PORCINE CIRCOVIRUS TYPE 2: ENZYME-LINKED IMMUNOSORBENT ASSAY (ELISA) FOR THE DETECTION OF ANTIGEN AND ANTIBODIES IN FECES

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Introduction:

Postweaning Multisystemic Wasting Syndrome (PMWS), a swine disease first identified in 1991, in Canada, has been since observed in the United States, Europe, and other Asian countries. Ultimately, the definitive diagnosis of a PCV-related disease will be based on the presence of the PCV2 associated with lesions and clinical symptoms.

Objectives:

Due to high prevalence, detection of serum antibodies is usually of poor value. This study report on the development of an antibody-detection blocking ELISA and an antigen-detection capture ELISA, both optimized for an original use on fecal samples. Concordance with the disease was investigated.

Materials and Methods:

Fecal samples (n=49) were collected from pigs with suspicious clinical signs (diarrhea, progressive weight loss, dermatitis, nephritis) in 22 herds in Britania, France, with history of PMWS demonstrated by immunohistochemistry. For the specificity study, fecal samples (n=139) were collected from pigs from one herd with no history of PMWS. Samples were collected on dry swabs. Following an extraction step of the swabs, samples were incubated in the two ELISAs and compare to disease status established by PCR, histology investigation and clinical symptoms. Both tests use an HRP-labeled specific anti-PCV2 monoclonal antibody for improved specificity.

Results:

Amongst 27 PCR positive fecal samples from infected herds, 21 were found positive for PCV2 antigen and 14 for antibodies. However, a sensitivity of 100% was achieved by adding results from both tests (antigen and/or antibody positive results). When run on samples from herds with no history of PMWS, the antigen detection and antibody detection tests showed a specificity of 96% and 95%, respectively. The specificity of the antibody detection test was significantly higher for young pigs (<8 wk), 98%, than for older ones, 90%. A very broad distribution of the responses obtained with the two tests when sampling infected herds (STD=0.972 for antigen detection and 0.609 for antibody detection), compared to the narrow one for healthy herds (STD=0.080 for antigen detection and 0,288 for antibody detection) was also observed.

Discussion:

Some samples from pigs with severe clinical signs had no ELISA detectable antigen in their feces but presented a very strong fecal antibody response, involving mainly IgA antibodies. This could be explained by a blocking action of the epitopes of the antigen by an excess of antibody, as can be observed for canine parvovirus.

Conclusions:

The original combined use of antigen and antibody ELISAs on fecal samples has demonstrated high concordance with PMWS status at the herd level and may provide ante-mortem tools. More valuable information was obtained than with conventional serology approach and this may assist in herd management decision.

Keywords: Diagnostic, PCV2, PMWS, Feces, ELISA

ESTRATEGIAS PRÁCTICAS PARA EL CONTROL DE PRRS

***Ménard, Julie, Québec, Canada.**

ESTRATEGIAS PRACTICAS PARA EL CONTROL DE PRRS

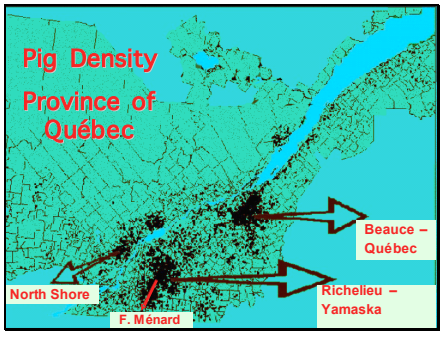
Por Julie Ménard, Agr, DVM

AMVEC, Ixtapa

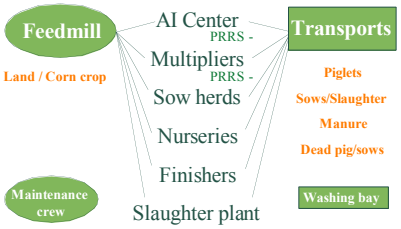
19 de Agosto de 2006

My history on PRRS

- Swine practitioner since 1987
- F. Ménard (family + integrated Cie)
  - *30 000 sows – 650 000 marketed pigs*
- First PRRS case = *1988*
- Since 18 years = *More than 350 PRRS cases*
- Share own experience



F. Ménard structure



My main role

- ⇩ Number PRRS outbreaks
- ⇩ Impact of PRRS

Impact PRRS outbreak depends on :

- ♉ PRRS strain (own or new)
- ♉ Immunity of herd
- ♉ Infection pressure
- ♉ Structure & Management

Control of PRRS

- Depends on respect of < Procedures / Principles

Control measures - PRRS

PHASE I : First clinical signs

- Anorexia in sows
- Few late term abortions
- Uncontrollable piglet scours
- Respiratory distress in piglets
- Early detection
- Diagnostic confirmation
  - Serology on aborted sows
  - Necropsy : Lung lesions / PCR

