OESOPHAGEAL MANOMETRY IN EXPERIMENTAL PIGS: METHODS AND INITIAL EXPERIENCE

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Summary: The aim of this study project was to prepare our own method of porcine oesophageal manometry. Ten mature experimental pigs entered the study. Conventional water-perfused system was decided for manometry. Porcine resting and relaxed pressures of the lower oesophageal sphincter are fully comparable with healthy human subjects. Evocable swallowing is doable and oesophageal peristalsis is quantifiable. Basic manometric parameters were different in male and female animals. Oesophageal manometry in experimental pigs is feasible. Porcine oesophageal manometry will be usable for preclinical studies in future.

Keywords: *Experimental pigs; General Anaesthesia; Oesophageal Manometry*

Introduction

Pigs, as an omnivorous representative, can be used in various preclinical experiments due to their relatively very similar gastrointestinal functions compared to humans (1, 2). Our group demonstrated that gastric myoelectrical studies (using electrogastrography) are reliable and feasible in experimental pigs (3–7). The aim of this current project was to work out our own method of oesophageal manometry in experimental pigs.

Material and methods

Animals

Five mature male and five mature female experimental pigs entered the study (*Sus scrofa f. domestica*, hybrids of Czech White and Landrace breeds; 3–4 month old; weighing 27.5–41.5 kg, mean 32.0 ± 4.6, median 30.5 kg). Animals were fed twice a day (standard assorted food A1) and were allowed free access to water. All manometry investigations were performed under general anaesthesia in the morning after 24 hours of fasting. Intramuscular injections of ketamine (20 mg per kg; Narkamon, Spofa, Praha, Czech Republic) and azaperone (2.2 mg per kg; Stresnil, Janssen Animal Health, Saunderton, UK) were used as an introduction. General anaesthesia was carried out by propofol (2.2 mg/kg; Fresenius Kabi Deutschland GmbH, Bad Homburg, Germany).

Oesophageal manometry

Water-perfused disposable catheters were used (MMS G-88402, conventional 12 French, 8 channels with central lumen; MMS – Medical Measurement Systems B.V., Enschede, the Netherlands). Catheters were introduced into the oesophagus through mouth (using a dedicated mouthpiece). Their correct position was verified endoscopically. We used video-gastroscope Olympus GIF160 dedicated for animal use only (Olympus Optical Co, Tokyo, Japan). All animals were lying in supine position. Oesophageal manometry was performed for 10 minutes by means of the Polygraf UPS 2020 (UPS-2020 manometry system from MMS – Medical Measurement Systems B.V., Enschede, the Netherlands). Dry swallowing was induced by massage of lower part of the neck. All evaluated parameters were assessed as an average measure of four consecutive values.

Statistics

The data were analysed using SigmaStat software (Version 3.1, Jandel Corp., Erkrath, Germany). Descriptive statistics, Fisher's exact test and un-paired t-test were used.

Ethics

The Project was approved by the Institutional Review Board of the Animal Care Committee of the University of Defence, Faculty of Military Health Services, Hradec Králové, Czech Republic (Protocol Number 14/2012). Ani-

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mals were held and treated in accordance with the European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes (8).

Results

Oesophageal manometry was successfully accomplished in all animals. Lower oesophageal sphincter was easily identified in all animals (Fig. 1). Its middle part is 50 to 55 cm far from incisors. It was possible to evaluate all recordings in all 10 animals. In the absence of oesophageal contractions, artefacts can be readily identified, caused by respiration (18 cycles per minute in average) (Fig. 2). Oesophageal peristalsis during dry swallowing was evocable in all animals, with substantial relaxation of the lower oesophageal sphincter (Fig. 3). Values of basic parameters are given in Table 1. Male and female pigs were comparable in age and weight. Baseline pressure of the lower oesophageal sphincter and peristaltic wave pressure were different in male and female experimental pigs (see Figs. 4 and 5).

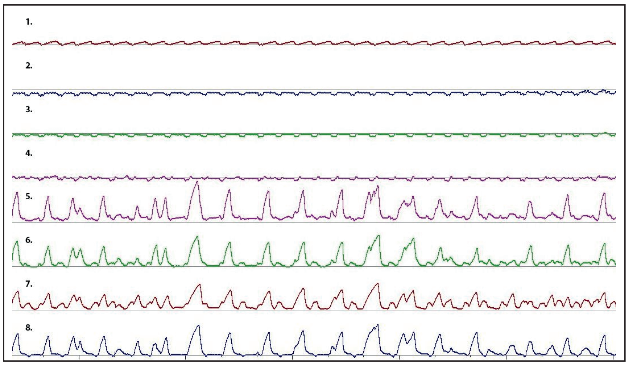

Fig. 1: Porcine oesophageal manometry using a water-perfused system.
Four proximal pressure sensors are localised in the oesophageal body (1.–4.) and four distal sensors are placed in the area of the lower oesophageal sphincter (LOS; 5.–8.). Manometry without swallowing shows a high-pressure LOS zone in distal four sensors (5.–8.). Pressure values displayed on the Y-axis; time course on the X-axis.

