

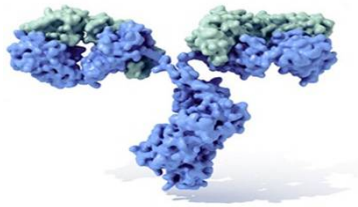
The "ideal" diagnostic test is accurate, reliable, sensitive, specific, and consistent (repeatable) and is cost effective (i.e. the costs are balanced by the usefulness of the information derived). Preferably the ideal diagnostic test will work on easily obtainable and easily preserved specimens. The specimens should preferably come from living animals (e.g. blood, feces, nasal swabs, etc.). Additionally, ideal diagnostic tests can be accurately performed locally (on farm, in veterinarian's clinic or at a regional lab).

An ideal diagnostic test for swine health would possess several key characteristics to ensure accurate and reliable detection of diseases or health issues in pigs. Here are some of the essential criteria for an ideal swine diagnostic test:

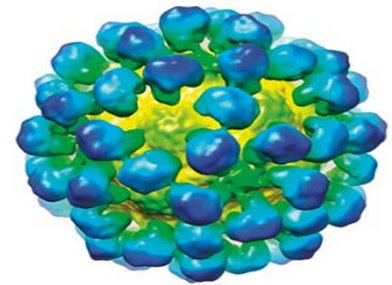
1. Sensitivity
2. Specificity
3. Rapid Results.
4. Cost-Effectiveness
5. Non-invasive or Minimally Invasive
6. User-Friendly
7. Accuracy
8. Long Shelf Life
9. Portability (on farm).
10. Ability to Detect Multiple Pathogens
11. Validation and Standardization
13. Regulatory Approval
14. Epidemiological Data to be used and take decisions

What?

INDIRECT DIAGNOSTIC
Antibodies detection



DIRECT DIAGNOSTIC
Antigen detection



INDIRECT DIAGNOSTIC

Antibody detection

ELISA



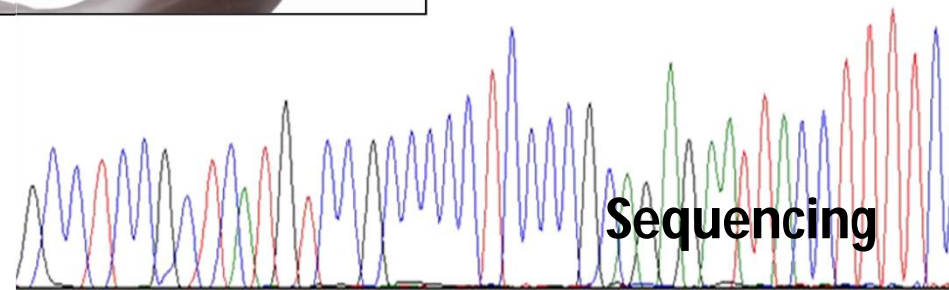
DIRECT DIAGNOSTIC

Antigen detection

Bacteriology



PCR



INDIRECT DIAGNOSTIC

Antibodies detection

ELISA



- Herd screening
- Serum profiles
- Vaccination response
- Gilt acclimatization success

Pros:

- Cheap
- High throughput volume
- Speed

Cons:

- Variable specificity and sensitivity depending on the test

- Individual Disease diagnosis
- Antibiotic resistances and treatment
- Autogenous vaccines development

Pros:

- Cheap

Cons:

- Sensitivity and Specificity depending on the bacteria (alive or not)
- Sampling is crucial

DIRECT DIAGNOSTIC Antigen detection



etection and selective amplification of a
ment of the virus/bacteria:

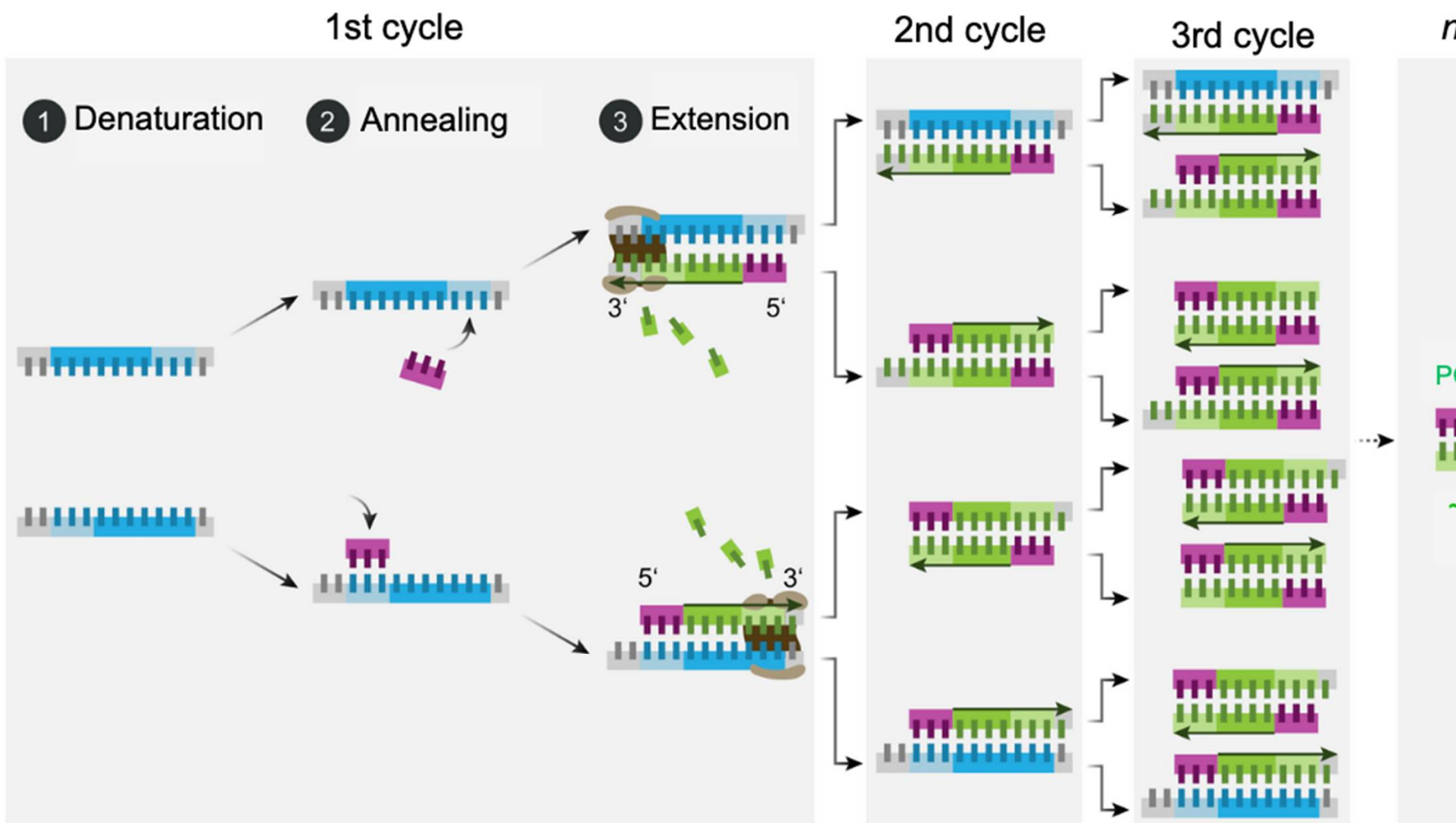
Known
Unique / Specific
Conserved

DIRECT DIAGNOSTIC Antigen detection

PCR



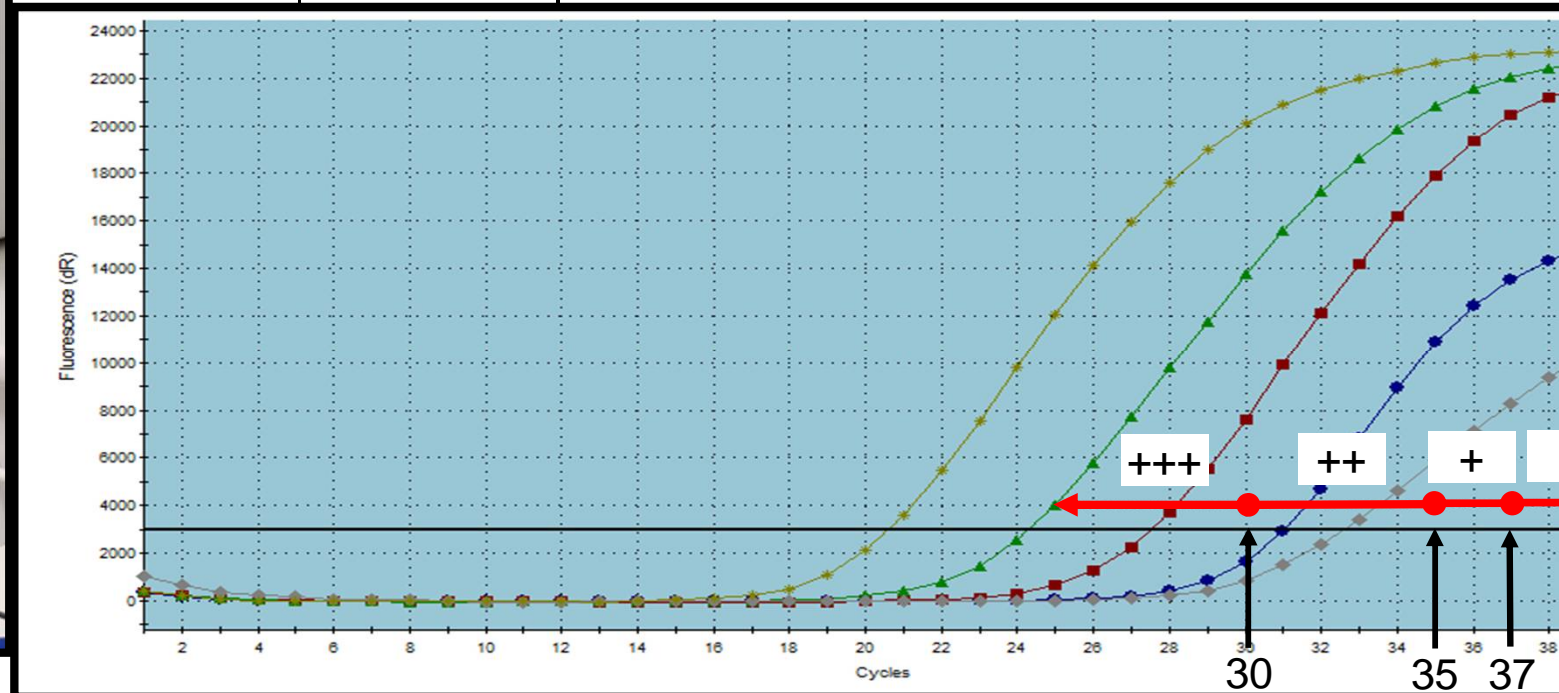
plate
ence of
t
3'
5'
primers
erase



Ct value



Ct value	Score	Interpretation
≤ 30	+++	High quantity of DNA detected
$>30 \leq 35$	++	Medium quantity of DNA detected
$>35 \leq 37$	+	Low quantity of DNA detected
>37	-	No DNA detected



