Preventive Veterinary Medicine 147 (2017) 58–65

Contents lists available at ScienceDirect

Preventive Veterinary Medicine

journal homepage: www.elsevier.com/locate/prevetmed

Individual and pen-based oral fluid sampling: A welfare-friendly sampling method for group-housed gestating sows

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ARTICLE INFO

Keywords:
Sow
Behaviour
Group housing
Welfare
Oral fluid sampling

ABSTRACT

The aims of this study were to assess the feasibility of individual and pen-based oral fluid sampling (OFS) in 35 pig herds with group-housed sows, compare these methods to blood sampling, and assess the factors influencing the success of sampling. Individual samples were collected from at least 30 sows per herd. Pen-based OFS was performed using devices placed in at least three pens for 45 min. Information related to the farm, the sows, and their living conditions were collected. Factors significantly associated with the duration of sampling and the chewing behaviour of sows were identified by logistic regression. Individual OFS took 2 min 42 s on average; the type of floor, swab size, and operator were associated with a sampling time > 2 min. Pen-based OFS was obtained from 112 devices (62.2%). The type of floor, parity, pen-level activity, and type of feeding were associated with chewing behaviour. Pen activity was associated with the latency to interact with the device. The type of floor, gestation stage, parity, group size, and latency to interact with the device were associated with a chewing time > 10 min. After 15, 30 and 45 min of pen-based OFS, 48%, 60% and 65% of the sows were lying down, respectively. The time spent after the beginning of sampling, genetic type, and time elapsed since the last meal were associated with 50% of the sows lying down at one time point. The mean time to blood sample the sows was 1 min 16 s and 2 min 52 s if the number of operators required was considered in the sampling time estimation. The genetic type, parity, and type of floor were significantly associated with a sampling time higher than 1 min 30 s. This study shows that individual OFS is easy to perform in group-housed sows by a single operator, even though straw-bedded animals take longer to sample than animals housed on slatted floors, and suggests some guidelines to optimise pen-based OFS success.

1. Introduction

Oral fluid (OF) has gained interest in recent years in veterinary medicine for diagnostic purposes (Prickett and Zimmerman, 2010). In pigs, it is used as a matrix for the detection of hormones such as cortisol (Colson et al., 2012), antibodies such as those against the porcine reproductive and respiratory syndrome virus (PRRSV) (Prickett et al., 2008a; Olsen et al., 2013; Kittawornrat et al., 2014), and infectious pathogens which are excreted via saliva such as foot-and-mouth disease virus (Mouchantat et al., 2014b; Vosloo et al., 2015), PRRSV (Prickett et al., 2008b; Olsen et al., 2013), porcine circovirus type 2 virus (Prickett et al., 2011), influenza A virus (Romagosa et al., 2012; Panyasing et al., 2016) or classical swine fever virus (Dietze et al., 2017). Several studies have indicated that OF sampling may be a promising method to detect and monitor pig health under experimental conditions (Prickett et al., 2008b; Vosloo et al., 2015). Pen-based OF samples can also be used for virus detection in field conditions or in wild boar which need to be sedated to be individually sampled (Olsen et al., 2013; Mouchantat et al., 2014a, 2014b). OF sampling is acknowledged as a more welfare-friendly and alternative method to blood sampling, which is widely used for monitoring and control purposes.

In growing pigs, natural chewing behaviour, reflecting a feeding and exploratory motivation (Day et al., 1996), facilitate the collection of OF. In many studies, OF was successfully sampled from group-housed growing pigs, through the use of a cotton rope. Prickett et al. (2008b) and White et al. (2014) described the best age and the best duration for this collection. Seddon et al. (2012) studied the effects of housing conditions on chewing behaviours, showing that increasing the number of ropes increased the mean total chewing time per pig only in pigs reared on fully slatted floors.

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http://dx.doi.org/10.1016/j.prevetmed.2017.08.011
Received 23 November 2016; Received in revised form 7 August 2017; Accepted 16 August 2017

In older pigs, such as gestating sows or boars, assays have mainly been conducted on individually housed animals (Kittawornrat et al., 2010; Pepin et al., 2015), but not on group-housed animals. Because older animals are less curious and less motivated to explore manipulable materials or objects (Elmore et al., 2012), they may not be interested in chewing the material providing OF. Other factors, like the presence of a litter, may also influence the behaviour of animals faced with a material newly introduced into the pen (Scott et al., 2006). This may therefore interfere with the interest in the new material for OF collection and the success of sampling.

In the EU, where group housing is mandatory for gestating sows (Council, 2008), monitoring the health status of herds using these methods might be faster and easier to implement than blood sampling in group-housed sows. Indeed, blood sampling, which involves restraining the animal with a rope around the nose, can be unsafe for the operators and is painful for the animal.

The aims of the current study were therefore to assess the feasibility of individual and pen-based OF sampling in group-housed sows (time to sample and easiness to perform) and to compare these methods with blood sampling, which can be considered as the gold standard in the field. The factors influencing the success of sampling were also assessed.

