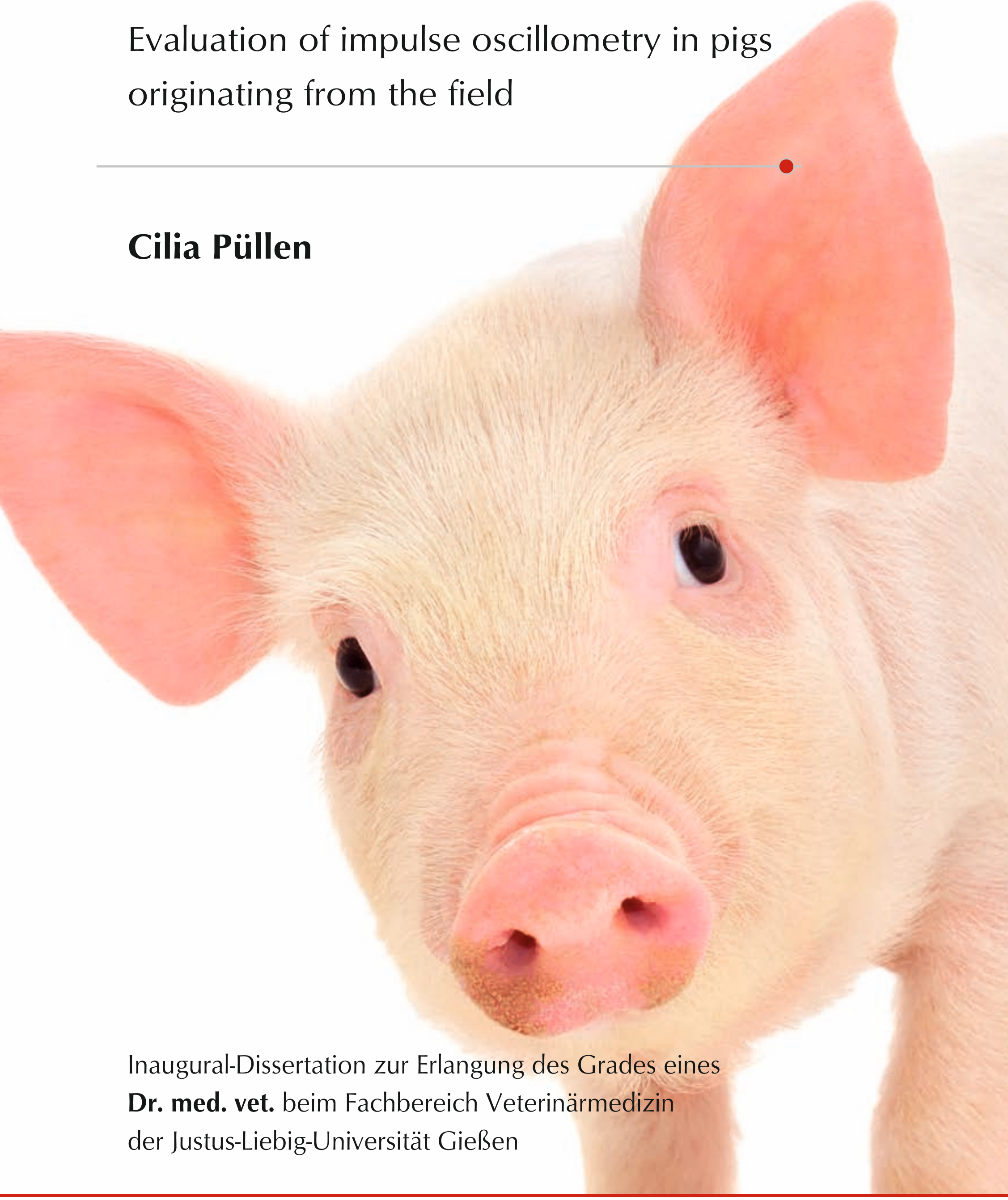
Evaluation of impulse oscillometry in pigs originating from the field

Cilia Püllen



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zur Erlangung des Grades eines
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Submitted from

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Mit Genehmigung des Fachbereichs Veterinärmedizin
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To my family

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Biomarkers for lung soundness and disease in swine – step 1: advanced clinical and pathological findings
ESPHM, Bruges, 25-27.04.2012

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Abbreviations

AF	breathing frequency
APP	*Actinobacillus pleuropneumoniae*
AX	reactance area
B.	*Bordetella*
BAL	bronchoalveolar lavage
BALF	bronchoalveolar lavage fluid
C	compliance; in FOT (e.g. IOS): capacitance (based on the analogy between pneumatic and electrotechnical parameters)
CO_2	carbon dioxide
CT	computed tomography
E	elastance
e.g.	exempli gratia
FOT	forced oscillation technique
f_{res}	resonant frequency
HE	hematoxylin and eosin
HPS	*Haemophilus parasuis*
HPV	hypoxic pulmonary vasoconstriction
Hz	hertz
i.e.	id est
IOS	impulse oscillometry system
kg	kilogram
kPa	kilopascal
L	inertance
L	liter
M.	*Mycoplasma*
MFO	monofrequent oscillometry
min	minute
ml	milliliter
O_2	oxygen
P.	*Pasteurella*
P	pressure
PCR	polymerase chain reaction
PCV2	porcine circovirus type 2
PMNs	polymorphnuclear neutrophils
PRDC	porcine respiratory disease complex

PRRSV	porcine reproductive and respiratory syndrome virus
R	resistance
RR	respiratory rate
RAO	recurrent airway obstruction
R_{dist}	resistance of distal airways
R_{prox}	resistance of proximal airways
R_{rs}	respiratory resistance
R5	resistance at 5 Hz
R10	resistance at 10 Hz
R15	resistance at 15 Hz
R20	resistance at 20 Hz
R5-R10	difference between resistance at 5 Hz and resistance at 10 Hz
R5-R15	difference between resistance at 5 Hz and resistance at 15 Hz
R5-R20	difference between resistance at 5 Hz and resistance at 20 Hz
s	second
S.	*Streptococcus*
SIV	swine influenza virus
spp.	species
V’	flow
V_{min}	minute ventilation
VT	tidal volume
X_{rs}	respiratory reactance
Z_{rs}	respiratory impedance
€	Euro

1 Introduction

Diseases of the respiratory tract are one of the most important health and economic problems in the modern swine industry (VanAlstine, 2012). Besides direct economic impacts, respiratory diseases lead to reduced animal welfare and issues with residues and antibiotic resistances. Therefore, they comprise a significant part of the principal problems in social and health policy.

Respiratory diseases in swine are often polyaetiologic, including infectious and non-infectious risk factors. Therefore, they are generally called the “porcine respiratory disease complex” (PRDC) (Honnold, 1997; Thacker and Thanawongnuwech, 2002).

Diagnosis ideally involves the inclusion of several diagnostic methods, i.e. clinical examination, dissection with gross pathology and histology as well as the detection of pathogens in the affected tissues (Nathues et al., 2012). In general, dissection combined with molecular and microbiological analyses are needed for a thorough diagnosis, as clinical signs are often non-specific. On the other hand, euthanizing pigs raises economic and welfare issues and places critical limitations on the statistical significance of the results.

Impulse oscillometry is a new and non-invasive method, yielding information on respiratory mechanics in the spontaneously breathing animal within a short time. Validated in different species, including calves and horses (Reinhold et al., 1996; Reinhold et al., 1998b; van Erck et al., 2004a), impulse oscillometry has also proved a promising tool for diagnosis and research in veterinary medicine.

To date, studies on impulse oscillometry in pigs are sparse. After its validation by Klein and Reinhold (2001) and Klein et al. (2003), the impulse oscillometry system (IOS) has been applied to analyse the effects of *Chlamydia spp.* and porcine reproductive and respiratory syndrome virus (PRRSV) on porcine pulmonary mechanics under experimental conditions (Reinhold et al., 2005b; Wagner et al., 2011).