PHASE II : Beginning of outbreak

- **Minimize the impact**
  - Isolation of sick animals
  - Check environment = ↓stress
  - Stop cross fostering of sick litters
  - Medication - Water (aspirin, AB)
    - Injectable (sows off feed)
  - Stop vaccination for few weeks
  - Needles : 1 / litter
    1 / 5 sows

PHASE III : During the outbreak

1. Stop completely cross fostering (producer insurance)
2. Kill all sick piglets
   - Heavy scour
   - Respiratory distress
   - Weak piglets at birth
   - Starved/runts
3. Collect lungs of killed piglets (freezer)
   - Feedback (vaccination)
   - Gilts acclimatization
   - PCR for sequencing

PHASE III : During the outbreak

4. Internal procedures to reduce spreading (boots, hands, coverall, dedicated personnel)
5. Intensive washing disinfection procedures
6. Strong respect of AIAO between farrowing rooms
7. Advise supplier/transport (biosecurity)

PHASE III : Nurseries and Finishers

1. Temporary nursery (6-8 wks)
   - Prevent contamination PRRS neg by PRRS pos.
2. Strict AIAO by building
3. Biosecurity protocole between rooms/buildings (boots/coverall/hand)

PHASE IV : After the storm – Recovery phase

1. Eliminate subpopulations
   - ♂ Adults – if needed
   - ♂ Gilts acclimatization

   } Feedback
2. Cull problem sows
   - ♂ Aborted + one more reason of culling
3. ↑ number of matings
   - ♂ Start again strongly
4. Start minimum cross fostering
   - ♂ And check!

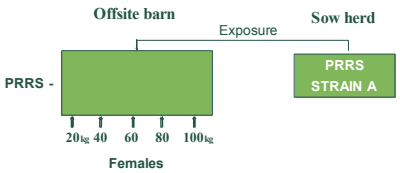
PHASE V : Post mortem crisis and Rebuilding

1. Result of PRRS sequencing
   - ♂ ? Source
2. Biosecurity / Management
3. Routine nursery serology – Empty site
4. Maintain gilts acclimatization

Gilts acclimatization – Following outbreak

- ♂ Expose future replacement gilts to new PRRS strain
- ♂ Offsite finishing barn/Isolated
- ♂ Enter replacements for next 5 months
- ♂ 5 different ages and weights

Gilts acclimatization



Gilts acclimatization - Advantages

1. Protect replacement gilts against new strain
   - Homologous vaccine
   - Immunity
2. Help stabilize sow herd
   - No subpopulations
3. Good performances
4. Strategy for eradication

PRRS Stabilization strategies for sow, nursery and finishers

Elimination or Stabilization

- 1rst step is stabilization
- Long term stabilization plan -> Eradication

Stabilization program

Aim :

Produce PRRS negatif piglets

Stabilization strategy

- **Respect of key principles**
  1. Source of semen and replacement gilts
  2. Gilts acclimatization
  3. Sow building design
  4. 3 sites – AIAO – Single source

My practitioner history

1. Depop – Repop
2. Farrow to finish VS 3 sites
3. Multiples sources
4. PRRS vaccination

Depop - Repop

- **Dense area – Forget it**
  - Unless have to do main repairs

  OR
  - Get rid of many PRRS strains
- **Good if :** Very isolated

  Gilts source and semen PRRS neg

  Very good biosecurity control

Farrow-wean / Farrow to finish

- **Good if :**
  - Isolated
  - PRRS neg or low disease challenge
  - Strict procedures
  - AIAO / room
- **In dense area :** Danger of high disease challenge
- **Our system :** All 3 sites

Multiple sources - Mixing

- Very bad experiences
- We quit
- High disease challenge
- Different sow herds = Different PRRS strains
- Other bugs : Myco, Influenza, Strep, Glassers …

PRRS vaccination

- **Gilts** :
  - We opted to quit in '97
  - Acclimatization to PRRS own strain
- **Finishers** :
  - We use in specific situation
  - In PRRS negative pigs
  - In dense area
  - Protection

Stabilization - Eradication

- **Key principles**
  1. Source of semen and replacement gilt
  2. Gilts acclimatization
  3. Sow building design
  4. 3 sites / Single source / AI AO

Principle #1

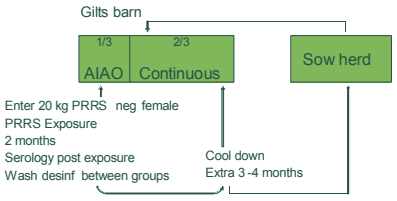
- Boar studs and gilts multipliers must be PRRS negative

Principle #2 – Gilts acclimatization

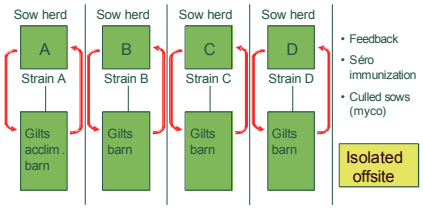
- One of the most important principle if not the most!

