

Tongue tip fluid (TTF): A welfare friendly directed population-based monitoring technique.

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Keypoints:

- The TTF is a directed monitoring technique focusing on animals with higher likelihood of harboring the virus.
- Individual diagnostic results comparing tongue tips and serum from same animals were similar.
- The TTF offers a good spatio-temporal representation of the litters in the farrowing room.

INTRODUCTION

Monitoring the Porcine Reproductive and Respiratory Syndrome (PRRS) virus (PRRSv) has become a key element for surveillance and decision making in breeding herds. Bleeding due-to-wean piglets together with the use of processing fluids (PF) are probably the most common monitoring strategies used in U.S. breeding herds. However, those types of sampling have certain limitations. Bleeding requires trained personnel, and it is time consuming as pigs need to be handled properly. A certain number of samples are needed to detect a specific level of prevalence. The lower the prevalence you want to detect, the higher the number of piglets to be bled. The standard number of pigs to be bled at weaning are 30 to detect a prevalence of 10%. When prevalence falls below that threshold (10%), the number of sampled animals needs to be increased substantially to detect at least 1 positive animal. The use of aggregated samples, such as PF, together with the sensitivity of the current diagnostic techniques (e.g., PCR) has helped in improving the monitoring in those low prevalence scenarios. However, the European Union legislation strictly regulates castration and tail docking procedures which makes the use of PF largely unavailable in European countries.

WHY TIP TONGUES? ADVANTAGES AND DISADVANTAGES

With all the limitations and monitoring information in mind we brainstormed to develop a population monitoring technique that could be applied under the E.U. regulations. We focused on the monitoring tissues of stillborn and preweaning mortality as we assumed that the likelihood of finding the virus in those two populations would be high. Furthermore, sampling dead animal had other perks, such as training to perform the sampling is not needed, it is a welfare friendly technique and live animals do not need to be disturbed, the number of animals available to sample are large (around 15% of the total born animals are available for sampling taking into account stillborn and preweaning mortality), most of the litters are represented in the final sample, samples could be aggregated daily, weekly, per batch or per room, and finally, this technique could be used in other stages of the swine production (e.g., nursery).

An aggregate sample with the tails of the dead piglets was tried first. Tails were selected for the easiness to get the tissue and because information regarding the litter sensitivity with this sample type was available. However, several limitations were observed: 1) The bag had little or no fluid to be analyzed, 2) Some media had to be added creating a dilution effect and 3) if scours were present during the collection the sample was not a clean sample and could interfere with the detection. The sample was homogenized with the use of a paddle blender to increase the final fluid. However, results were not that satisfactory.

The tip of the tongues was proposed as a good candidate tissue to be sampled after our first attempt on sampling dead animals (<https://doi.org/10.3390/vetsci8110259>). Some of the positive factors of sampling the tongue are: the tongue is easily accessible and easy to be sampled (remove the tip), the tongue tissue is well irrigated, and when you collect the tongue tip you are also collecting some oral fluids (or traces of amniotic fluid in the case of stillborn). It is recommended to use a filtered bag for the tongue tip fluid (TTF) collection and storing the tissues in the freezer. The result of thawing the bag is a sero-sanguineous clean fluid that can be collected and transferred to a sterile container easily (Figure 1). Furthermore, the CTs obtained from the tongue tip fluid PCR are similar to the CTs obtained with serum. Due to all the above, the quality of the ORF5 sequence is similar to serum also.

The limitations of this sample type are not different from other aggregated sampling. A loss of individual sensitivity due to the dilution effect will occur if only one tongue from a positive animal is collected among a very large number of animals. However, we will overcome the loss of individual sensitivity by sampling larger numbers of animals and therefore increasing the sample sensitivity. Whilst in eradication scenarios the sera were negative to PRRS by PCR, the TTF were still positive.

Finally, the TTF have been tested in the field and has been proven to be a very valuable tool in all the PRRSv monitoring scenarios. This new aggregated technique opens a new possibility in all the scenarios where a strict welfare regulation exists.

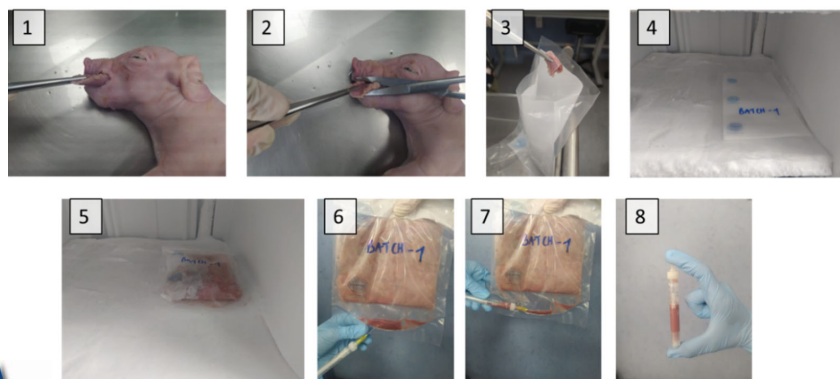


Figure 1. Recommended protocol to obtain the tongue tip fluid (TTF). 1) Extract and 2) remove the tip (aprox. 2cm), 3) introduce it in a new filtered bag, 4) store the bag in the freezer until 5) the monitoring period or batch is completed. 6) Thaw the bag and the tissues, the filter will allow the liquid to concentrate at the bottom of the bag. 7) Remove it with a syringe and 8) place it in a clean and sterile container for submitting the sample to the lab