The objective of the present study was to evaluate impulse oscillometry as an advanced clinical method in pigs originating from conventional pig farms where PRDC is predominant. The first manuscript evaluates the methodology and sensitivity of IOS, targeting the repeatability and variability of test results. The reactance area (AX) is also assessed as a new impulse oscillometric parameter. The second manuscript evaluates the contribution of clinical findings to the variability of impulse oscillometric traits.

2 Literature review

2.1 The Porcine Respiratory Disease Complex (PRDC)

In the past, the terms for respiratory diseases in pigs have undergone several changes due to the evolution of these diseases. This is comprehensively reviewed by Done and White (2003). Nowadays, diseases of the porcine respiratory tract are generally multifactorial, caused by interacting infectious and non-infectious risk factors. Due to their predominantly highly complex pathogenesis, respiratory diseases in pigs are often called the “porcine respiratory disease complex” (Done and White, 2003; Hansen et al., 2010).

2.1.1 Prevalence and relevance

PRDC is considered one of the major problems in the pig industry worldwide (VanAlstine, 2012). However, there are no precise data about the global prevalence of PRDC due to its varying manifestations and different housing conditions (Bochev, 2007). Cross-sectional studies conducted in slaughterhouses in several countries revealed average prevalence of pneumonia and pleurisy ranging from 23.85% to 72.4% and from 14.0% to 30.0%, depending on the lung scoring system applied (Enøe et al., 2002; Leneveu et al., 2005; Fraile et al., 2010; Meyns et al., 2011; Fablet et al., 2012). The prevalence of pleurisy has not changed over the last decade (Maes et al., 2001; Martínez et al., 2009; Meyns et al., 2011), despite improved management and preventive strategies. Pagot et al. (2007) reported a negative correlation between pneumonia and growth rates of fattening pigs.

Besides reduced performance, the impairment of pulmonary health leads to increased medication costs and elevated mortality rates (Maes et al., 1996). Major economic losses and animal welfare issues result. The economic impact of pulmonary lesions is significant, with a financial loss of up to € 3 per affected fattener (Aubry et al., 2010). Moreover, product quality decreases with the degree of lung lesions and the increased bacterial contamination of organs in affected pigs highlights the zoonotic relevance of the overall problem (Fehlhaber et al., 1992).

2.1.2 Risk factors

Several studies have previously been conducted to detect and characterise infectious and non-infectious risk factors involved in the development of PRDC.

Viral and bacterial pathogens associated with this disease commonly potentiate the clinical manifestation by coinfection and interaction (Thacker et al., 1999; Cho et al., 2006; Maes, 2010; Opriessnig et al., 2011). Sixty-three different combinations of pathogens have been reported so far (Hansen et al., 2010). Potential pathogens involved in PRDC include porcine circovirus type-2 (PCV2), porcine reproductive and respiratory syndrome virus (PRRSV), swine influenza virus type-A (SIV), *Mycoplasma hyopneumoniae* (*M. hyopneumoniae*), *Bordetella bronchiseptica* (*B. bronchiseptica*), *Pasteurella multocida* (*P. multocida*), *Haemophilus parasuis* (HPS*), Streptococcus suis* (*S. suis*) and *Actinobacillus pleuropneumoniae* (APP), among others (Choi et al., 2003; Kim et al., 2003; Palzer et al., 2008; Martínez et al., 2009; Hansen et al., 2010; Opriessnig et al., 2011; Fablet et al., 2012). A detailed literature survey of the individual pathogens is provided by Zimmermann et al. (2012). Many pathogens are ubiquitous, circulating in and between farms and even vary within production units (Dee, 1996).

The main source of infection is subclinical or mildly infected pigs (Bochev, 2007; Hennig-Pauka et al., 2007) which are purchased or mixed with pigs from other facilities, resulting in new pathogen combinations. Weaners and fattening pigs are most susceptible due to an insufficient immune defence system (Bochev, 2007).

Most bacterial lung infections occur when pulmonary defence mechanisms are compromised. Predisposing factors are viral or mycoplasmal infections as well as stress or poor climatic conditions (Du Manoir et al., 2002). In a previous study, minimal airborne contaminants on farms, i.e. dust, endotoxin or peptidoglycan, were sufficient to induce cellular changes in bronchoalveolar lavage fluid (BALF), which can serve as a precursor to viral and bacterial infections (Jolie et al., 1999a; Jolie et al., 1999b). Other non-infectious risk factors leading to an increased susceptibility to viral and bacterial infections are environmental, management and housing conditions, comprehensively illustrated in a review by Stärk (2000).

2.1.3 Clinical signs

Due to its polyaetiological character, clinical signs of PRDC are variable and can be characterised by sneezing, coughing, dyspnoea, nasal discharge, fever, anorexia, depression, accelerated respiratory rate and reduced performance such as poor growth rate (Bochev, 2007). Even though all pigs are susceptible, pigs around 16-20 weeks are most affected (Dee, 1996; Halbur, 1997; Done and White, 2003).

2.1.4 Diagnostic methods

Exact diagnosis of the causes of PRDC is often difficult since the aetiology is regularly multifactorial (Honnold, 1997). A combination of clinical signs of respiratory disease, dissection and the isolation of the involved pathogens in the affected tissues is recommended for an adequate diagnosis (Nathues et al., 2012).

According to Done and White (2003), clinical diagnosis is based on six major clinical signs: a) death after peracute or acute infection, b) pyrexia, c) sneezing after infection of the upper respiratory tract, d) coughing when the middle respiratory tract is affected, e) dyspnoea when small airways and alveoli are affected and f) production failure. However, clinical examination cannot be used on its own, as many respiratory infections do not exhibit distinct signs (Christensen and Mousing, 1992) and subclinical or persistent infected animals usually show no or only mild clinical signs (Pringle et al., 1988; Hennig-Pauka et al., 2007).

Gross pathology and histopathology as well as laboratory diagnostics provide the most complete information about the causative agents (Bochev, 2008; Szeredi et al., 2013).
Pig slaughter inspection is widely used for epidemiological studies and herd health inspection (Leneveu et al., 2005; Meyns et al., 2007; Pagot et al., 2007; Sibila et al., 2007; Martínez et al., 2009; Fraile et al., 2010; Fablet et al., 2012). Pathologic-histological examination can be a valuable tool to assess the relevance of the isolated pathogens (Hansen et al., 2010), which can be identified via polymerase chain reaction (PCR) or cell culture. However, relying solely on bacteriological examination is not recommended, since potential pathogenic bacteria have been detected in healthy pigs as well (Hensel et al., 1994; Hennig-Pauka et al., 2007).
Moreover, some bacteria require special media or grow very slowly, highlighting the advantages of PCR: it is quicker than cell culture, independent of antibiotic treatment (Maes et al., 1996) and can also detect viral genetic material (Woeste, 2007).
Serological testing can be applied for herd screening, epidemiological surveys or for controlling vaccination efficacy (Maes et al., 1996; Maes et al., 2000). It should not be applied as the sole diagnostic test in individual affected animals, since serum titres can derive from various effects, e.g. maternally derived antibodies, after recovery from disease or endemic latent disease (Hennig-Pauka et al., 2007).

Bronchoalveolar lavage (BAL) is routinely applied as a minimally invasive diagnostic method in affected pigs. It is a potential tool to detect early indices of pneumonia by assessing the percentage of polymorphonuclear neutrophils (PMNs) (Hensel et al., 1994) and to identify disease markers for bacterial infections in the recovered fluid (Hennig-Pauka et al., 2007). However, this technique only represents a restricted local area of the lung (Reinhold et al., 2005a) and the lavaged region does not correspond to the site of the lung lesion (Moorkamp et al., 2008).