- Detect of DNA/RNA of targeted pathogen
- Disease surveillance and monitoring
- Biosecurity monitoring
- Gilt acclimatization success

Pros:

- High sensitivity
- Multiplexing
- Early disease detection
- Quantification

Cons:

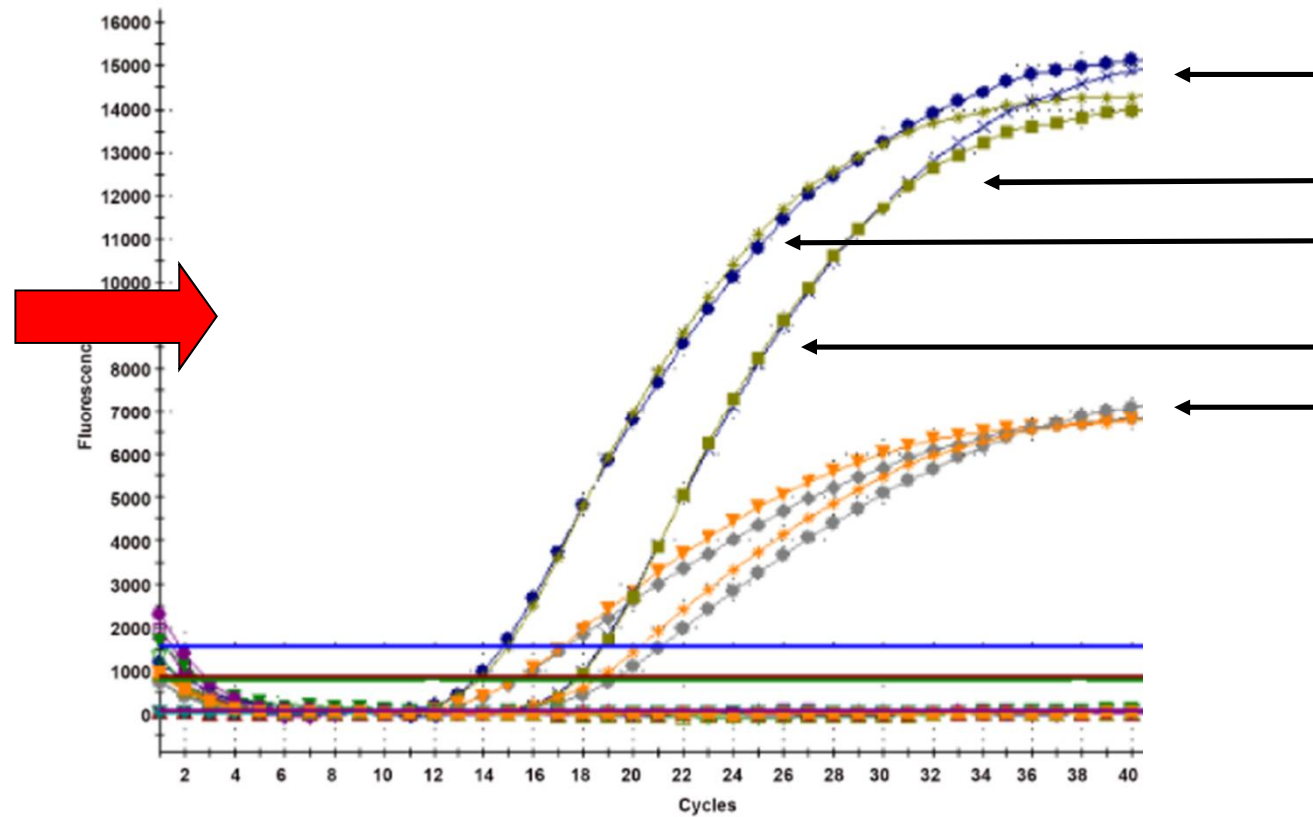
- Expensive
- Sample contamination

DIRECT DIAGNOSTIC Antigen detection



ultiplexing: Enteric colibacillosis

Detection of virulence factors. PCR



Piglet Enteric Diseases FTA card sampling

E.coli

F4 / F5 / F6 / LT

C.perfringens

α / β / ϵ

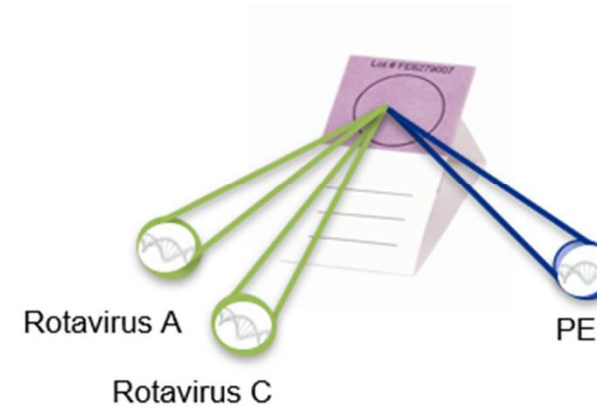
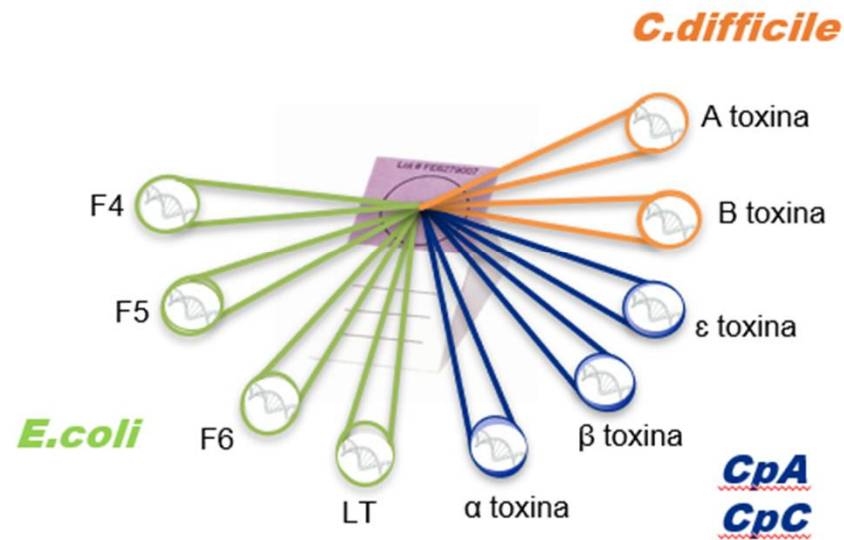
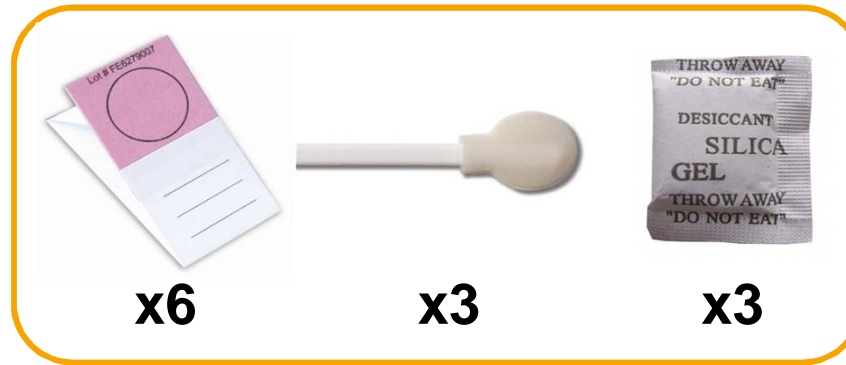
C.difficile

A / B

Rotavirus

A / C

PED



PCR from ORAL Fluids

PRRSV, SIV, ASFV, CSFV, PPV, PCV2, PCV3, **hev**, PEDV, PDCoV, SVA
