Adrenergic and cholinergic innervation of pulmonary tissue in the pig

Aleksandra Wojtarowicz, Piotr Podlasz, Krzysztof Czaja
Department of Animal Anatomy, University of Warmia and Mazury, Olsztyn, Poland
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Both the adrenergic and the cholinergic components of the autonomic nervous system (ANS) have been found to be an important source of nerve fibres supplying the lungs. On the other hand, data regarding the innervation of the pulmonary tissue in breeding animals are surprisingly scarce. Therefore, in the present study noradrenergic and acetylcholinesterase-positive (AChE-positive) innervation of the lungs of sexually immature pigs was studied using histochemical methods. Studies were performed on six juvenile female pigs (aged 9 weeks, body weight 15–20 kg). Samples of the tissue were collected from the caudal lobe of the right lung. 10µm cryostat sections were processed for the sucrose-potassium phosphate-glyoxylic acid technique to determine the occurrence and distribution of noradrenergic nerve fibres, while AChE-positive nerves were detected according to the acetylcholinesterase histochemistry. The present results revealed a dense network of noradrenergic nerve fibres localised mainly in the muscular membrane surrounding the epithelium of the bronchuli while AChE-positive nerve terminals supplied functional capillary vessels localised in the inter-alveolar septum and mucous membrane of the bronchi and bronchuli. The results of the present study confirm those of physiological experiments reporting the influence of the adrenergic and cholinergic components of the autonomic nervous system on the lung functions of pigs.

key words: lungs, adrenergic nerves, cholinergic nerves, pig

INTRODUCTION
The autonomic nerves that control the lungs generally show a pattern that consists of sensory afferent nerves originating in the epithelium of the airways, the lung parenchyma, and the muscle [5]. Efferent fibres may be myelinated or non-myelinated and end in the vagal nuclei, while afferent nerve terminals run to the muscle cells and glands and originate in ganglia [5, 9]. The latter are controlled preganglionically from the vagal nuclei and are part of the parasympathetic nervous system. The ganglia are situated close to the effector organ, lying external to the smooth muscle and the cartilage. The preganglionic fibres of the sympathetic nervous system leave the spinal cord and synapse with prevertebral ganglia, while postganglionic fibres emerge from the ganglia and are said to innervate the airway smooth muscle glands and pulmonary blood vessels [5, 7]. The above-mentioned data show that pulmonary tissue is well supplied by both adrenergic and cholinergic components of the ANS. However, data regarding the innervation of the pulmonary tissue in breeding animals are surprisingly scarce [2, 8, 10].

MATERIAL AND METHODS
Studies were performed on six juvenile female pigs (aged 9 weeks, body weight 15–20 kg) of the Large White Polish breed. The animals were injected...
with 10 mg/kg b.w. of proponylpromazine. After 30 mins the animals were deeply anaesthetised with 30 mg/kg of sodium pentobarbital (Vetbutal, Biovet, Poland; iv.). Samples of tissue were collected from the caudal lobe of the right lung. 10 µm cryostat sections were processed for the sucrose-potassium phosphate-glyoxylic acid technique [1] to determine the occurrence and distribution of noradrenergic nerve fibres. AChE-positive nerves were detected according to acetylcholinesterase histochemistry [3] and later stained with the Hematoxilin-Eozin (HE) method to visualise the background tissue. Sections were studied and photographed with a Zeiss-Axiophot microscope equipped with both the bright field for AChE-positive innervation and the epi-illumination fluorescence for detection of noradrenergic fibres. The principles of animal care (National Institutes of Health Publications No. 86–23, revised 1978) as well as the governmental regulations of Poland on the protection of animals were followed in the present study.

RESULTS AND DISCUSSION
The pulmonary blood vessels were richly innervated with noradrenergic nerves. Numerous nerve terminals located in the adventitia of the arteries formed a dense network (Fig. 1, 2). The present results revealed also noradrenergic nerve fibres localised mainly in the muscular membrane surrounding the epithelium of the bronchuli (Fig. 1, 3). Some nerve fibres were observed in the parenchyma of the lungs (Fig. 2–4). The muscular membrane surrounding the epithelium of the bronchi and bronchuli was innervated by an AChE-positive network of fibres (Fig. 5–8). Numerous AChE-positive nerve bundles and fibres were observed in the parenchyma of the lungs (Fig. 5, 6, 8). AChE-positive nerve terminals also supplied functional vessels localised in the inter-alveolar septum. The majority of the AChE-positive nerve fibres supplying the arteries were localised in their adventitia and ran circumferentially around these vessels (Fig. 7). The above-mentioned results confirm the results of physiological experiments reporting the influence of both the adrenergic and the cholinergic subdivision of ANS on the regulation of lung functions [4, 7, 9]. However, some controversy exists as to the control exerted by the adrenergic nervous system. Although the parasympathetic nerves are the major motor control system for airway smooth muscle, there is no certainty as to whether the sympathetics are responsible for relaxation. There is no doubt that β-adrenoceptors are present in the airways and that β-adrenoceptor agonists are powerful bronchodilators. The extent to which the receptor function is controlled by adrenergic nerves is not clear. It has been reported that there is no neural control and

Figure 1. Noradrenergic nerve fibres (arrowhead) localised in the muscular layer (ml) surrounding the epithelium of the bronchuli (b). Noradrenergic nerve terminals (arrow) located in the adventitia of artery (a). Magn. ¥ 400.

Figure 2. Noradrenergic nerve terminals (arrow) located in the adventitia of artery (a). Nerve fibres observed in the parenchyma (p) of the lungs (arrowhead). Magn. ¥ 400.

Figure 3. Noradrenergic nerve fibres (arrow) localised in the muscular layer (ml) surrounding the epithelium of the bronchi (b). Nerve fibres observed in the parenchyma (p) of the lungs (arrowhead). Magn. ¥ 400.
that the major relaxing system is that of the so-called non-adrenergic non-cholinergic inhibitory system [9]. Sympathetic nerves are said to be mainly destined for vascular smooth muscle when found traversing the airway. Less variation exists in cholinergic innervation to the airways than in the adrenergic. However, this had to be modified in view of the recognition of a non-adrenergic non-cholinergic excitatory system in some species [6]. The putative transmitter is substance P and the system is capsaicin-sensitive. No evidence for the existence of adrenergic inhibitory nerve fibres was reported in the airways collected post mortem, but the presence of non-adrenergic inhibitory nerves in the smooth muscle of the human trachea and bronchi has been demonstrated pharmacologically [9]. The preliminary results presented in the paper confirm the importance of further studies on the innervation of the pulmonary tissue in breeding animals. Further studies may help in the elucidation of the mechanisms of lung physiology and disorders and may establish a reference useful in functional studies of the lungs.
REFERENCES