2.2 Characterisation of lung function in swine

2.2.1 Anatomical and physiological characteristics of the porcine pulmonary system

Structural differences in the respiratory system across various species influence lung function and can lead to a different susceptibility to respiratory disorders.

Based on their subgross anatomy, there are three types of mammalian lungs (McLaughlin et al., 1961; Robinson, 1982). Along with sheep and cattle, the porcine lung belongs to category I. This categorisation is based on several anatomical and physiological features (Robinson, 1982):

- High degree of lung lobulation/segmentation
- Thick pleura
- Lack of collateral ventilation (channels of Lambert, pores of Kohn, channels of Martin)
- Strong *Tunica muscularis* in the vascular walls of small pulmonary arteries

The lobulation of bovine and porcine lungs is very well developed. Every secondary lobule is a macroscopically defined unit which is distinctly separated from adjacent segments by complete fascial sheaths, ventilated via a lobar bronchus and perfused by a functionally corresponding blood system (Reinhold, 2005). Due to this high degree of lung lobulation, the spread of infections and inflammations throughout the lung is limited (Caswell and Williams, 2008), creating a coexistence of affected and unaffected segments within the same lung lobe. Findings from lung biopsies or bronchoalveolar lavage are hence only representative for the sampled lung region, not the entire pulmonary system.

The high level of segmentation also leads to increased tissue resistance and decreased pulmonary elasticity (dynamic compliance) in pigs and cattle (Reinhold, 2005; Kirschvink and Reinhold, 2008). Consequently, breathing work is already elevated during basal respiration in both species in order to overcome this resistance (Reinhold, 2005).

Collateral ventilation is a compensatory mechanism during ventilation inhomogeneities, supplying the affected region with oxygen via accessory airways.
Cattle and pigs lack collateral ventilation which frequently leads to atelectasis during airway obstruction. The result is ventilatory asynchronisms with marked atelectasis in some parts and hyperinflated regions in other regions of the lung, leading to alveolar hypoventilation. These regional inhomogeneities result in considerably impaired gas exchange efficiency with hypoxemia, hypercapnia, an elevated alveolar-arterial oxygen difference and an increased right-to-left vascular shunt (Kirschvink and Reinhold, 2008).

In the small pulmonary arteries, the degree of hypoxic pulmonary vasoconstriction (HPV) is positively correlated with the thickness of the *Tunica muscularis*. HPV is a compensatory mechanism which ensures perfusion of better ventilated regions by diverting blood flow from poorly ventilated areas. The *Tunica muscularis* is very thick in cattle and swine, resulting in severe pulmonary hypertension during alveolar hypoxia and other vasoconstrictive stimuli (Reinhold, 2005).

2.2.2 Mechanics of breathing

The theoretical description in this chapter is mainly based on Reinhold (1997a) and Robinson (2007).

Pulmonary ventilation is composed of inspiration, which transports oxygen (O_2) to the alveoli, and expiration, which transports carbon dioxide (CO_2) from the alveoli into the environment. This mechanism is realised by the respiratory system, which consists of the lungs and the thorax. Both are mechanically connected to each other by pleural liquid. Therefore, the respiratory system behaves as a single unit, creating differences between the intrapleural and barometric pressure during the process of breathing. During inspiration, this pressure difference has to be established actively through muscular strength in order to overcome restrictive forces and frictional resistances of the pulmonary system. Expiration is a passive mechanism due to the lungs' elastic recoil.
The parameters of ventilation are breathing frequency (AF), tidal volume (VT) and minute ventilation (V_{min}). The physiological breathing frequency of weaned pigs averages 26±5

breaths per minute (Plonait, 2004). Tidal volume represents the volume which is inhaled and exhaled within one breath of air, which is about 10 ml/kg body weight in mammals and approximately 8-10 ml/kg body weight in pigs (Stahl, 1967).
Minute ventilation is the product of tidal volume and breathing frequency (V_{min}=VT x AF). It represents the total volume which is inhaled and exhaled in one minute. V_{min} is also the sum of dead space and alveolar ventilation. The anatomic dead space reaches from the nares to the bronchioles, which are the conducting airways. They do not take part in gas exchange. Alveolar ventilation is the physiological prerequisite for alveolar gas exchange.

Pulmonary ventilation is determined by mechanical properties of the respiratory system, i.e. resistance, compliance and inertance.
Resistance (R) describes flow resistances in the conducting airways. It results from the ratio between pressure and flow (kPa/(L/s)). Airflow resistance is determined by the calibre and length of the conducting airways. Airways with a small diameter exhibit a greater resistance than those with a large calibre. The diameter of individual airways decreases with increasing division of the conducting airways; however, the sum of individual cross-sections increases, which decelerates airflow and gradually reduces total airway resistance towards the lung periphery. Due to the Hagen-Poiseuille law, reduction in airway calibre will lead to a significant increase in resistance, e.g. during airway obstruction.

As a result of this branching pattern, about 80% of total pulmonary resistance is due to airways larger than 2 to 5 mm in diameter (tracheobronchial tree). Only 20% of pulmonary resistance is contributed by the bronchioles.

Compliance (C) is derived from the ratio of volume and pressure (L/kPa), reflecting thoracopulmonary elasticity. C is the reciprocal of elastance (E), which corresponds to the elastic resistance of the lung which has to be overcome during inspiration. Within the pulmonary tissue, lung elastic recoil is generated by collagen and elastic fibres and the surface tension forces of alveoli and terminal bronchioles. Compliance increases with progressive pulmonary elasticity.

Inertance (L) represents inertia of the moving air column in the airways and of the thoracopulmonary tissue. During basal breathing, L can be neglected, whereas it can crucially influence the dynamic compliance during fast and laboured breathing (Lekeux, 1988; Art et al., 1989).

2.2.3 Methods of evaluating lung function disorders in pigs

From a clinical perspective, the following examinations can be applied to detect possible indices for respiratory disorders (Jackson and Cockcroft, 2002): physical examination including general condition, posture and appetite, body temperature, heart rate, mucous membranes, ocular and nasal discharge, upper airway noises, sneezing, respiratory frequency, depth, character and rhythm, coughing, expiratory grunting, palpation (to identify thoracic pain), abnormal breathing sounds (auscultation) and percussion.
Further investigations include bronchoalveolar lavage, serological testing, nasal swabs and saliva to identify the aetiological agent, radiography, ultrasonography, lung biopsy, thoracocentesis, pulse oximetry and acid/base blood gas analysis.
The limiting factors of clinical examination are that only a symptomatic diagnosis can be made (Bryson, 1985) and the absence of subclinical pneumonia cannot be guaranteed (Pringle et al., 1988).

Special techniques are needed to detect lung function changes which are not clinically apparent and to quantify the degree of functional disorders. In veterinary medicine, the range of these techniques is limited, as only methods which do not require the patient's cooperation can be applied (Reinhold, 1997a).

Conventional techniques used to analyse respiratory mechanics in animals are based on a volume and pressure relationship or a pressure and flow relationship, including whole body barometric plethysmography, oesophageal balloon and interrupter technique (Reinhold 1997a). Their principles and applications in animals are explicitly described in a review by Reinhold (1997a). However these techniques require a high methodological effort, making them expensive and partially invasive.
The three measuring techniques mentioned above have already been applied in pigs (Intraraksa et al., 1984; McFawn et al., 1999; Halloy et al., 2004a; Halloy et al., 2004b; Halloy et al., 2004c; Halloy et al., 2005).

Alternative, non-invasive methods include the forced oscillation technique (FOT). The following descriptions of the measurement principle and the classification of the measuring systems are based on Reinhold (1997b).