MHYO, *Bordetella bronchiseptica*, *Pasteurella multocida*, *E. Coli* VTEC.

RNA viruses very temperature and time sensitive

Easy monitoring of large populations



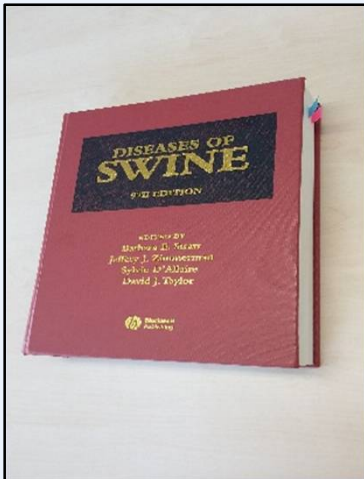
EDEMA Disease Oral Fluids Test



Basics of Sequencing (ORF5 PRRS)

How is it performed?

PARTIAL



Specific known Genome
segment
Sanger

COMPLETE



Complete Genome of all the
microorganism from the sample
Next generation sequencing (NGS)

Basics of the sequencing



RSV genome: 15,300 bases

ORF7: 387 b (2,5%) → RT-qPCR → **DETECTION**

ORF5: 606 b (4%) → RT-qPCR + Sanger sequencing → **CHARACTERIZATION**

ORF2-7: 3.200 b (21%) → RT-qPCR + secuenciación NGS → **RECOMBINANTS ANALYSIS**

How is it performed?

