TRANSLATIONAL REVIEW

Anatomy and Bronchoscopy of the Porcine Lung

A Model for Translational Respiratory Medicine

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Abstract

The porcine model has contributed significantly to biomedical research over many decades. The similar size and anatomy of pig and human organs make this model particularly beneficial for translational research in areas such as medical device development, therapeutics and xenotransplantation. In recent years, a major limitation with the porcine model was overcome with the successful generation of gene-targeted pigs and the publication of the pig genome. As a result, the role of this model is likely to become even more important. For the respiratory medicine field, the similarities between pig and human lungs give the porcine model particular potential for advancing translational medicine. An increasing number of lung conditions are being studied and modeled in the pig. Genetically modified porcine models of cystic fibrosis have been generated that, unlike mouse models, develop lung disease similar to human cystic fibrosis. However, the scientific literature relating specifically to porcine lung anatomy and airway histology is limited and is largely restricted to veterinary literature and textbooks. Furthermore, methods for *in vivo* lung procedures in the pig are rarely described. The aims of this review are to collate the disparate literature on porcine lung anatomy, histology, and microbiology; to provide a comparison with the human lung; and to describe appropriate bronchoscopy procedures for the pig lungs to aid clinical researchers working in the area of translational respiratory medicine using the porcine model.

Keywords: porcine; bronchoscopy; respiratory; translational; lung

The domesticated pig has made a significant contribution to the improvement of many areas of human health over the past number of decades and has been used as a source of biological material for experimental and therapeutic purposes and as a large-animal model for veterinary and medical studies (1). In the last two decades, pigs have been genetically modified for a variety of commercial, agricultural, and biomedical purposes (2–6). Recently, a major limitation with the porcine model compared with rodent models has been overcome by the generation of gene-targeted pigs and by the publication of the pig genome (7). These developments have significantly increased the importance of the porcine model in biomedical research.

In the field of respiratory medicine, the porcine model is becoming an increasingly important bridge between traditional small laboratory animal models and human medicine. A major advance was made with the generation of the porcine cystic fibrosis (CF) model. CF is an autosomal recessive disease caused by mutations in the gene encoding the CF transmembrane conductance regulator (CFTR) anion channel (8, 9). Patients develop lung, pancreatic, intestinal, vas deferens, and hepatic disease. Suitable animal models had not been previously available to study the pathogenesis of this disease (10, 11). In 2008, heterozygote male piglets with null allele and ΔF508 alleles (the most common CFTR mutation) were produced for the first time (12). This targeted disruption of both CFTR alleles replicated abnormalities seen in newborn humans with CF (13). Subsequent analysis of CF pigs has shown that the CF pig lung has a defect in eradicating bacteria within hours of birth and that CF pigs spontaneously develop lung disease that largely replicates that in humans, including inflammation, remodeling, mucus accumulation, and infection (14). It is hoped that these apparent similarities between human and

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porcine CF lungs will help to provide further insight into the pathogenesis of this disease over time (15).

In addition to CF, the porcine lung model has been used in other areas of translational pulmonary research. For example, it is being used in the study of respiratory diseases, such as chronic obstructive pulmonary disease, pneumonia, and ventilator-induced lung injury (16–18). The porcine lung is also being used in anesthesia, in critical care and lung transplantation research, and in studies of airway structure, function, and microbiome (19–23). It is also being increasingly used in the growing field of interventional pulmonology, where the porcine airway has been used to compare and develop diagnostic and therapeutic endobronchial techniques (24–27).

In this review we present a collation of the disparate literature on porcine lung anatomy, histology, and microbiology and provide a comparison with the human lung. We propose a new nomenclature for the porcine airways based on similar systems used for other animals. We also describe appropriate bronchoscopy procedures for porcine lungs, including anesthesiology, with the ultimate aim of aiding clinical researchers aiming to use the porcine model in the area of translational respiratory medicine.

Lung Anatomy

In contrast to human respiratory bronchioles, which are often connected to two or three primordial alveolar ducts at birth, neonatal porcine respiratory bronchioles are typically connected to paired alveolar ducts (28). Porcine alveoli multiply rapidly in the first 2 to 4 weeks of life, whereas similar development occurs over the first 3 years of human life (28, 29). The average length of the perinatal porcine acinus (~ 5 mm) is shorter than the acinus of the human infant (~ 11 mm) (28). The morphological structure and distribution of the porcine airways vary according to the age and breed of pig but are broadly similar to the human lung. The porcine trachea is notably longer and more cartilaginous than the human trachea (Figure 1). For example, Belgian landrace pigs (6–8 wk old, 20–25 kg) have a tracheal length of 15 to 20 cm and a tracheal wall structure comprising of 32 to 45 hyaline cartilaginous rings, compared with the mean adult human tracheal length of 12 cm with 16 to 20 cartilaginous rings (30, 31). Porcine airways are more cartilaginous in structure, but a similar number of bronchial generations (n = 23) have been identified in humans and pigs (28, 32, 33). The general decrease in diameter and length seen with bifurcations of the human bronchial tree is also observed in porcine airways (33, 34). However, in contrast to the human airway tree, which has a bipodial branching pattern, the porcine airway tree has a monopodial branching pattern where each larger "parent" bronchus gives rise to smaller side-branches (bronchi) that branch off at obtuse angles (35).