The FOT is based on a test signal which is generated by an external generator and superimposed on the spontaneous breathing at the mouth of a human patient or the nose of an animal. The respiratory system reacts to the signal with a shift in pressure (P) and flow (V') during breathing. The differences between P and V' are registered by a computer and analysed and interpreted according to time and frequency. Since this technique is performed without forced respiratory manoeuvers or specific respiratory actions, the smooth muscle tone of airways is unlikely to be modified by FOT and can be analysed without the patient's cooperation (Navajas and Farré, 1999).

Depending on the physical characteristics of the signal, the FOT can be categorised into monofrequent oscillometry (MFO), polyfrequent oscillometry (which is further classified into the pseudo-random noise and random noise techniques) and multifrequent oscillometry.

2.2.4 The application of the impulse oscillometry system (IOS) in human and veterinary medicine

The current project employs the impulse oscillometry system, which is based on the multifrequent oscillometry. The theoretical background and interpretation of test results have been described comprehensively by Reinhold (1997b) and Smith et al. (2005).

IOS principle and interpretation of test results

In contrast to the classic measuring techniques mentioned in Chapter 2.2.3, the IOS test signal is generated via an external loudspeaker generator.

The advantages of an externally generated test signal include its precise definition and reproducibility, easier measurement of externally applied forces and the generation of frequencies which cannot be produced by respiratory muscles (Peslin, 1989).

The IOS generator superimposes small impulse shaped pressure signals on the spontaneous breathing of the examined animal. The signal has a frequency content from 0 to 100 Hz. Airflow and pressure signals are measured by a pneumotachograph and a pressure transducer, which are closely connected to the animal's face mask (Fig. 1). The pulmonary system reacts to the impulse pressure with a shift in P and V'. Changes in these variables are registered after each impulse and then analysed and interpreted based on the frequency of the applied signal. The resulting index is respiratory impedance ($\underline{Z}_{rs}$), which is calculated using a Fast Fourier

Transform. Among the assessment of mechanical parameters, the pneumotachograph can register pulmonary ventilation variables such as respiratory rate (RR) and tidal ventilation (VT).

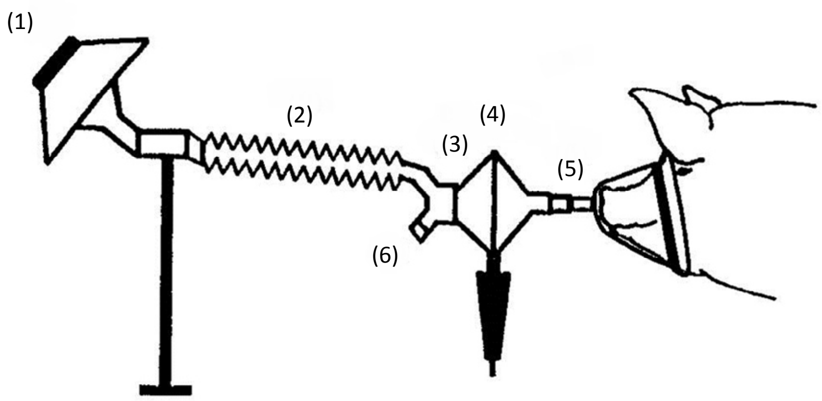

Fig. 1: Principal design of IOS measuring technique in swine (Klein and Reinhold, 2001)
(1) loudspeaker; (2) folded hose; (3) Y adapter; (4) pneumotachograph and pressure sensor; (5) connection to facemask; (6) opening with closing resistor

$\underline{Z}_{rs}$ is a complex parameter, consisting of 'real' (respiratory resistance (R_{rs})) and 'imaginary' components (respiratory reactance (X_{rs})).

R_{rs} reflects resistive properties of the conducting airways (R) and is primarily frequency independent under physiological conditions. X_{rs} can be separated into capacitance (C) and inertance (L). Capacitive resistances represent elastic properties of the thoracopulmonary system whereas inertance reflects the inertia of the air column in the conducting airways and the thoracopulmonary tissue (Fig. 2).

Based on the frequency of the applied test signal, R_{rs} and X_{rs} are graphically displayed after each IOS run (Fig. 3). Capacitive resistances are located in the negative area of X_{rs}. At low frequencies, they approach the y-axis. At high frequencies, they approach the x-axis. L is always positive and increases with increasing frequency. Due to this frequency dependence, inertive and capacitive resistances can be discriminated and therefore improve diagnostic significance. Both L and C determine the spectral course of X_{rs}, which generally starts negative and gradually becomes positive with increasing frequency.

When the curve of reactance crosses the abscissa, the capacitance and inertance have equal magnitude. This point is the resonant frequency (f_{res}).

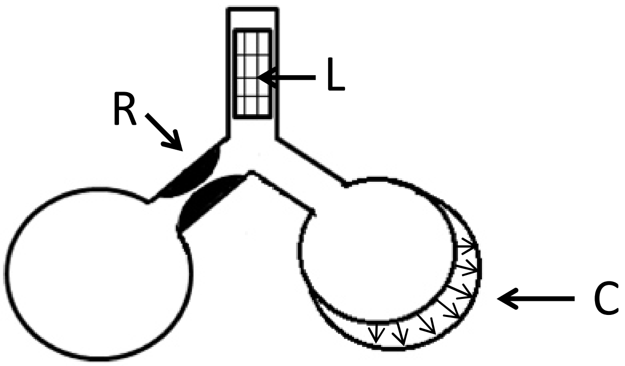

Fig. 2: Schematic presentation of the components of respiratory impedance ($\underline{Z}_{rs}$) (based on Wagner (2010))

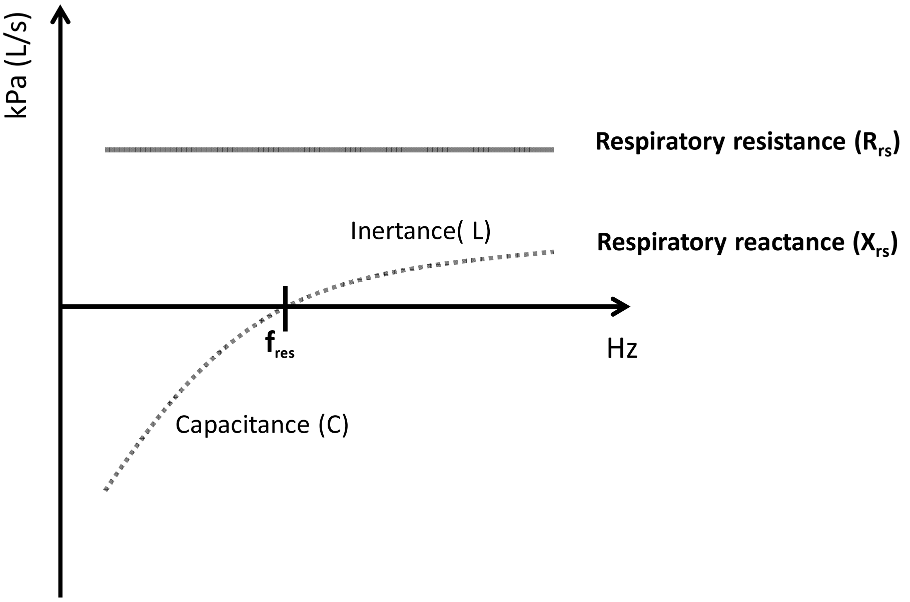

Fig. 3: Schematic presentation of the spectral course of R_{rs} and X_{rs} depending on the frequency of the test signal (based on Reinhold (1997b))

Compared to other methods, the advantage of analysing IOS results is that there are various ways to interpret impulse oscillometric parameters for diagnosis (absolute values, the spectral course of R_{rs} and X_{rs}, model parameters and the combination of parameters) (Bisgaard and Klug, 1995).