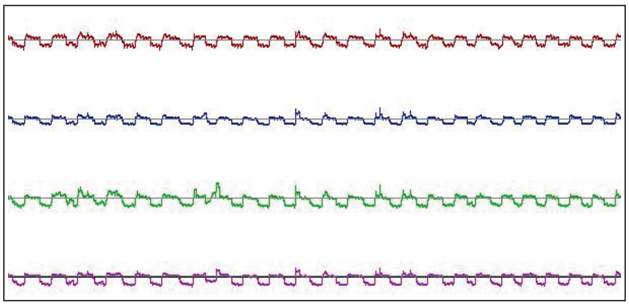

Fig. 2: Respiratory artefacts.
Four pressure sensors localized in the oesophagus show respiratory artefacts in the absence of oesophageal contractions.

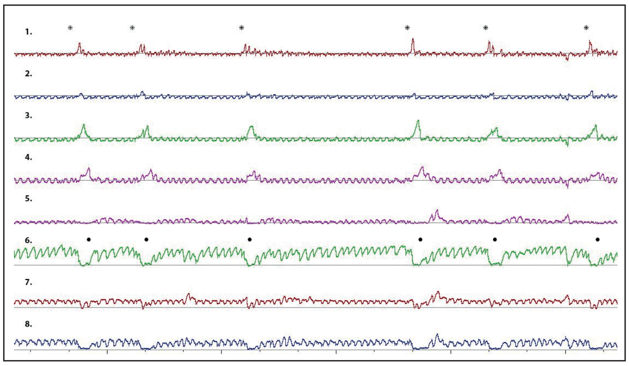

Fig. 3: Propulsive peristaltic contraction of the porcine oesophagus. Zone of high pressure is produced by the lower oesophageal sphincter and diaphragm (5.–8.). Several peristaltic sequences are recorded, with a propulsive increase of oesophageal pressure in the oesophageal body (sensors 1.–4.). Asterisks indicate dry swallows. Relaxation of the lower oesophageal sphincter is marked with black closed circles.

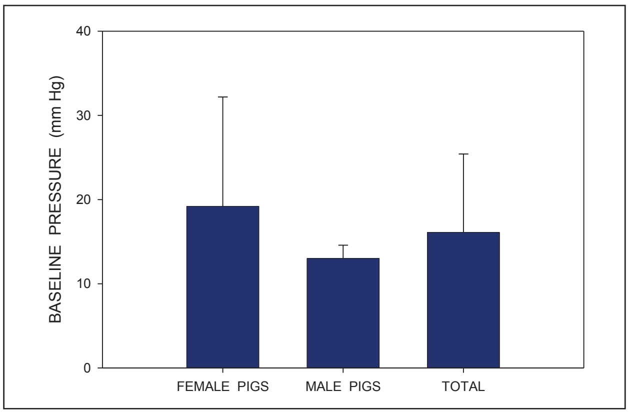

Fig. 4: Baseline pressure of the lower oesophageal sphincter in total and separately in female and male experimental pigs. The difference is not statistically significant (with power below desired value of 0.8).

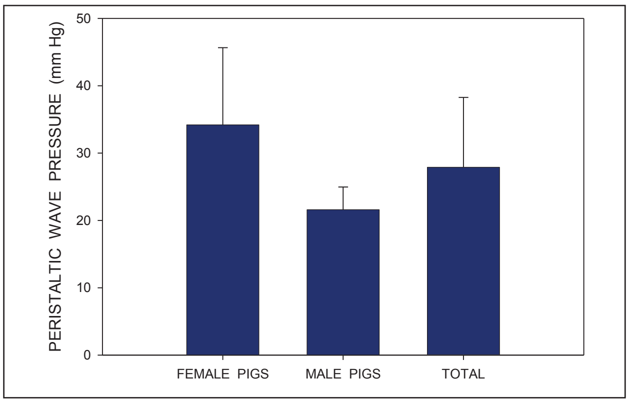

Fig. 5: Peristaltic wave pressure in total and separately in female and male experimental pigs. The difference between female and male pigs is statistically significant (p = 0.046).