le datos
analizada

cunales

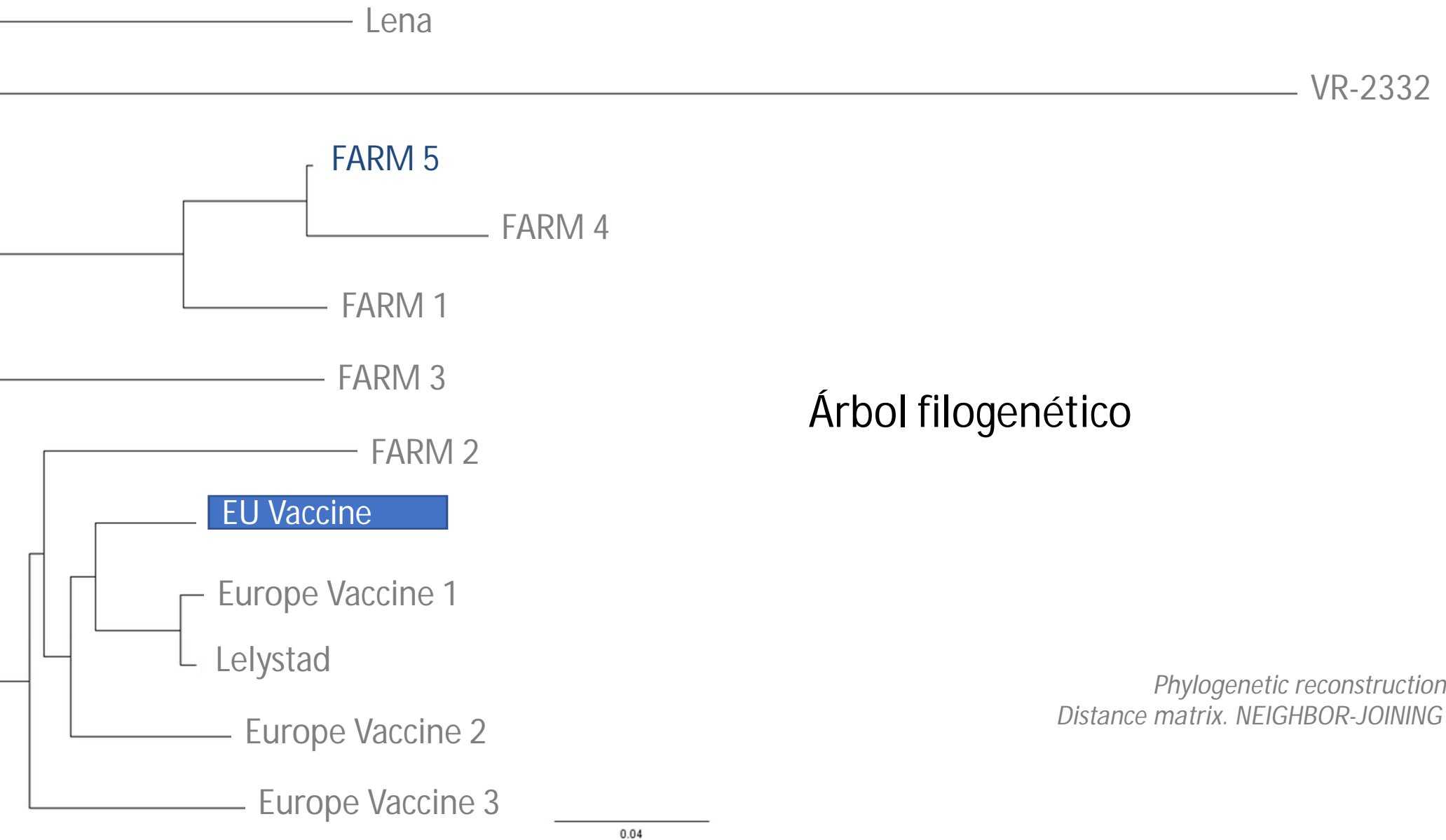
epas de
ferencia

	FARM 5	FARM 4	FARM 3	FARM 2	FARM 1
FARM 5		94.884%	83.003%	83.828%	93.069%
FARM 4	94.884%		79.703%	80.033%	88.449%
FARM 3	83.003%	79.703%		80.693%	84.158%
FARM 2	83.828%	80.033%	80.693%		83.828%
FARM 1	93.069%	88.449%	84.158%	83.828%	
EU Vaccine	87.459%	83.333%	84.323%	89.439%	86.469%
Europe vaccine 1	87.459%	83.993%	85.149%	88.779%	86.799%
Europe vaccine 2	86.304%	82.343%	84.818%	86.139%	85.314%
Europe vaccine 3	83.333%	79.538%	84.818%	85.809%	82.343%
Lelystad strain	87.624%	83.828%	85.314%	89.109%	86.964%
Lena strain	81.353%	77.393%	81.188%	82.673%	80.693%
VR-2332 strain	64.521%	61.551%	63.531%	63.366%	63.531%

Identidad nucleotídica (%)



Cómo se hace la secuenciación?