Alterations in respiratory function can be differentiated and allocated to proximal and peripheral airways depending on the frequency and the choice of resistive or reactive components. Low frequencies represent the entire respiratory tract and the functional properties of peripheral airways as they reach deep into the pulmonary system. In contrast, high frequencies remain in the upper conducting airways, reflecting extrathoracic regions of the respiratory system.

Due to this frequency dependence, $\underline{Z}_{rs}$ exhibits particular changes depending on the localisation of pulmonary functional disorders (Fig. 4):

During proximal obstructions, the spectral course of R_{rs} increases throughout the whole frequency range whereas reactance remains unchanged.

Obstructions in the peripheral airways are characterised by increased values of R_{rs} at low frequencies (3 and 5 Hz) which decrease with increasing frequency, displaying a negative frequency-dependence of respiratory resistance. Frequency-dependence increases with rising peripheral resistance. Values for respiratory reactance at low frequencies simultaneously decrease, resulting in a drop in the spectral course of X_{rs}.

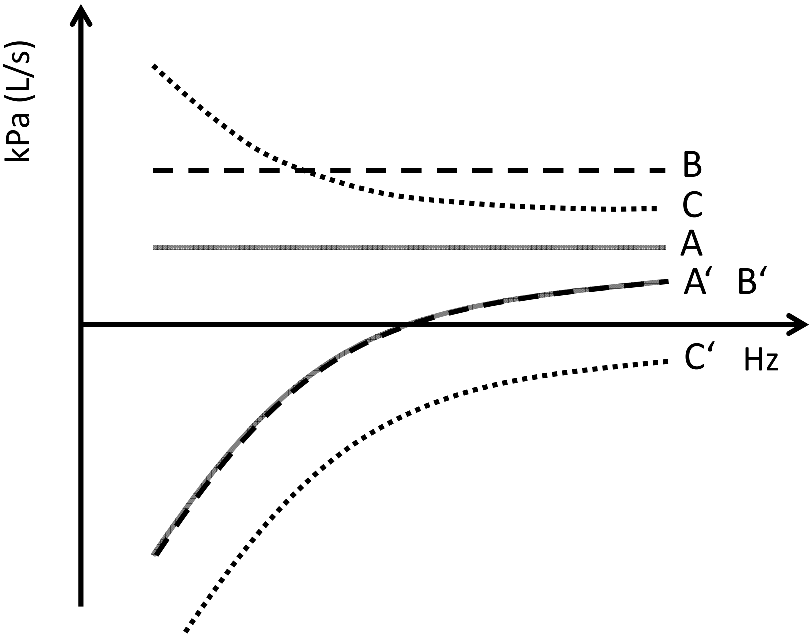

Fig. 4: Schematic presentation of the spectral course of R_{rs} and X_{rs} during phsyiological and pathophysiological conditions depending on the frequency
Spectral course of R_{rs} (A) and X_{rs} (A‘) during physiological conditions (solid lines)
Spectral course of R_{rs} (B) and X_{rs} (B‘) during proximal obstructions (dashed lines)
Spectral course of R_{rs} (C) and X_{rs} (C‘) during peripheral obstructions (dotted lines)

Frequency range for IOS analysis in pigs and model parameters

Depending on body size, the most useful frequency range for clinical evaluation differs among species (Smith et al., 2005). In pigs, it is recommended to restrict IOS parameter analysis to 3-15 Hz. Impulse oscillometric indices ≥ 15 Hz mainly represent extrathoracic airways in swine and calves. They can be distorted by the capacitive shunt effect caused by the face mask which connects the animal to the measuring system. Therefore, they are not suitable for analysing R_{rs} and X_{rs} (Reinhold et al., 1998b; Klein et al., 2003).

Besides the spectral parameters, model-derived parameters (resistance of proximal and distal airways (R_{prox}, R_{dist})) are provided by the IOS for further interpretation of findings and to simplify the complex oscillometric data. Based on a seven-element model of the lung by Mead (1961), R_{prox} (representing proximal airways, i.e. nasal cavities, larynx and pharynx) and R_{dist} (reflecting the lung periphery) have previously been described and validated in pigs and calves (Reinhold et al., 1998a; Reinhold et al., 1998b; Klein et al., 2003). Both parameters proved to be of diagnostic value in these species (Reinhold et al., 1998b; Klein and Reinhold, 2001; Reinhold et al., 2005b; Jaeger et al., 2007; Wagner et al., 2011)

IOS parameters applied in human medicine

Impulse oscillometry was first applied in human medicine by Müller and Vogel (1981). Ever since, this technique has been established in the research and diagnostics of human respiratory diseases (Oostveen et al., 2003; Smith et al., 2005).

The following parameters were reported to be sensitive for functional alterations in the lung periphery of humans: respiratory resistance (R_{rs}) at low frequencies (5 Hz); the difference between R5 (resistance at 5 Hz) and R15 (R5-R15) or the difference between R5 and R20 (R5-R20); respiratory reactance (X_{rs}) at low frequencies (5 Hz); the reactance area (AX) and the resonant frequency (f_{res}) (Goldman et al., 2002; Skloot et al., 2004; Larsen et al., 2009; Meraz et al., 2011; Shi et al., 2011; Frantz et al., 2012; Gonem et al., 2012; Qi et al., 2013; Schulz et al., 2013; Sugiyama et al., 2013).

F_{res} has been shown to be a sensitive parameter for the assessment of obstructive alterations in peripheral airways (Oostveen et al., 2003; Smith et al., 2005).

R5-R15 and R5-R20 indicate the frequency dependence of R_{rs}. They have been reported to be sensitive indices for increased peripheral airflow resistance (Brochard et al., 1987; Goldman et al., 2002).

The reactance area (AX) is a quantitative index which integrates all negative values of reactance between 5 Hz and the resonant frequency. This integration creates an area between the x-axis and X_{rs}. The AX index was established and validated by Goldman (2001) and Goldman et al. (2002). Correlating with R5-R15 and R5-R20, this parameter reflects changes in the degree of pulmonary restriction and/or peripheral airway obstruction (Smith et al., 2005; Meraz et al., 2011). In various studies, both indices proved to be sensitive parameters representing small airway function (Skloot et al., 2004; Goldman et al., 2005; Meraz et al., 2011; Shi et al., 2011).

IOS in veterinary medicine

In veterinary medicine, impulse oscillometry has been validated as a potential method for improving research and diagnostics in different species, e.g. calves and horses (Reinhold et al., 1996; Reinhold et al., 1998a; Reinhold et al., 1998b; van Erck et al., 2004a; van Erck et al., 2004b). This technique had a superior sensitivity to clinical examinations and established reference techniques in both veterinary and human medicine (Strie et al., 1997; Goldman, 2001; van Erck et al., 2004b; van Erck et al., 2006; Larsen et al., 2009; Richard et al., 2009; Frantz et al., 2012; Onmaz et al., 2013; Shi et al., 2013).

Within the relevant frequency range, most parameters of respiratory mechanics and ventilation known from human medicine can be applied to characterise and localise changes in the animal's pulmonary system (Reinhold, 1997b; Klein et al., 2003; van Erck et al., 2006). In addition to R_{rs}, X_{rs}, and f_{res}, the R5-R10 and R5-R15 differences have also been applied to animal studies, confirming their potential to reflect the intensity of peripheral bronchoconstriction (Young et al., 1997; Klein et al., 2003; van Erck et al., 2003).

The reactance area, however, is yet to be validated in animal studies.

Until now, studies on impulse oscillometry in pigs have been sparse. Initial experimental validations were performed by Klein and Reinhold (2001) and Klein et al. (2003). Further studies focused on experimentally induced changes in the respiratory system, i.e. the impact of *Chlamydia spp.* and porcine reproductive and respiratory syndrome virus infection (PRRSV) (Reinhold et al., 2005b; Wagner et al., 2011). The sensitivity of this method to alterations in the peripheral airways was confirmed via histopathological examinations (Wagner et al., 2011).