Principle #2 – Gilts acclimatization

- Homologous strain exposure



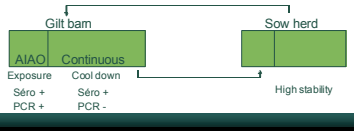
Principle #2 – Gilts acclimatization



Good for PRRS, Mycoplasma, Influenza …

Principle #2 – Gilts acclimatization

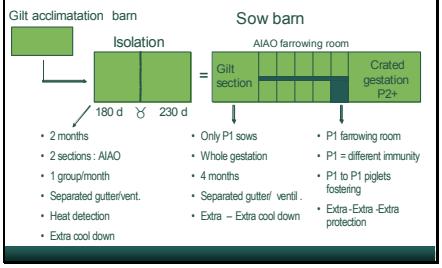
- Production of PRRS negative piglets after 5 to 6 months
- **Eradication** without production break
- Keep acclimatization 1 year
- Then empty gilt barn -> stop exposure
- Long term = Immunity for sow herd



Principle #3 – Sow building design

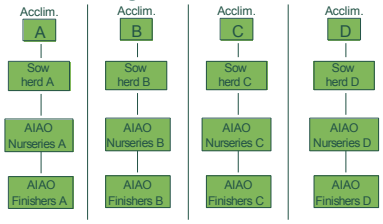
- Keep P1 isolated
- Gilts are the weakest point of our system

Principle #3 – Sow building design



Principle #4

3 sites – Single source - AIAO



Principle #4

3 sites – Single source - AIAO

- **Advantages** :
  - Decrease duration PRRS outbreak – no nursery and finisher on site
  - Production PRRS neg piglets from PRRS pos herds
  - Easier to eradicate PRRS
  - AIAO + single source :
    - Nursery and finisher
    - ↓ disease challenge

Eradication - Stabilization

- Every single principle is important
- Isolated site : Eradication is good
- Dense area : Keep immunity trough acclimatization
- Don't forget new introduction of bugs

"BIOSECURITY"

Biosecurity

- Essential to prevent new outbreaks
- Regular rules : Strict follow up
  - Introduction of materials/visitors/rodents
  - Shower/door lock
  - Pyramid follow up/log book
- Key element = People

***Educate them**

Biosecurity

- 1998 – PRRS sequencing project
  - *Larochelle – Magar – Dallaire – HC and Vet school*
- Great tool : Epidemiology

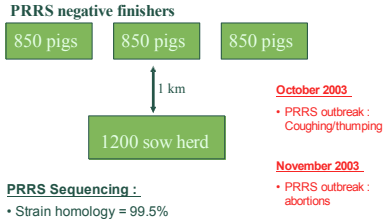
***Source of contamination**

- '98 till 2005 : Over 200 strains identified and sequenced

Biosecurity – PRRS contamination

1. Site – Site – Site
2. Transport
3. Fomites / Environment
4. Unknown

Biosecurity - Site



Biosecurity - Site

- **Regional control**
  - Protect sow herds
    - *Same source of pigs*
  - Sick pigs in restricted area
  - Gilts acclimatization barn isolated
  - Agreements with neighbors
    - *Same source/same strain*

Biosecurity - Transport

1999 – 2000

- Comingling of 10 sources
- Garbage run
- Same transport
- Feb '99 till April 2000
- 5/10 sow herd abortion breaks
- PRRS sequencing 98.5 to 99.5% homology

Consequences of these findings

- **Transport reform**
  - As soon as evidence of PRRS outbreak
    - *Transport viremic pig end of the day*
  - Move to single source transport
    - *No pigs in truck from other source close to sow herds*
  - Some contamination trough fresh feces and shavings
  - **Very important :** washing/disinfection procedures and drying > 8 hrs

Biosecurity - Fomites

2001

Nursery #1 | Nursery #2 | Nursery #3

- Very isolated
- PRRS negative
- Respiratory problem in barn #1
- No PRRS outbreak in sow herd
- Strain identification :
  - *Homology 99.7% to sow herd in Ange-Gardien (170 km away)*

Biosecurity - Fomites

- **Possible explanation**
  - Technician did hernia repair 7 days before beginning clinical signs
  - Use poorly disinfected materials (rope)
  - Did hernia repair the day before on piglets from Ange-Gardien sow herd

What we also learned with sequencing

- New PRRS introduction with few clinical signs

Sequencing M16 Ange-Gardien

		Sequence	229480
05-26-2004	M16 Ange-Gardien	229480	ID
11-09-2004	M16 Ange-Gardien	257115	90.2%
10-13-2005	M16 Ange-Gardien	308775	89.0%
01-06-2006	M16 Ange-Gardien	320973	93.6%

- Adjustment of gilts acclimatization

PRRS sequencing

- Great tool to understand
- Bring some explanations to contamination
- Help convince to take preventive measures ($$)
- Prevention is best
- Biosecurity is fondamental

Summary

1. In PRRS outbreak, minimize the impact
2. Following the outbreak, restabilize sow herd trough gilts acclimatization
3. In any time, strict biosecurity to prevent PRRS introduction