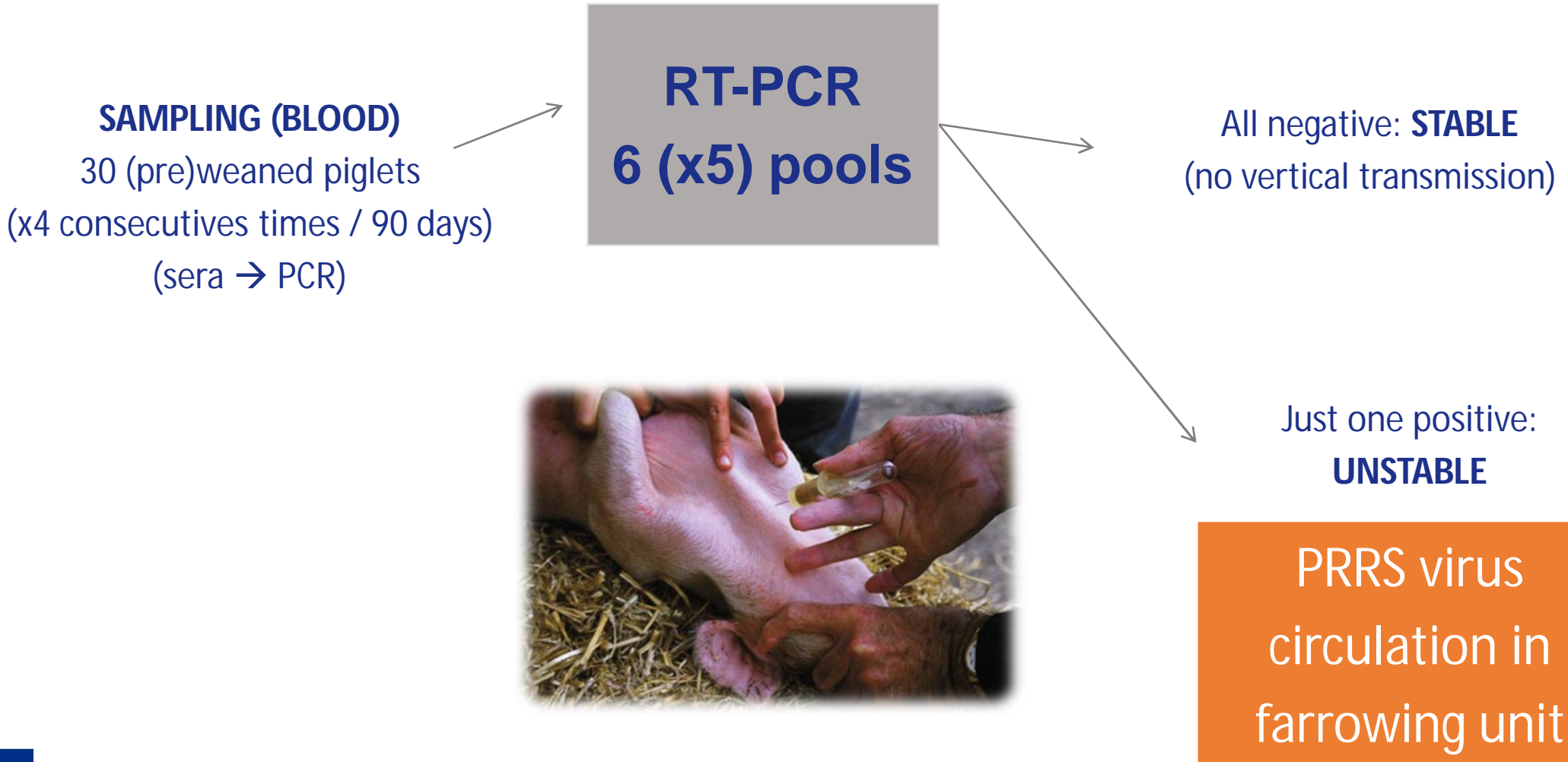
Árbol filogenético

*Phylogenetic reconstruction me
Distance matrix. NEIGHBOR-JOINING alg*

Why? Start with the Basics.

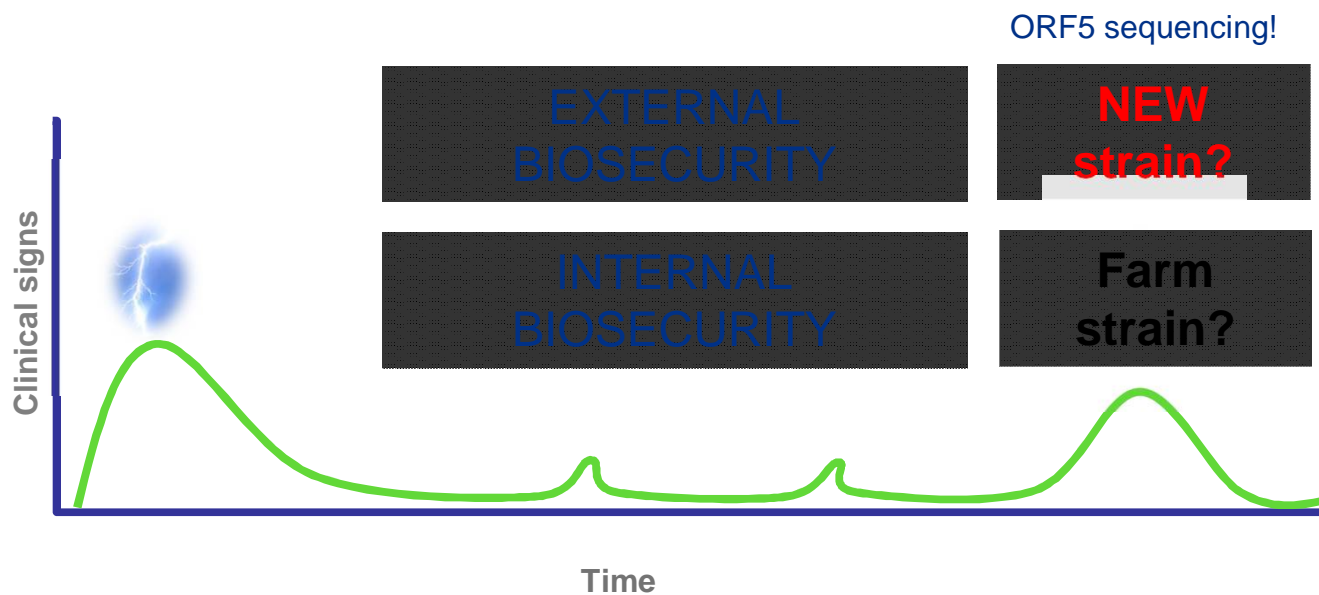
1. Surveillance: Testing expected negative farms for a particular pathogen. (Farms expected negative)
2. Monitoring: Testing positive farms for a particular pathogen
3. Disease Investigations: Farms with clinical disease of unknown causes