The lungs of pigs and humans are highly lobulated, with well-defined pulmonary lobules demarcated by interlobular septae. In the human lung, the collagenous component of these interlobular septae is incomplete, and interalveolar pores (of Kohn) and other communicating channels have been described in alveolar walls (36, 37). These factors may lead to collateral ventilation between lobules, especially in emphysematous lungs (38). In contrast, the collagenous component of the porcine interlobular septa is more complete, and collateral ventilation is substantially less likely (39).

The lobar and bronchial anatomy of pigs is similar to that of humans. The porcine left lung is similar to the human left lung because it consists of a cranial lobe and a caudal lobe. The left cranial lobe bronchus divides into cranial and caudal segmental bronchi. This is similar to the lingular and left upper lobe divisions in humans. The left caudal bronchus divides into four dorsal and four ventral segmental bronchi, which ventilate the left caudal lobe. In contrast to the human right lung, which has three lobes, the porcine right lung is divided into four lobes (cranial, middle, accessory, and caudal). The right cranial lobe is served by a cranial lobe (tracheal) bronchus, which arises from the right wall of the trachea, proximal to the bifurcation of the trachea (Figure 1). The cranial lobe bronchus then subdivides into cranial and caudal segmental bronchi (40). Tracheal bronchi are anatomical variants found in humans, usually arising within

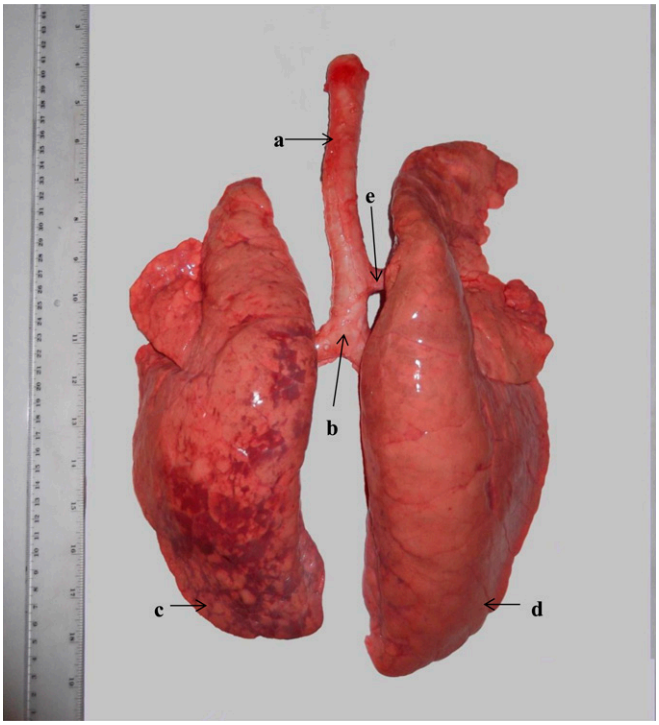

Figure 1. Porcine lung anatomy. Photograph (dorsal aspect) of lungs from a pig (age, ~ 22 wk; size, ~ 105 kg) showing trachea (*a*), carina (*b*), left lung (*c*), right lung (*d*), and cranial lobe bronchus (*e*). Standard 12-inch/30-cm ruler shown for scale.

2 cm of the carina and up to 6 cm of the carina (41). The porcine right middle bronchus, which ventilates the right middle lobe, arises just caudal to the bifurcation of the trachea on the ventrolateral side. The right accessory bronchus, which ventilates the small right accessory lobe, arises just caudal to the middle lobe bronchus and passes ventromedially (40). The right accessory lobe is situated at the base of the heart and diaphragm and surrounds the terminal intrathoracic portion of the caudal vena cava (30). The right caudal lobe is ventilated by the right caudal bronchus, which divides into four dorsal and four ventral segmental bronchi (42).

The bronchial anatomy of the pig has been classified into dorsal, lateral, ventral, and medial bronchiolar systems, each named according to the order in which the bronchioles come off the main bronchus on each side (40). Here we propose a system similar to that which has been used in clinical and research fields for dogs, horses, cats, and humans (43–46). Each lobar bronchus is named according to the side of the lung it ventilates and according to the order in which it is encountered bronchoscopically (Figure 2). The right cranial lobe bronchus is therefore named RB1, the middle lobar bronchus RB2, accessory lobe bronchus RB3, and right caudal lobe bronchus RB4. The left cranial and caudal bronchi are named LB1 and LB2, respectively. Branches from the lobar bronchi are named according to their predominant orientation (dorsal [D], ventral [V], cranial [Cr], caudal [Cd]) in order of proximal to distal, with lowercase letters (a, b, etc.) representing subsegmental bronchi (e.g., RB1, RB1Cr1, RB1Cr1a, etc.).