2. Materials and methods

2.1. Study population

The study was carried out in 35 pig herds located in Brittany, western France, from February 2014 to June 2015. It was part of a largest study on the validation of the detection of PRRSV antibodies by ELISAs on OF or blood matrix (Fablet et al., 2017). The following criteria were required to select and stratify the herds: 1) the type of floor: concrete slatted vs. straw bedded, 2) the group size: small group (fewer than 15 sows per pen) vs. large group (more than 15 sows per pen), 3) the presence of at least one hundred breeding sows in the herd, and 4) the willingness of the farmer to participate. When technical results were available, they were recorded. In each herd, at least 30 gestating sows and three pens were selected at random. To avoid reproduction failure due to blood sampling, the selected sows were between 4 and 14 weeks of gestation.

2.2. Biological sampling under field conditions

All observations and samplings were done in the morning in every herds.

2.2.1. Individual OF sampling

Samples were collected from the buccal cavity of each sow using a folded sterile synthetic dry swab of 270 cm^2 (large swab) or 160 cm^2 (small swab) (Sodibox, Nevez, France). The swab was gently presented to the sow with a clamp (25 cm or 41 cm) by an operator to make the animal chew. The time spent to sample the sow, i. e. from the sow selection by the operator to the removing of the swab from the mouth of the sow, the size of the swab, the size of the clamp, and the name of the operator were recorded for each sample.

After collection, the chewed swabs were stored in a sterile tube and placed in a cool box. At the laboratory, OF was transferred to a sterile tube by wringing the swab with a sterile syringe. The volume collected was measured.

2.2.2. Pen-based OF sampling

Pen-based OF was collected after individual OF collection according to a method derived from Prickett et al. (2008a). The chewing devices were either a cotton rope with two strands (Idexx, Cergy-Pontoise, France, Fig. 1a) or with multiple strands, like a mop (Calipro, Lamballe, France, Fig. 1b), or a ten-strand synthetic swab (Sodibox, Nevez, France, Fig. 1c). They were placed in each pen for 45 min at approximately 70 cm above the floor, to hang at sow shoulder height. They were suspended over pens, 1 m from the wall via a metallic frame or hung directly from the pen fence. The number of chewing devices per pen depended on the group size: one device was given per 15 animals. Therefore, in large groups of more than 15 animals, several devices were suspended in the same way, and spread through the various areas of the pen. For each sample, the type of device and its position in the pen were recorded. All sows were standing up at the beginning of sampling.

OF was extracted from each device immediately after collection. It was placed in a sterile plastic bag and wrung-out by hand. The fluid was then stored in a sterile tube placed in a cool box. The volume collected was measured.

During the 45 min of sampling, the time spent by each sow to chew each device was recorded. To assess pen activity, the number of lying down or sitting sows was recorded at 15, 30 and 45 min after the beginning of the sampling procedure.

2.2.3. Individual blood sampling

Blood was sampled last. It was collected by venipuncture of the jugular vein of each OF sampled sow. The animals were restrained by a rope around the snout. The time spent to sample the sow, i. e. from the sow selection by the operator to the end of the venipuncture, was recorded. If the initial catch of the sow was not successful, the operator did it again until the sow was caught.

2.3. Information collected on the farm related to the sows and their living conditions

Group size, type of floor, type of feeding, feeding schedule, and presence of manipulable material other than straw bedding were recorded for each pen. Parity, gestation stage, number of days after introduction into the group, and genetic type were recorded for each sow.

2.4. Statistical analysis

2.4.1. Descriptive analysis

The technical results of the sampled herds were compared with the technical results of a reference group of Brittany farms by a Student *t*-test ($P < 0.05$)

Correlation between individual chewing time and sampled volume was tested by a Spearman correlation test ($P < 0.05$).

2.4.2. Factors influencing OF and blood sampling

2.4.2.1. Definition of the outcomes. For individual OF sampling, the outcome was the time spent to sample the sow, which was categorised as more than 2 min or less than or equal to 2 min. For blood sampling, the outcome was the time spent to sample the sow, classified in two categories: more than 1 min 30 s or less than or equal to 1 min 30 s. Since blood sampling required at least two operators and sometimes more (e.g. to protect the operators when other sows of the group were aggressive), a second synthetic variable multiplying the time spent to blood sample and the number of operators required for the sampling was defined and named “summarised operator time”. This outcome was categorised in two classes: more than 2 min 30 s-operator or less than or equal to 2 min 30 s-operator.

For pen-based OF sampling, three outcomes were defined at the sow level: 1) the act of chewing the device (yes or no) named “chewing behaviour“, 2) the latency to interact with the device ($\leq$10 min or > 10 min), and 3) the total duration of chewing ($\leq$10 min or > 10 min).

Another outcome was defined to describe pen activity. This outcome was categorised as more than 50% sows lying down at 15, 30 or 45 min after the beginning of the pen-based sampling, or less than or equal to 50% of sows. The cut-off points of the quantitative dependent variables were determined according to the shape of the variable distributions.