Classifying PRRSV Farm status using PCRs



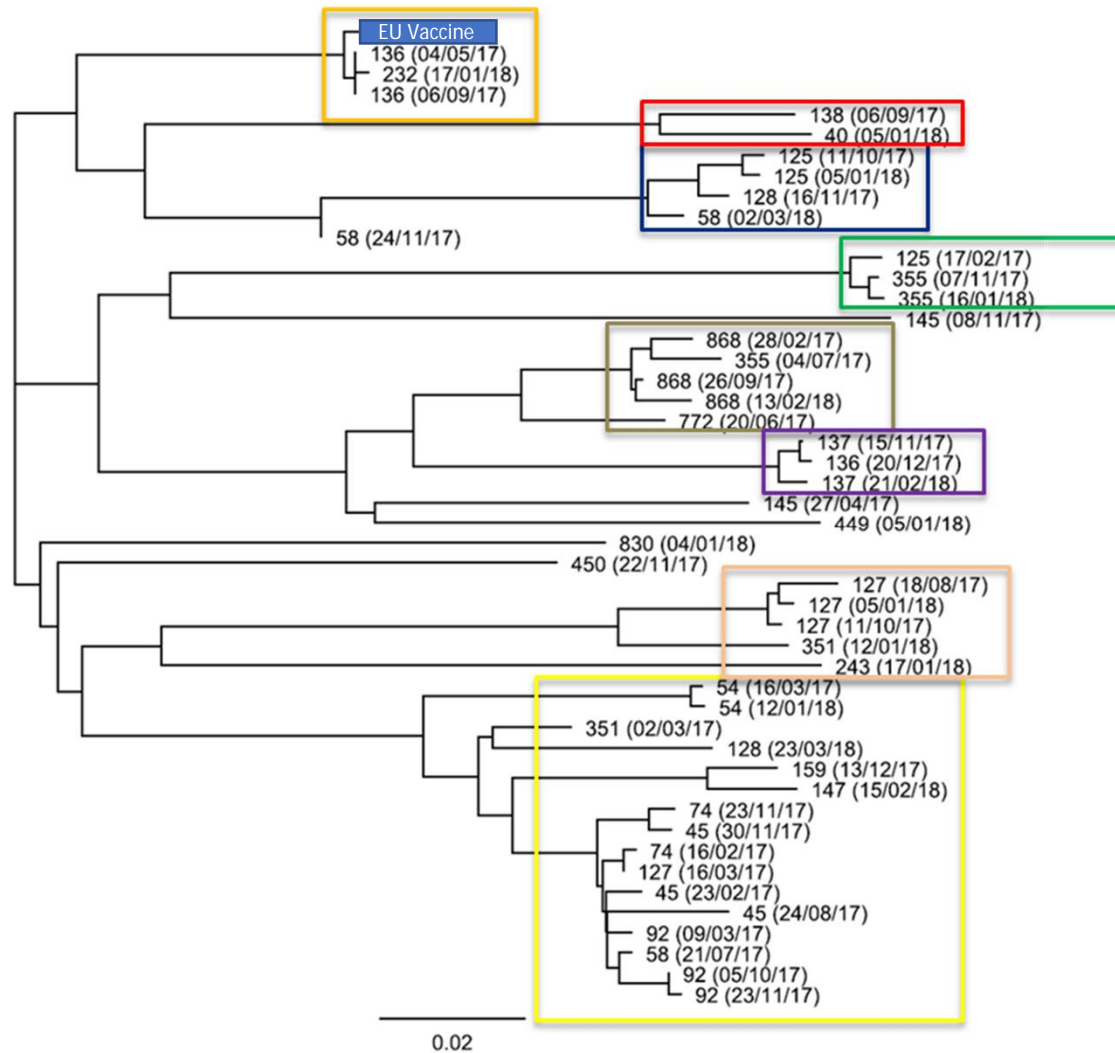
ORF5 sequencing: when?

- 1. Homology between strain and phylogenetic tree:**
 - ¿Similarity to a vaccine strain? ¿Or a reference strain?
 - ¿Similarity to a previously detected strain in the same farm/company? ¿Region?
- 2. Taking better decisions about management and biosecurity:**
 - ¿The outbreak has been caused by a new strain? ¿Already detected strain in the past?
- 3. PRRS strains mapping in a company/region**
 - ¿Which strains do they have in common? ¿Which strain shows the highest or quickest dissemination?
- 4. It's not useful for predicting vaccine efficacy**
 - A higher similarity between field strain and vaccine strain doesn't mean higher protection/efficacy.