Other morphological similarities exist between porcine and human lungs. Both species have abundant but varying quantities of visceral pleural collagen, leading to thicker pleural membranes than other species (37). The visceral pleural vascular supply is from the bronchial arteries in both species, and visceral pleural lymphatics are extensive.

Histology

Histological structure is similar in the human and porcine respiratory tract. The type of epithelium lining the airway lumen of pigs depends on the level of the respiratory tract. The porcine nasal passages are lined by several types of epithelia, including squamous epithelium, ciliated pseudostratified columnar epithelium (respiratory epithelium), nonciliated columnar epithelium (transitional epithelium), and olfactory epithelium (47). Pseudostratified respiratory epithelium lines the large airways, and ciliated columnar epithelium lines the small bronchi and bronchioles (47). In contrast to other animals (e.g., dogs), large numbers of submucosal glands are found in the bronchi of pigs (48). This is an important consideration for the development of large animal models of airway inflammatory disease and CF. Submucosal glands are associated with the smooth muscle layer at all bronchial levels on the luminal and cartilaginous sides. The remaining tissue adjacent to the glands consists of cartilage and associated connective tissue, and this probably accounts for more than 80% of the bronchial wall mass. The entire noncartilaginous layer lining the bronchial lumen is rich in submucosal glands.

Differentiation of porcine bronchial epithelia into ciliated cells, goblet cells, and basal cells by 80 days gestation has been reported (15, 49). Primordial submucosal glands have been observed by 92 days gestation and continue to develop in maturation and number through gestation and into the postnatal period; this is comparable to gestational and postnatal developmental regulation observed for human submucosal glands (49, 50). An abrupt decrease in submucosal gland concentration has been identified in smaller airways beyond the bronchial–bronchiolar junction, and porcine submucosal glands have been classified according to the morphology of the terminal collecting ducts into antral, linear, or convoluted ducts (51–53). All three subtypes demonstrate a secretory response to acetylcholine, although differences in the level of secretory activity between these subtypes have been found (53). The epithelia of the porcine airway have a relatively high permeability to water, and porcine tracheal epithelia spontaneously absorb liquid (15, 54). Primary cultures of porcine and human airway epithelia also show quantitatively similar electrolyte transport (55). These factors are significant, particularly in the context of porcine lung models of CF.

Microbiology

Porcine respiratory pathogens have been extensively studied due to their potentially significant impact on commercial pig production. Consequently, most studies relate to animals reared on commercial farms and not those reared in laboratory environments. Farm-related factors, such as the type (indoor or outdoor) of housing, herd size, and vaccination rate, may affect the respiratory health (and airway microbiology) of pigs (56, 57). The sourcing of pigs is therefore a significant consideration for all researchers using the porcine airway model. Animals sourced from farms with low respiratory disease rates or those raised in controlled laboratory environments may therefore be required for some research studies where high levels of animal health are a prerequisite.

Gram-positive and gram-negative bacteria can cause respiratory disease in pigs and humans. However, humans are not normally susceptible to the bacterial pathogens that commonly cause respiratory illness in pigs, such as *Haemophilus parasuis*, *Mycoplasma hyopneumoniae* *Streptococcus suis*, *Pasteurella multocida*, and *Actinobacillus pleuropneumoniae* (58, 59). *Pseudomonas aeruginosa*, a common cause of morbidity and mortality in humans with CF, is not commonly found in pigs, although a recent study suggests that pigs with CF may become colonized or infected with this respiratory pathogen (60). Acute infections have also been induced in pigs to evaluate ventilator-acquired pneumonia, with developed models resembling the human model in terms of bacteriology, histology, and pathogenesis (61, 62). *Staphylococcus aureus* is less commonly cultured from porcine airways compared with human airways. However, recent studies conducted on commercial pig farms have identified methicillin-resistant *S. aureus* in porcine respiratory tracts and have shown that those who work closely with pigs are at a significantly higher risk of becoming colonized with this pathogen (63, 64).

