# PRDC - Validation of a new diagnostic procedure for the diagnosis of PRDC in pigs

**Jensen, M. T.; Hjulsager, Charlotte Kristiane; Kristensen, C. Sonne; Pedersen, K. S.; Haugegaard, S.; Lind, Peter; Larsen, Lars Erik**

*Published in:*
Proceedings of the 23rd IPVS Congress

*Publication date:*
2014

*Document Version*
Publisher's PDF, also known as Version of record

C.044

# PRDC - Validation of a new diagnostic procedure for the diagnosis of PRDC in pigs

MT Jensen[1], CK Hjulsager[1], CS Kristensen[2], KS Pedersen[2], S Haugegaard[2], P Lind[1], LE Larsen[1]

[1]*National Veterinary Institute, Technical University of Denmark, Denmark;* [2] *Pig Research Centre, Danish Agriculture & Food Council, DK-8620 Kjellerup, Denmark,* lael@vet.dtu.dk

## Introduction

Respiratory disease is one of the most important diseases in pigs worldwide (1). Porcine Respiratory Disease Complex (PRDC) is the term for pneumonia caused by multiple pathogens. PRDC has been found to cause morbidity ranging from 30-70% and mortality ranging from 4-20% (1, 2, 3). *M.hyo*, PCV2, SIV and PRRSV are all pathogens that are often diagnosed as primary pathogen of PRDC in swine herds (5, 6).!

In Denmark the laboratory diagnosis of PRDC is routinely performed by a "Porcine Respiratory disease PCR package" which includes testing of 1-3 lung samples from each herd by single tube real-time PCR/reverse transcriptase-(RT-)PCRs specific for *Mycoplasma Hyopneumonia* (*M.hyo)*, Porcine Circovirus type 2 (PCV2), Swine influenza virus (SIV) and Porcine reproductive and Respiratory Syndrome virus (PRRSV). The aim of the present study was to compare the qualitative and quantitative results of the PCR package performed on lungs and on lung swabs.

## Materials and Methods

The study compared the outcome of real-time PCR/RT-PCR performed on pieces of lung tissue samples in parallel with swabs obtained from the same lung lobe. The lungs included in the study were submitted from Danish herds for the diagnosis of PRDC. The standard sample consisted of lung tissue homogenized by bead-beating on TissuelyzerII (QIAGEN) prior to DNA and RNA purification. The other sample for parallel testing was generated by swabbing the same lung lobe with a standard cotton swab. The swab sample was collected in 2 mL of 0.9% saline with 0.1% peptone (peptone). DNA and RNA from both samples were purified using RNeasy Mini Kit and QIAamp DNA Mini kit, respectively (QIAGEN) and tested by real-time PCR/RT-PCR for *M.hyo*, PCV2, SIV and PRRSV. The use of peptone for collection swabs was validated prior to the study and no indication of PCR inhibition was observed. The results were analyzed qualitatively and quantitatively (comparison of Ct-values) for each sample and on herd level.

## Results

A total of 99 paired lung and lung swab samples from pigs with respiratory symptoms were collected and tested. The samples originated from 44 herds. The qualitative results of the real-time PCRs/RT-PCRs of the diseased pigs are listed in table 1. A kappa analysis revealed results between 0.71 and 1.00 when comparing the two sampling methods. The quantitative results were graphically illustrated and analyzed by paired t-test (data not shown).

**Table 4.** The number of positive and negative samples and herds for each of the pathogens: *M.hyo*, PCV2, SIV, PRRSV Type 1 and PRRSV Type 2.

| | | Tissue | | Swabs | |
|---|---|---|---|---|---|
| | | + | - | + | - |
| ***M.hyo*** | Samples | 28 | 71 | 25 | 74 |
| | Herds | 16 | 28 | 14 | 30 |
| **PCV2** | Samples | 28 | 71 | 34 | 65 |
| | Herds | 17 | 27 | 21 | 23 |
| **SIV** | Samples | 25 | 74 | 22 | 77 |
| | Herds | 13 | 31 | 11 | 33 |
| **PRRSV** | Samples | 11 | 88 | 9 | 90 |
| **Type 1** | Herds | 7 | 37 | 5 | 39 |
| **PRRSV** | Samples | 6 | 93 | 6 | 93 |
| **Type 2** | Herds | 3 | 41 | 3 | 41 |

## Conclusions and Discussion

A good agreement was found between the qualitative results of test of the paired samples when compared by kappa analysis but the correlation was not perfect. The quantitative results revealed generally lower threshold cycles (CTs) for the lung tissue samples compared to the lung-swab sample by that indicating that lung tissue is more sensitive than swabs. Ongoing test on alternative swab material and adjusted swabbing technique may increase the sensitivity of this procedure which is cheaper and less resource demanding than tests on lung samples where a homogenization step is needed.

## Acknowledgments

Technicians from *National Veterinary Institute* virology and diagnostic department and *Pig Research Centre*

## References

1. Opriessnig T et al. 2011. Animal Health Research Reviews;12(02):133-48.
2. Harms PA et al. 2002. Journal of Swine Health and Production;10(1):27-32.
3. Kim J et al. 2003. The Veterinary Journal. 11;166(3):251-6.
4. Fachinger V et al. 2008. Vaccine;26(11):1488-99.
5. Fablet C et al. 2012. Vet Microbiol;157(1):152-63.
6. Hansen MS et al. 2010. J Comp Pathol; 143(2):120-31.