2.3 Objectives of the present study on current scientific knowledge

There is currently a paucity of information on respiratory function in affected pigs originating from the field. In the field, effects of mixed respiratory pathogens and suboptimal housing conditions strongly modify the high complexity of porcine respiratory diseases (Brockmeier et al., 2002; Bochev, 2007; Opriessnig et al., 2011).

The aim of the present study was to evaluate impulse oscillometry as a further clinical method in pigs coming from different conventional herds.

The study focused on methodological aspects and the sensitivity of impulse oscillometry to variations in lung function as well as the relationship between clinical findings and IOS data.

3 Manuscripts

3.1 Manuscript #1

Evaluation of impulse oscillometry in pigs of unknown disease status originating from the field

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Abstract

The aim of this study was to assess impulse oscillometry as a method to characterise lung function in 58 German hybrid pigs from 29 different herds of unknown respiratory status. The variability of repeated lung function measurements increased significantly after the sixth run and therefore the average of the first six runs was used for analysis. The presence of peripheral respiratory alterations in some pigs was indicated by the negative frequency dependence of the 95th percentile of respiratory resistance (R_{rs}), with highest values at 3 Hz and the sharp drop of respiratory reactance ($X_{rs)}$ across the whole frequency range (3-15 Hz). Respiratory resistance and reactance were negatively correlated. Reactance area was correlated with (1) R_{rs} at 3, 5 and 10 Hz; (2) X_{rs} at 3, 5, 10 and 15 Hz; (3) the frequency dependence of resistance compared between 3 and 5 Hz (R3-R5), 5 and 10 Hz (R5-R10), and 5 and 15 Hz (R5-R15); and (4) tidal volume. High repeatability and low intra-individual variability of impulse oscillometry indicate that this method is a promising tool for advanced characterisation of the pulmonary system of pigs and has potential for use for herd health monitoring.

Keywords: Impulse oscillometry; Lung function; Pigs; Respiratory mechanics; Reactance area

3.2 Manuscript #2

Relationship between clinical signs and results of impulse oscillometry in pigs originating from the field

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Abstract

The aim of the present study was to assess the contribution of clinical data to the variability of impulse oscillometric test results observed previously by Püllen et al. (2014). Fifty-eight German hybrid pigs from 29 different herds with unknown respiratory status were examined in the context of routine diagnostics as part of herd health service. Routine clinical examination was extended to a total set of 29 parameters, representing detailed clinical signs of the respiratory system, and to lung function testing applying the impulse oscillometry system (IOS). The resulting linear relationship between clinical data and variables of pulmonary mechanics had a mean r^2 of 0.52. Clinical parameters predominantly representing the lower respiratory tract closely correlated with established impulse oscillometric indices reflecting peripheral airways. Because of a restricted relationship between pulmonary functional disorders and clinical data, additional diagnostic methods are required to reveal the proportion of variance undefined by clinical examination.

Keywords: Impulse oscillometry; Clinical examination; Respiratory disease; Pigs; Reactance area

4 Discussion

In previous experimental projects, impulse oscillometry proved to be sensitive to alterations of respiratory mechanics in pigs (Klein et al., 2003; Reinhold et al., 2005b; Wagner et al., 2011). The present study evaluated the viability of impulse oscillometry in pigs originating from the field.

Pigs chosen for the examination

Two pigs were chosen from one pen per herd during routine diagnostics in the context of herd health service. One of the objectives of the present study was to evaluate the sensitivity of IOS in pigs with PRDC, which is predominant in conventional herds. We can assume that the aetiological factors (biotic and abiotic) and the respiratory status of the pigs were not homologous since they originated from different herds. Yet pigs with varying clinical signs and respiratory status were preferred to ensure the interindividual variations in respiratory function which were expected to be detected by IOS.

Results yielded a marked interindividual variability within the examined pigs, suggesting IOS is sensitive to variations in pulmonary mechanics. Clinical data served as control for this hypothesis by assessing the correlation and relationship between the results of clinical data and impulse oscillometry. Therefore, control animals, which are commonly used in experimental designs, were not needed as reference for IOS results and analysis in the present study.

Breathing frequency

Compared to the physiological respiratory rate (RR) of weaned pigs (26±5 breaths/min (Plonait, 2004), the mean breathing frequency of the examined pigs was elevated in all three IOS categories (Category 1: 36±10.28 breaths/min; Category 2: 34.2±8.46 breaths/min; Category 3: 46.3±17.11 breaths/min).

There are several possible explanations for the increased breathing frequency:
a) thermoregulation, b) excitement, c) pain and/or d) functional disorders of respiratory mechanics (Wagner, 2010).

Due to their lack of sweat glands, pigs regulate their thermal balance via evaporation of water in the respiratory tract (Ingram, 1974). The observed increase in breathing frequency could

therefore serve as a compensatory mechanism to cope with elevated body temperature, i.e. during fever, caused by infection or hyperthermia (Plonait, 2004).
Thermoregulation due to hyperthermia can be excluded as the pigs were housed and examined under standardised environmental conditions.
Because the pigs were chosen during routine diagnostics, only animals with clinical signs representing the actual herd problem were examined. Therefore, we can assume that, on average, the pigs were affected by respiratory tract infections which can lead to an elevated body temperature, among others. In the current study, ten out of 58 pigs exceeded the physiological body temperature of 39.3±0.3 °C (Plonait, 2004; Dewey and Straw, 2006).

Elevated breathing frequency could also have been the consequence of excitement during clinical examination as well as IOS measurement. Even though diazepam reduces breathing frequency (Klein and Reinhold, 2001), the pigs' excitement could have increased their respiratory rate during lung function testing. This hypothesis is congruent with Wagner (2010) who observed a habituation effect in pigs, expressed as a decrease in the mean RR with an increasing number of IOS measurements. Pain (e.g. from pleurisy (Frewein, 1999)) could also have influenced the respiratory rate in some of the pigs.

Overall, we can assume that the increased respiratory rate resulted from impaired respiratory mechanics. Since pigs were chosen during routine diagnostics, we can suppose that on average, their respiratory systems were affected.

In order to assess further pathophysiological causes which are directly located in the pulmonary system such as elevated oxygen demand, increased carbon dioxide release and impaired blood oxygen transport capacity, additional examination methods are needed, i.e. blood gas analysis and the rebreathing method (Wagner, 2010). However, these methods were not conducted during routine diagnostics in the current study.

Possible factors leading to a limited relationship between clinical and IOS data

Correlations between clinical data and IOS parameters were statistically significant, particularly for disturbances in the lower respiratory tract. Yet, only a restricted relationship between the clinical data and IOS variations could be displayed. The discrepancy between clinical and IOS results was also reported in calves originating from the field (Strie et al., 1997) as well as in calves with defined respiratory infections (Hildebrandt, 1999; van

Bömmel, 2000). One possible reason for this restricted relationship could be the superior sensitivity of IOS to subclinical or ongoing functional respiratory impairments, undetected by clinical examination. This hypothesis is consistent with Robinson et al. (2000), who analysed the relationship between clinical signs and lung function in horses with recurrent airway obstruction (RAO). In this study, alterations in pulmonary mechanics were substantial before the onset of clinical signs which could implicate an underdiagnosis of respiratory disease. The potential of IOS for early detection of subclinical respiratory diseases with absent clinical signs was confirmed by Richard et al. (2009).

The limited relationship between clinical and IOS findings in the present study could also have been influenced by the clinician who conducted the examination. Even though the examinations were all conducted by the same person to prevent interobserver variations, clinical findings are still subjective. Another observer could have made different clinical observations.