Viruses have been shown to be important causes of respiratory illness in

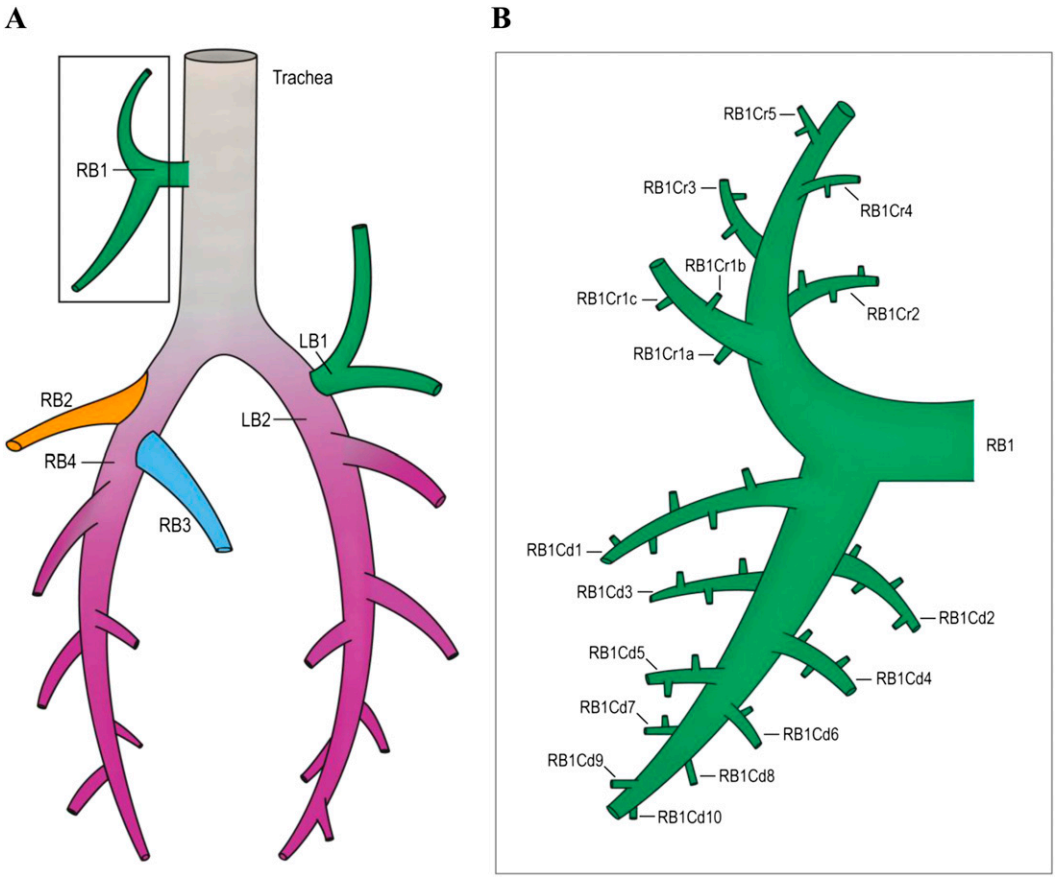

Figure 2. Nomenclature of the porcine airway tree (ventral aspect). (*A*) Larger airways with different colors denoting different lung lobes (*green*, cranial lobes; *orange*, right middle lobe; *blue*, right accessory lobe; *purple*, caudal lobes). LB1, left cranial lobe bronchus; LB2, left caudal lobe bronchus; RB1, right cranial lobe bronchus; RB2, right middle lobe bronchus; RB3, right accessory lobe bronchus; RB4, right caudal lobe bronchus. (*B*) Smaller branches of the right cranial lobe with labels attached. Branches from the lobar bronchi are named according to their predominant orientation (dorsal [D], ventral [V], cranial [Cr], and caudal [Cd]) in order of proximal to distal, with lowercase letters representing subsegmental bronchi (e.g., RB1, RB1Cr1, RB1Cr1a, etc.). Adapted from Reference 93.

humans and in pigs. Common causes of viral respiratory infection in pigs include Porcine Reproductive and Respiratory Syndrome virus, Porcine Circovirus type-2, and Porcine Coronavirus, and common viral respiratory pathogens in humans include Adenovirus, Parainfluenza virus, Coronavirus, and Respiratory Syncytial virus. The Influenza virus is a common cause of respiratory disease in both species. Interspecies transmission of influenza viruses between pigs and humans has been well acknowledged, and the 2009 pandemic H1N1 virus (influenza type A) arose from a reassortment event between a North American triple reassortant swine virus and a Eurasian swine virus (65, 66). To reduce potential bacterial and viral cross-transmission, appropriate hygiene precautions, such as the wearing of gloves when handling pigs and the wearing of facemasks during procedures (e.g., bronchoscopy), are recommended.

Bronchoscopy

Bronchoscopy is an effective means of visualizing the porcine tracheobronchial tree and for performing a variety of diagnostic and therapeutic procedures within the porcine lung.

