimator of upper airway resistance.

The Effects of Sleep

Our findings of high resistance without apneas in the obese VPB pigs differ from the obstructive apneas and hypopneas in NREM and REM sleep in obese Yucatan miniature pigs reported by Lonergan et al. (12). In addition to breed differences, the differences in methods used to quantitate respiratory effort and airflow may contribute to the dissimilar findings. The present findings also differ from another model of sleep-disordered breathing, the English bulldog, which experiences central and obstructive apneas during REM sleep (8). The reason for apneas in the bulldogs but not in the VPB pigs is unclear but may relate to differences in the underlying pathology. Airway obstruction in the bulldog derives from an abnormal upper airway anatomy, which may not be present in the obese pig.

Because of the absence of obstructive apneas, the sleep-disordered breathing in the obese VPB pig does not resemble OSA per se but does resemble high upper airway resistance syndrome. This syndrome is characterized by abnormally high upper airway resistance without apneas or hypopneas and is associated with frequent arousals that lead to sleep fragmentation and excessive daytime sleepiness (4). We were only able to study pigs when they were obese, as they were apparently hypersomnolent and much more tolerant of the face mask, whereas when the pigs were lean they would not sleep in the laboratory or wear the face mask. We hypothesize that the apparent hypersomnolence exhibited by the obese pig was a result of breathing-related sleep disruption (i.e., Fig. 4).

The GG, a tongue protruder, appears to play an important role in patients with obstructive sleep-disordered breathing (5). A sleep-related decrease in GG EMG is thought to play a pathogenic role in pharyngeal collapse (17). Similarly, we describe a substantial sleep-related suppression of GG EMG during sleep in the obese pig, which is likely related to the rise in resistance also seen during sleep.

In conclusion, we describe methods for chronic instrumentation of the obese VPB pig for the investigation of respiratory function during sleep. This animal displays spontaneous sleep-disordered breathing similar to high upper airway resistance syndrome and therefore may be a useful model of obstructive sleep-disordered breathing. This animal preparation may be useful for studying the pathophysiology of high upper airway resistance during sleep and for exploring possible pharmaceutical treatment strategies.

The authors thank Heather Finch and Lorraine Poulton for surgical assistance.

This research was supported by the Respiratory Health Network of Centres of Excellence, Inspiraplex. S. A. Tuck was supported by an Alberta Heritage Foundation for Medical Research studentship.

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