In order to reveal the proportion of variance undefined by clinical data in the present study, further routine diagnostic methods have to be applied, e.g. pathohistological examination. Pathological examination will also be a useful tool to differentiate the causes for the decrease in X_{rs} which does not discriminate between pulmonary obstructions and restrictions (Oostveen et al., 2003; Smith et al., 2005). Both pulmonary obstruction and restriction influence the compliance of the lung and lead to a change in X_{rs}. Furthermore, pathology will be valuable to validate the new parameter AX.

In a previous study in calves, correlations between X_{rs} and pathological findings were better than correlations with respiratory resistance. This observation was based on the closer relation of mechanical properties represented by X_r, to inflammatory and pneumonic lesions of the lung tissue which were more likely detectable by post-mortem examination (Reinhold et al., 2002). On the other hand, overall correlations between parameters of respiratory mechanics and pathological findings were poor. A possible reason for this lack of correlation might have been that functional impairments did not necessarily have to be detected by pathological examination.

According to Lekeux et al. (1993a), airway obstructions can be induced by a) intraluminal changes, i.e. excessive secretions; b) intramural causes, i.e. excessive contractions of bronchial smooth muscle, hypertrophy of mucous glands, inflammation or oedema of the walls or c) extraluminal alterations, i.e. destruction of lung parenchyma leading to loss of radial traction or compression of bronchi by enlarged lymph nodes or neoplasms.

Restrictions can be caused by a) alterations in lung parenchyma, i.e. interstitial pneumonia; b) intraparenchymal changes, i.e. intraalveolar accumulation of fluid or cells or c) extraparenchymal causes, i.e. thoracic tumours, accumulation of pleural fluid or skeletal deformities (Lekeux et al., 1993b).

Other than morphological correlations for bronchospasms, the pulmonary changes mentioned above are likely to be detected by pathology. Airway muscle contractions, however, are triggered by irritant receptors with vagal afferent nerves (Robinson, 2007).

In a previous study in pigs, changes in respiratory mechanics, especially restrictions, and histopathological findings corresponded with each other whereas bronchospasms could only be confirmed by the exclusion of further obstructive causes (Wagner et al., 2011).

Differential cytology of bronchoalveolar lavage fluid (BALF) might be another possible diagnostic method to detect factors leading to the described pronounced interindividual variations of IOS results. Significant correlations were observed between cytological findings and IOS measurements in a previous study in horses with inflammatory airway disease (Richard et al., 2009). However, to date there are no data on the relationship between cytological and IOS results in pigs.

Previous studies assessed the potential of applying differential cytology to BALF to discriminate between healthy animals and those with respiratory disorders. In general, total and differential cell count proved to be a valuable tool to assess inflammation in the respiratory tract. In healthy animals, macrophages are the predominant cell type (>90%) (Henderson and Muggenburg, 2001). During respiratory disease, total cell concentration increases while the ratio between macrophages and neutrophils changes in favour of polymorphonuclear neutrophils (PMNs) due to a massive influx of this cell type (Reinhold et al., 1992; Henderson and Muggenburg, 2001). A cut-off of 8% PMNs in BALF proved to be the most reliable parameter for indicating pneumonic pigs (Ganter and Hensel, 1997; Mombarg et al., 2002). The high number of neutrophils serves as an indicator for pulmonary inflammation (Allen et al., 1992; Henderson and Muggenburg, 2001) and reflects subclinical respiratory disorders (Pringle et al., 1988), whereas an increase in eosinophils or lymphocytes indicates an immune-based inflammatory response (Henderson and Muggenburg, 2001).

In contrast to clinical observation, BAL proved a sensitive tool for detecting subclinical pneumonia in pigs (Mombarg et al., 2002). Therefore, differential cytology of BALF could be a useful method for indicating commencing or subclinical respiratory tract infections as well as immunological reactions which lead to alterations in pulmonary mechanics in the absence of clinical signs or noticeable pathologic changes. However, a differentiated evaluation of lung function disorders, i.e. obstructions and/or restrictions, will probably not be possible with this method.

The insensitivity of clinical examination to subclinical respiratory diseases and the restricted diagnostic possibility of pathological and cytological examination to ventilatory disturbances suggest that the variability of IOS results may not be fully explained by these diagnostic methods. On the other hand, this limitation highlights the advantage of IOS, in that it generates additional information on disorders of the respiratory system.

Potential of IOS as a diagnostic method

Clinical signs of respiratory disease, dissection and the isolation of the involved pathogens are currently the gold standard for diagnosis of respiratory infections (Nathues et al., 2012). Although clinical and pathological findings may yield restricted information on pulmonary function disorders, impulse oscillometry will unlikely replace routine diagnostic methods.

It is possible to partly describe a relationship between isolated pathogens and impulse oscillometric results in calves with enzootic bronchopneumonia (Hildebrandt, 1999). This limited relationship could be due to the polyaetiological character of enzootic bronchopneumonia, which resembles PRDC in pigs. Experimental designs in gnotobiotic animals are hence favourable for analysing the impact of viral and bacterial infections on lung function (Hildebrandt, 1999). Under field conditions, however, the breed and immunity status of the animal as well as housing and climatic conditions crucially influence the infection and disease course. This can hardly be reproduced under standardised clinical conditions (Maes, 2010). Due to these multiple influences on the respiratory system in the field, it is questionable whether PRDC pathogens could be characterised by specific IOS traits.

Depending on the frequency and the choice of resistive or reactive components, IOS enables the allocation of qualitative and quantitative mechanical limitations to the upper and lower respiratory tract. However, the differentiation of restrictive and obstructive changes, of intra and extrapulmonary disorders and their aetiology is not possible with this technique (Oostveen et al., 2003). Therefore, pathological and clinical information is still required.

Alternatively, the rebreathing method or imaging techniques, i.e. computed tomography (CT), could be further non-invasive methods for differential diagnostics (Kneucker, 2008; Wagner et al., 2011; Brauer et al., 2011; Brauer, 2012; Brauer et al., 2012). Even though sonography and radiography can detect gross pathological lung changes in affected pigs (Klein, 1999; Höltig et al., 2008; Brauer et al., 2012), CT examination is superior to both techniques (Brauer, 2012; Brauer et al., 2012) and enables the diagnosis of pulmonary consolidations and alterations in pulmonary tissue (Brauer, 2012).

Since various primary and opportunistic pathogens are commonly involved in PRDC, pathology is further needed to differentiate the primary agents which ultimately lead to respiratory disorders.

In conjunction with routine diagnostic methods, however, IOS can be used as a valuable non-invasive technique, providing complementary information for the detection and quantification of functional respiratory impairments in an animal.

Applicability of IOS

The findings of the present study suggest impulse oscillometry is a valuable tool to complement conventional diagnosis of respiratory disorders in naturally infected pigs.
On the other hand, IOS will not be applicable as a diagnostic method in the field, since the prerequisites for good measurement practice cannot be ensured on most farms:

- Although the mobility of the system is high due to the use of a notebook, the methodological effort is still considerable since pigs have to be placed in a canvas sling which has to be mounted and transported (Fig. 3).
- At least two people are needed for lung function testing (one person to handle the system and another holding the head of the animal as well as the measuring head) (Fig. 3).
- Sedation of the pig proved necessary for its cooperation. Even though pigs can be trained to be examined without sedation, sedation significantly improved the quality of test results (Klein and Reinhold, 2001). Before and after the application of the sedative, pigs have to be kept and measured in a quiet environment to ensure a successful sedative effect and prevent paradoxical reactions (Löscher et al., 2006). Therefore, an appropriate room is needed which often cannot be provided on a conventional farm.

- The effort to prevent the system from spreading pathogens from one farm to another is considerable and expensive.