Bronchoscopic Devices for Pig Studies

As in humans, rigid and flexible (fiberoptic) bronchoscopy can be effectively performed in pigs. Rigid bronchoscopy is less commonly performed and uses a straight, hollow, stainless steel bronchoscope that is available in a variety of sizes. These bronchoscopes are manufactured in various lengths ranging from 33 to 43 cm depending on type and manufacturer, with external diameters of 8 to 14 mm and internal diameters of 7 to 13 mm (67). Rigid bronchoscopes with similar dimensions have been successfully used in pigs. For example, a rigid bronchoscope with an 11-mm external diameter was used for 23-kg Largewhite-Landrace piglets (68). Rigid bronchoscopy is useful for visualization of the trachea and proximal bronchi, hence its use in porcine models of tracheal stenosis (69). The large lumen of the rigid bronchoscope also facilitates stronger suctioning, removal of debris/objects, and the introduction of catheters, lasers, and other equipment. Hence, the rigid bronchoscope has been used in other porcine models of interventional pulmonology, such as stent insertion and photodynamic therapy (68, 70).

Flexible bronchoscopy is more commonly used in pigs for commercial and research purposes. A flexible bronchoscope is thinner than a rigid bronchoscope. It contains a fiberoptic system that transmits light from a light source to the tip of the instrument. The image from the tip of the bronchoscope is transmitted back to an eyepiece (fiberscope) through fiberoptic wires. In contrast, other bronchoscopes (videoscopes) have a chip camera at their tip from which a signal is transmitted to a video processor and then to a monitor for viewing. Because fiberscopes do not require a video processor or monitor, they are often more practical for use in field conditions and are usually less expensive than videoscopes. Fiberscopes have been used successfully in a number of anesthetic, critical care, and otolaryngological porcine models (71, 72). They are also frequently used for performing bronchoalveolar lavage (BAL) in pigs (73, 74) and in bronchoscopic examinations of piglets and smaller pigs (75). Videoscopes are commonly used in human bronchoscopy. Although these scopes are considerably more expensive than fiberscopes, the images produced are sharper and more magnified with a higher resolution and wider field of vision (Figure 3). They also have a larger working channel than fiberscopes, thereby facilitating a number of bronchoscopic procedures. For these reasons, videoscopes should be strongly considered for porcine bronchoscopy, especially when an airway procedure or detailed study of the airways is intended. More recently available ultrathin flexible bronchoscopes (external diameter of insertion tube < 3 mm) may be a suitable option for small pigs or for peripheral airway examinations or procedures. Pediatric gastroscopes that have longer insertion tubes ($> 1{,}000$ mm) than bronchoscopes may be used to access the peripheral and distal airways of larger animals.

Sedation and Anesthesia

Anesthesia and intubation is usually required when bronchoscopy is performed in the research setting to facilitate safe and efficient insertion of the bronschoscope into the airway. Pigs have a complex upper airway anatomy, which makes endotracheal intubation and insertion of a bronchoscope without intubation difficult. For example, the mouth of a pig has a narrow opening with a large tongue, the larynx is long and mobile, and the large epiglottis has a free extremity that extends ventrally to the palate (30). The larynx also forms an obtuse angle with the trachea, and the arytenoid