Tab. 1: Oesophageal manometry in experimental pigs.

Animal No. / sex	**Baseline pressure of LES** (mm Hg)	**Relaxation of LES** (%)	**Duration of relaxation** (s)	**Peristaltic wave pressure** (mm Hg)	**Duration of peristaltic wave** (s)
1 / F	12	85	5.7	46	3.3
2 / F	23	100	23.0	20	4.5
3 / F	40	89	8.4	46	2.8
4 / F	7	100	2.4	29	2.3
5 / F	14	100	3.5	30	1.5
6 / M	15	95	5.5	24	1.7
7 / M	13	98	7.5	19	1.4
8 / M	11	100	1.5	26	1.5
9 / M	14	100	5.4	21	2.0
10 / M	12	100	4.4	18	2.0

F – female; M – male
LES – lower oesophageal sphincter
s – seconds

Mean relaxation of the lower oesophageal sphincter was 96.7 ± 5.4%, duration of relaxation was 6.7 ± 6.1 s. Mean duration of peristaltic wave was 2.3 ± 1.0 s. These two time intervals were longer in female pigs (Table 1), however, the difference did not reach a statistical significance. Propulsive peristalsis was found in 86%, there were simultaneous (3%), interrupted (4%) or non-transmitted ones (7%) in remaining cases.

Discussion

Our group elaborated our own method of oesophageal manometry in experimental pigs. It was a feasibility study in fact. Porcine hiatal and gastro-oesophageal anatomy and physiology are similar to human ones. The muscle is thicker at the point where the clasp (on the right side) and sling fibres (on the left) concentrate. The pressure profiles are axially and radially asymmetric in coincidence with the thickness variations of the corresponding muscle layers. Sphincteric pressure is recorded as a plateau, whereas diaphragmatic crural pressure appears as phasic oscillations in synchrony with respiration. The sphincter relaxed upon deglutition (9).

We were able to find out only two papers published so far that can be partly compared with our study. Ciotola et al. (10) performed peroral endoscopic myotomy (POEM) in five pigs. Mean pre-myotomy pressure of the lower oesophageal sphincter was 36 ± 8 mm Hg. After myotomy, the pressure significantly dropped to 10.6 ± 3.2 mm Hg (10). Perretta et al. (11) found mean preoperative pressure 22.2 ± 3.3 mm Hg and mean pressure 11.3 ± 2.7 mm Hg after POEM in four experimental pigs.

In humans, various authors reported different normal range of basic manometric parameters in healthy subjects, beside other things influenced by ethnicity and/or age (12–18). There are also significant pressure differences between solid-state and water-perfused systems in lower oesophageal sphincter measurement (16). Common values of baseline pressure of the lower oesophageal sphincter are about 5–25 mm Hg and relaxation 90–100% (with duration 5–10 s). Common peristaltic wave pressure is about 30–160 mm Hg in humans (with duration of contraction 2–6 s) (18). Based on our study, the crucial parameters of porcine oesophageal manometry are comparable with those in healthy humans.

Surprisingly, basic manometric parameters were different in male and female experimental pigs in our current study. Peristaltic wave pressure was significantly higher in female pigs. There was also a clearly distinct trend in other parameters in favour of female gender (higher baseline pressure of the lower oesophageal sphincter, longer duration of relaxation and longer duration of peristaltic wave), although they did not reach a statistically significant difference, mostly because of a small number of subjects. Gender-related difference of the oesophageal motility has not been reported in porcine manometry yet, but it was already described in healthy humans. Differences have been observed in water ingestion, oropharyngeal transit, duration of opening of the upper oesophageal sphincter, and pressure duration in the oropharynx with swallows (for review see ref. 19). Women also had longer duration of oesophageal contraction in the distal oesophageal body (19). The explanation for the results observed may be anatomic and/or hormonal differences between genders (20).

We are fully aware of possible limits of our current study. Primarily, this is our very initial experience with porcine oesophageal manometry. Number of subjects was sufficient for a usual animal setting but not for detailed statistics, especially correlation analysis. We decided a conventional water-perfused system, not a high-resolution manometry. All measurements were accomplished under general anaesthesia that could also influence the acquired results. Nevertheless all the obtained data are consistent.

Conclusions

Oesophageal manometry in experimental pigs is feasible. Porcine resting and relaxed pressures of the lower oesophageal sphincter are fully comparable with healthy human subjects. Evocable swallowing is doable and oesophageal peristalsis is quantifiable. Porcine oesophageal manometry will be usable for preclinical studies in future.

Conflicts of interest

The authors declare no conflict of interest.

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