Fig. 5: IOS setup and pig during IOS measurement

In summary, preparations and lung function testing are time consuming and elaborate, and it is doubtful whether it will be accepted by farmers due to biosecurity issues.
Therefore, this technique should instead be used in a permanent location, e.g. universities, clinics etc., where pigs from the field can be examined in an appropriate environment.
IOS will unlikely be used in veterinary practice, due to the economic and biosecurity issues in conventional livestock farming.
IOS could be applied in valuable breeding animals as follow up or therapy control; however, for methodological reasons lung function measurement will be restricted to pigs weighing between 10-100 kg (Reinhold et al., 2005b).

Above all, impulse oscillometry can be employed to objectively characterise respiratory diseases in scientific research. Besides the analysis of lung function in experimentally or naturally infected animals, IOS could be used as a follow-up to therapy controls, to investigate the efficacy of pharmaceuticals or to verify the effect of different infection doses on the functional evolution of respiratory mechanics.

Recommendations for future studies

This was the first study to assess the methodological and diagnostic aspects of impulse oscillometry in pigs originating from the field. Additional lung function measurements should be conducted in further studies to generate more biological data in swine, to further validate the new parameter AX and to obtain more information on the impact of PRDC on respiratory mechanics.

In human medicine, reference values for IOS parameters for males, females and children are provided to interpret results of lung function measurements (Oostveen et al., 2003). In veterinary medicine, however, there are no comparable reference values which could be used to assess pulmonary alterations. However, it is questionable whether reference values can be used for lung function analysis. In a previous study in human medicine, predicted values for lung function testing derived from the standard deviation within populations were reported to be too wide to assess individual findings. Therefore, analysis of individual lung function results should primarily be based on values yielded by previous measurements of the patient (Baur et al., 1996). Hence, the recommendation is to compare the results of individual follow-up measurements or combine IOS results with findings from further diagnostic methods in order to assess results of lung function analysis for the individual animal.

As yet, there are no recommendations for the standardisation of lung function tests in animals like in human beings. In human medicine, recommendations on measurement technique, calibration and practical application of forced oscillation techniques have been published by the European Society Task Force (Oostveen et al., 2003). This would be necessary to ensure the comparability of IOS data on an international basis.

5 Summary

For the first time, the applicability of the impulse oscillometry system (IOS) was assessed for lung function analysis in pigs originating from the field. Fifty-eight German hybrid pigs from 29 different herds were chosen in pairs from one pen, each for routine diagnostics in the context of herd health service. The pigs were brought to the Clinic for Swine of the Justus-Liebig-University Giessen and examined according to standard clinical methods. Routine clinical examination was extended to a total set of 29 parameters, representing detailed clinical signs of the respiratory system. The overall examination was complemented by impulse oscillometry, bronchoalveolar lavage (BAL) and detailed pathology. Bronchoalveolar lavage fluid (BALF) and lung tissue samples were examined for relevant airway pathogens in swine using molecular and microbiological methods. In addition, BALF was examined by differential cytology.

The present study focused on the methodological aspects and sensitivity of IOS to variations in pulmonary mechanics as well as on the relationship between clinical findings and the results of lung function analysis.

The variability of repeated IOS runs increased significantly after the sixth run. Therefore, the average of the first six runs for each individual pig was used for further analysis. High repeatability and the pronounced interindividual variability of test results suggested impulse oscillometry as a potential method for improving clinical diagnostics in pigs with respect to respiratory diseases.

Eight clinical parameters exhibiting a marked relationship with the results of impulse oscillometry were extracted via multiple regression analysis. The resulting linear relationship between the clinical data and IOS parameters had a mean r^2 of 0.52. Correlations between clinical data and IOS parameters were statistically significant, particularly for disturbances in the lower respiratory tract. The correlations between clinical findings and the reactance area (AX) were similar to established IOS parameters, indicating the ability of AX to qualitatively and quantitatively assess functional disorders in the lung periphery.

Because of the limited relationship between pulmonary functional disorders and clinical data, the inclusion of the additional diagnostic methods, especially pathological and histological examination, is expected to detect further factors, leading to the high variability impulse oscillometric results.

Based on the present findings, IOS can be applied in conjunction with routine diagnostic methods as a valuable non-invasive technique, providing complementary information for the

detection and quantification of functional respiratory impairments in pigs originating from the field.

6 Zusammenfassung

In dieser Studie wurde die Anwendbarkeit der Impuls-Oszilloresistometrie erstmals als Methode zur Lungenfunktionsuntersuchung bei Schweinen aus dem Feld überprüft. 58 deutsche Hybridschweine aus 29 verschiedenen Herden wurden zu jeweils zwei Tieren aus einer Bucht für die Routinediagnostik im Rahmen des Gesundheitsmonitorings ausgewählt. Die Tiere wurden an die Klinik für Schweine der Justus-Liebig-Universität Giessen verbracht und dort mittels klinischer Standardmethoden untersucht. Die klinische Routineuntersuchung wurde um 29 Parameter erweitert, welche detaillierte respiratorische Krankheitsanzeichen repräsentierten. Die Gesamtuntersuchung wurde durch das Impuls-Oszilloresistometrie-System (IOS), der bronchoalveolären Lavage und einer ausführlichen pathologischen Untersuchung ergänzt. Bronchoalveoläre Lavageflüssigkeit (BALF) und Lungengewebe wurden mittels molekularen und mikrobiologischen Untersuchungsmethoden auf relevante Atemwegserreger des Schweins untersucht. Eine Differentialzytologie der BALF wurde ebenfalls durchgeführt.

Die Schwerpunkte der vorliegenden Untersuchung waren die Beurteilung der methodischen Aspekte und Sensitivität des IOS sowie die Bewertung des Bezugs zwischen klinischen Befunden und Ergebnissen der Lungenfunktionsanalyse.

Die Variabilität der wiederholten Lungenfunktionsmessungen stieg nach dem sechsten Messdurchgang signifikant an. Daher wurden die ersten sechs Messdurchgänge jedes Schweins für die weiteren Auswertungen gemittelt. Eine hohe Wiederholbarkeit sowie eine ausgeprägte interindividuelle Variabilität der Messergebnisse deuteten auf das Potenzial der Impuls-Oszilloresistometrie hin, klinische Diagnostikmethoden in Bezug auf porzine respiratorische Erkrankungen mit dieser neuen Methode optimieren zu können.

Mittels multipler Regressionsanalyse konnten acht klinische Parameter, die einen ausgeprägten Bezug zu IOS Daten aufweisen, extrahiert werden. Die lineare Beziehung zwischen klinischen Daten und Parametern der Atemmechanik wies ein mittleres Bestimmtheitsmaß (r^2) von 0,52 auf. Klinische Daten, die den unteren Atemtrakt reflektieren, korrelieren eng mit den bestehenden Parametern der Impuls-Oszilloresistometrie, welche die Lungenperipherie darstellen. Die Korrelationen zwischen klinischen Befunden und der Reactance-Fläche (AX) glichen denen gängiger IOS Parametern. Dies deutete auf das Potenzial von AX hin, Veränderungen in der Lungenperipherie qualitativ und quantitativ darstellen zu können.

Der Bezug zwischen pulmonalen Funktionsstörungen und klinischen Daten war begrenzt.

Durch Einbeziehung weiterer diagnostischer Methoden im Gesamtprojekt, vor allem der pathologischen Anatomie und Histologie ist zu erwarten, dass weitere Faktoren ermittelt werden können, die zu der hohen Variabilität der IOS Ergebnisse geführt haben.

Anhand der vorliegenden Ergebnisse bietet sich die Impuls-Oszilloresistometrie in Verbindung mit Methoden der Routinediagnostik als nützliches nicht-invasives Verfahren an, ergänzende Information zur Erkennung und Quantifizierung von funktionellen respiratorischen Störungen in Schweinen aus dem Feld zu liefern.

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