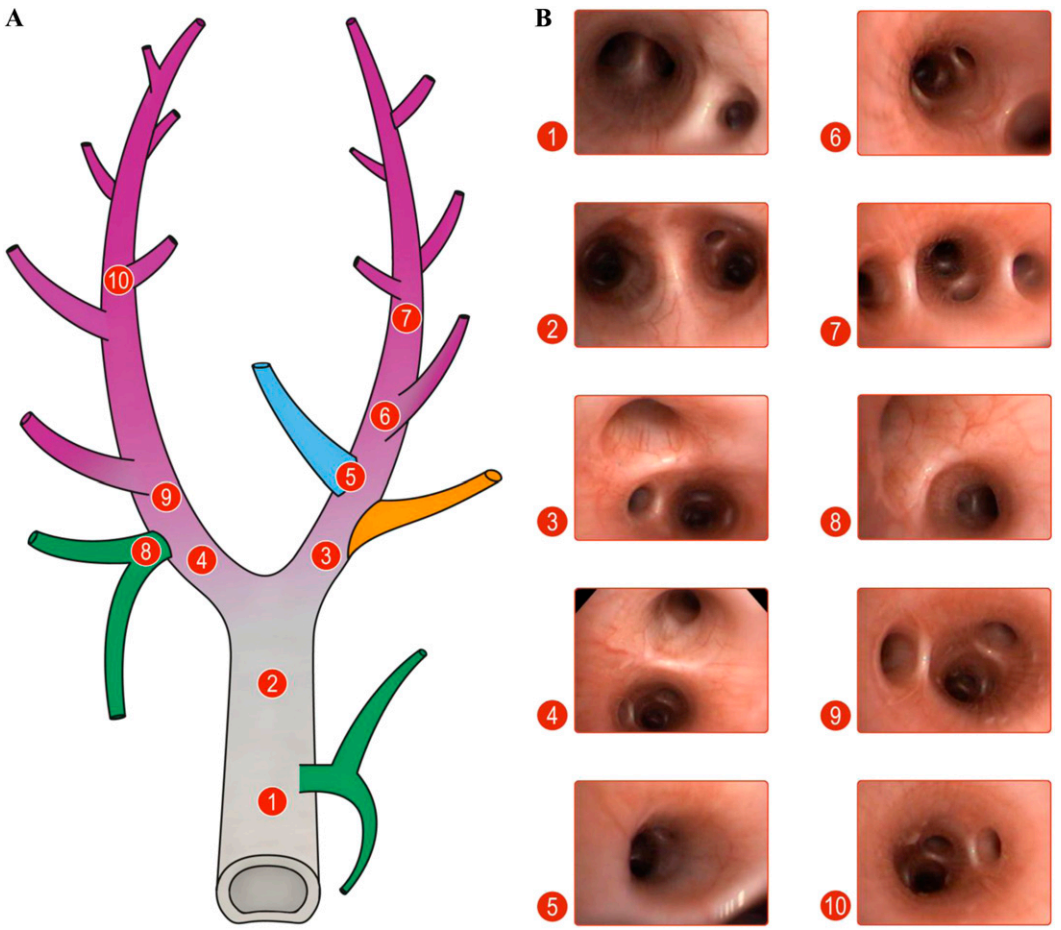

Figure 3. Bronchoscopic images of the porcine bronchial tree (dorsal aspect). (*A*) Each number denotes an airway position at which bronchoscopic images were obtained in our laboratory. Different colors denote different lung lobes (*green*, cranial lobes; *orange*, right middle lobe; *blue*, right accessory lobe; *purple*, caudal lobes). (*B*) Bronchoscopic images obtained by flexible bronchoscopy.

cartilages are large. In addition, the mobile larynx can easily be pushed caudally over several centimeters with a endotracheal tube. This movement could be confused with successful cannulation of the trachea (30). Therefore, attempted insertion of a bronchoscope into the trachea could lead to trauma and consequent morbidity and mortality without adequate anesthesia and endotracheal intubation performed by a skilled operator. A suggested anesthesia protocol used in our laboratory is included in Table 1. Tracheotomy may also be considered as an alternative method of endotracheal intubation, especially in larger animals, for difficult intubations via the oral route or to accommodate larger bronchoscopes with wider instrument channels required to perform interventional procedures such as cryoprobe biopsy (24).

Table 1. Suggested Protocol For Anesthetizing Pigs for Bronchoscopy

Procedure	Technique/Method
Preparation	Fast from food for ⩾ 6 h Ensure adequate hydration and warmth.
Intramuscular premedication combination	Detomidine 0.1 mg/kg; butorphanol 0.2 mg/kg, and ketamine 5 mg/kg; can be mixed in one syringe and administered together
Alternative intramuscular premedication combination	Midazolam 0.2–0.5 mg/kg, ketamine 5–10 mg/kg, and atropine 20–40 μg/kg; can be mixed in one syringe and administered together
Intramuscular premedication administration	The neck (behind the ear) is the preferred site. The rump muscles are readily accessible, but long needles (at least 4 cm) should be used for injections.
Intravenous access	Marginal ear vein*
Induction of anesthesia	Propofol 1–2 mg/kg to effect
Intravenous slowly	Administer 1 mg/kg slowly and top-up as required.
Endotracheal Intubation	Position pig in sternal, dorsal, or lateral recumbency (as preferred by the operator). Hold the mouth open using strong bandages (the neck is held in line with the spine but is not overextended). Insert a laryngoscope with a long straight blade (such as a Magill size 4) to visualize the larynx.† Spray the larynx with lidocaine (without adrenaline), approximately 0.5 mg/kg, to help reduce the incidence of laryngeal spasm and aid intubation. After 1–2 min, an ETT may be passed under direct visualization into the larynx.‡ –Once it engages the laryngeal wall, rotate the ETT through 90° while passing into the trachea and then rotating back If the ETT cannot be passed after one or two attempts, a long bougie may be used over which the ETT is threaded into the trachea. Guide for size of ETT < 20 kg up to 6 mm I.D. 20–50 kg up to 9 mm I.D. > 50 kg up to 14 mm I.D.
Maintenance of anesthesia	Isoflurane or sevoflurane in oxygen ± IPPV§ Propofol infusion intravenously 0.2–0.3 mg/kg/min, oxygen ± IPPV Cover pig during anesthesia with blanket to prevent hypothermia.
Analgesia	NSAIDs (e.g., meloxicam, ketoprofen). Opioids

Definition of abbreviations: ETT, endotracheal tube; I.D., internal diameter (of the endotracheal tube); IPPV, intermittent positive pressure ventilation; NSAIDs, nonsteroidal anti-inflammatory agents.
*A 5% emulsion preparation containing 2.5% each of lidocaine/prilocaine is well absorbed there and can be applied to desensitize the skin.
†Immediately after induction of anesthesia, the epiglottis is usually trapped behind the soft palate, and this should be released to allow the larynx to be visualized.
‡Care must be taken to avoid traumatizing the sensitive laryngeal tissue, which will cause postanesthetic swelling and obstruction.
§Pigs usually breathe adequately under anesthesia. However, depending on the positioning of the pig, the procedure, anesthetic agents, and duration of anesthesia, it may be advantageous to provide IPPV. Tidal volumes of 10–15 mg/kg will usually maintain normocapnia.

Bronchoscopic Procedures in Pigs

Bronchoscopy is generally performed in pigs to obtain diagnostic samples or to evaluate new therapeutic interventions. Methods used in pigs for diagnostic sampling include lung lavage, endobronchial biopsies, and transbronchial biopsies.

Lung lavage. Lung lavage is performed during bronchoscopy in pigs when samples are required for microbiological, cytological (total cell count and differential), and immunological purposes. Methods of lung lavage in pigs are transtracheal lavage, endotracheal lavage, and BAL. BAL appears to have a higher recovery rate of fluid and higher microbiological and cytological diagnostic yields (total cell count) than transtracheal and endotracheal lavage (74). However, contamination of lavage samples with commensal organisms and environmental contaminants is lower in transtracheal lavage (74). Transtracheal and endotracheal lavage are more commonly performed in commercial practice because they require less equipment and are more practicable. However, BAL is more commonly used for research purposes.

BAL differs from a bronchial wash (in which 10–20 ml of sterile saline is introduced via bronchoscope into an airway and quickly aspirated) by providing a predominantly alveolar sample rather than a bronchial sample. BAL samples are therefore less likely to be contaminated with upper airway commensal organisms and provide a better sample for cytological analysis. Recommendations have been made for BAL procedures in humans, and a standardized protocol has been suggested for BAL in pigs (73). In humans, the optimum location for BAL is in the right middle lobe or lingular bronchi due to anatomical accessibility and a 20% higher return of fluid (76). In pigs, BAL can be efficiently performed in any targeted area of interest within the lung, such as the caudal lobes or right middle lobe (61, 75). In humans, the volume of fluid returned can be quite variable. In pigs, this volume appears to be higher, with expected volumes above 80% when BAL is performed in the right cranial lobe (73). Respiratory disease (e.g., infection) may decrease the volume of BAL fluid (BALF)

recovered due to increased alveolar membrane permeability and reduced lung elasticity (73). However, recovery rates of BALF in pneumonic pigs appear to remain relatively high (~ 75%) (77). The dwell time required to aspirate fluid should be kept to a minimum, and an average dwell time of 30 seconds has been suggested in previous studies in pigs (73, 78).

Previous studies suggest that an elevated polymorphonuclear granulocyte (neutrophil) count above 8% can differentiate healthy pigs from pigs with respiratory disease (79, 80). A BALF cell differential reference range for healthy pigs has also been suggested, with alveolar macrophages representing 58 to 100% of cells, lymphocytes 0.01 to 16% of cells, and neutrophils 0.01 to 9% of cells (73). A previous study also compared the BALF cell differential in high-health pigs (i.e., pigs weaned from a swine farm with minimal disease) and low-health pigs (i.e., pigs weaned from a commercial farm with a 70% prevalence of macroscopic pneumonia lesions at slaughter) (75). The cell differential of the high-health pigs and was comparable to that of healthy nonsmoking humans, although porcine BALF contains a higher number of cells (Table 2).

Bronchial brushing. Bronchial brushing is used to collect cytological samples from the luminal surface of the airway for cytological and microbiological analysis. Nonprotected and protected brushes are used for this procedure. Nonprotected brushes may be open or enclosed within an open-ended sheath, which can be advanced or retracted by the bronchoscopist's assistant on request (81). The brush is not protected from upper airway contamination within the bronchoscope because the sheath is open ended, but cytological samples are protected from being lost during removal of the catheter and brush from the bronchoscope. Protected specimen brushes (PSBs) are also available and are primarily used for collection of samples for microbiological analysis. The PSB is enclosed within two telescoping catheters to ensure that any bacteria collected is from the lower respiratory tract and that it is not an upper airway contaminant (81). A study using pigs suggests that bristle diameter and not the diameter or length of the whole brush determines the efficacy of bronchial brush cell collection (82). Tracheobronchial brush samples and BAL samples were found to be optimal sampling methods for detection of *M. hyopneumoniae* using PCR analysis in necropsied pigs (83). In another study, PSB samples taken from the right middle lobe and apical segment of the right caudal lobe in pigs identified the presence of ventilator-acquired pneumonia as defined histologically in 69% of cases (versus 78% of cases with BAL), with a specificity of less than 50%, and an ability to identify the causative organism in less than 50% of cases (61).

Endobronchial biopsy. Endobronchial biopsy is the method used to sample the mucosal lining of the airway or endobronchial lesions. The main types of forceps available for this procedure are the toothed (alligator) forceps; the cupped-tip forceps, which has a cutting edge; and the needle forceps, which has a sharp prong positioned between the jaws to facilitate easier positioning of the forceps in the target area. Multiple biopsies should be taken because this improves the diagnostic yield. At least five endobronchial biopsies are recommended in cases of suspected malignant endobronchial tumors in humans (84). Biopsies can be taken from any location within the airways, and optimal sites suggested in humans (which are also suitable in pigs) are the right middle lobe carinae and the subsegmental carinae (4th generation bronchi) of the caudal lobes (85). Endobronchial biopsies performed in pigs have been shown to be of excellent quality for microscopic evaluation (24). The mean diameter of endobronchial biopsies taken with a 2.4-mm forceps from the trachea, main bronchi, and proximal cranial and caudal lobe bronchi were shown to be 2.4 ± 1.2 mm. This study also showed that cryoprobe biopsies produced larger samples than forceps biopsies with similar levels of bleeding (24).

Transbronchial biopsy. Transbronchial lung biopsy (TBLB) is the method used to obtain small samples of lung parenchymal tissue. This method of lung biopsy has been performed in pig models to assess the risk of performing TBLB on patients who are anticoagulated and to compare the efficiency of an electrocautery hot forceps with a traditional forceps (25, 86, 87). The methods used in pigs have been similar to those described in humans (25, 88). Although TBLB can be done without radiological guidance, fluoroscopic guidance is invaluable in obtaining higher-quality tissue samples by assisting the guidance of the forceps into the desired biopsy area (88). In addition, fluoroscopy is recommended to reduce the incidence of procedure-related pneumothorax (88). The incidence of TBLB-related complications, such as pneumothorax and bleeding, has

Table 2. Bronchoalveolar Cell Differential in Pigs and Humans

BALF Recovery	High-Health Pigs*	Low-Health Pigs†		Healthy Humans (Nonsmokers)
		Research Facility	Farm	
Total cell count (10^4/ml)	163 ± 73‡	171 ± 36	199 ± 59	12.9 ± 2.0
AM, %	85 ± 6	90 ± 3	88 ± 4	85.3 ± 1.6
PMN, %	7 ± 5	2 ± 1	4 ± 3	1.6 ± 0.7
Lymphocytes, %	8 ± 3	8 ± 2	8 ± 2	11.81 ± 1.1
Eosinophils, %	0	0	0	0.19 ± 0.06

Definition of abbreviations: AM, alveolar macrophage; BALF, bronchoalveolar lavage fluid; PMN, polymorphonuclear granulocyte.
*High-health pigs were weaned from a swine farm with minimal disease. All pigs were 10 wk old.
†Low-health pigs were weaned from a commercial farm with a 70% prevalence of macroscopic pneumonia lesions at slaughter. Low-health pigs were raised in a research facility or a farm. All pigs were 10 wk old.
‡Data are presented as mean ± SD and are taken from References 75 and 92.

also been shown to be higher in mechanically ventilated patients (89). Where pigs are mechanically ventilated during bronchoscopy, fluoroscopic guidance should be considered if performing TBLB, and previous studies support this use (25, 86, 87). Fluoroscopy can also be used to screen the pig for a pneumothorax after TBLB (87).

Complications of Bronchoscopy

Bronchoscopy is generally a safe procedure in humans and pigs. Data on the safety of this procedure primarily relate to humans but may be applicable to both species due to biological similarity. Complications reported include laryngeal, tracheal, and bronchial spasm; transient hypotension related to sedation; bleeding; cardiac arrhythmias; and pneumothorax. Major complications are very uncommon, with pneumothorax, hemorrhage, and respiratory failure occurring in less than 1% of procedures (90, 91). TBLB is the procedure with the highest rate of complications in humans (pneumothorax in 4% and hemorrhage in 2% of cases), with a higher reported frequency during mechanical ventilation (89, 90). However, studies where TBLB was performed on anticoagulated pigs produced no significant pulmonary hemorrhages and only one pneumothorax (86, 87). Transient fever is reported to occur in 10 to 30% of human patients after BAL (76). The risk of bleeding is slightly higher with bronchial brushing than with bronchial washing or BAL, especially where the mucosa is inflamed and more friable, and there is also a very small risk of pneumothorax when brushing peripheral lesions (81). Dislocation of the ETT causing hypoxia and necessitating reintubation and complications related to anesthesia causing death have also been reported in pigs (86). Careful preselection of animals, comprehensive anesthesia protocols, appropriate periprocedural animal monitoring, and good bronchoscopic technique are therefore essential to minimize potential complications during porcine bronchoscopy.

Conclusions

This article has discussed the anatomical, histological, and microbiological features of the porcine lung and describes how similarities between it and the human lung combine to make this a suitable large animal model for translational respiratory research. Bronchoscopic airway examination and bronchoscopic procedures can be efficiently and safely performed in pigs and should contribute significantly to future translational studies in this model. ■

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