Perform routine sequencing minimum once a year

ORF5 Phylogenetic study



ORF5 sequencing is a extremely good help to understand where we need to put the focus: external biosecurity or internal biosecurity

Farm	January	February	March	April	May	June	July	August	September	October	November	Dicember
1		Yellow									Yellow	
2	Green			Green								Green
3		Green			Green							
4												
5	Green						Blue					
6			Blue								Green	
7	Blue									Blue		
8		Orange										

crossprotection conferred by each
MLV PRRS VACCINES can be
predicted by using sequencing?

ORIGINAL RESEARCH

PEER REVIEWED

Genomic homology of ORF 5 gene sequence between
attenuated live vaccine virus and porcine reproductive and
respiratory syndrome virus challenge isolates is not predictive
of vaccine efficacy

Eric M. Vaughn, DVM, PhD; Francisco J. Pallarés, DVM, PhD; Dachrit Nilubol, DVM, PhD; Amy L. Vincent, DVM, PhD; Eileen L. Halbur, DVM, PhD, Diplomate ACVM; Eric M. Vaughn, DVM, PhD; Michael Roof, DVM, PhD; Patrick G. Halbur, DVM, PhD

No!



ELSEVIER

The Veterinary Journal

Volume 175, Issue 3, March 2008, Pages 356-363

Similarity of European porcine reproductive and respiratory syndrome virus strains to vaccine strain is not necessarily predictive of the degree of protective immunity conferred

Cinta Prieto, Esther Álvarez, Francisco J. Martínez-Lobo, Isabel Simarro, José M. Castro  



Next Generation Sequencing Applications



- 1) Viral Discovery
- 2) Pathogen Characterization

CASE 1: Outbreaks of vesicular disease in 2014 in Brazil



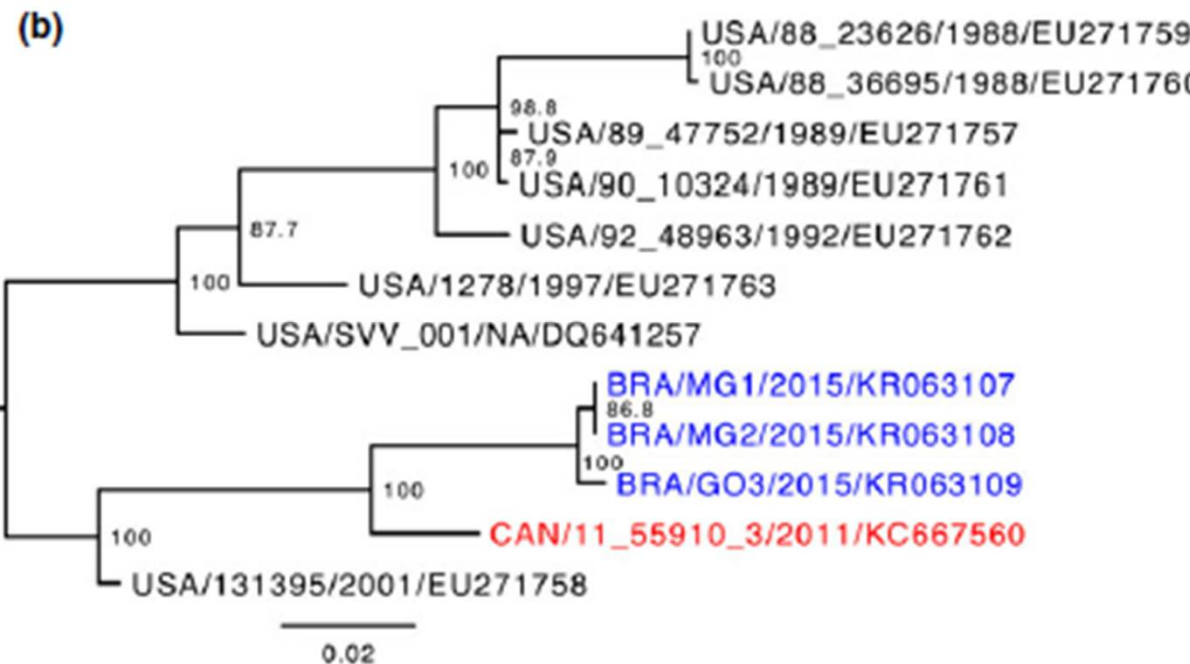
annucci et al. 2015. *Transbound Emerg Dis* 62(6):589-93.

CASE 1: Outbreaks of vesicular disease in 2014 in Brazil

INGS:

Seneca Valley Virus

Senecavirus A)



Vannucci et al. 2015. Transbound Emerg Dis 62(6):589-93.



) Viral Discovery

2) Pathogen Characterization

- Comparison of two or more genomes
- Recombination analysis
- Typing
- Prediction of virulence
- Prediction of antibiotic resistance
- Prediction of immune response

CASE 2: PRRSV re-break in a sow farm

Original outbreak: 1-7-4 PRRSV

Serum inoculation + herd closure, farm stable

Recent outbreak: 1-7-4 PRRSV, 98.5% similar (Orf5)

Is this the same strain?



CASE 2: PRRSV re-break in a sow farm

First PRRSV outbreak: PARTIAL genome sequence

- 9 Contigs (fragments) - 118-370 nt
- 11% of the genome

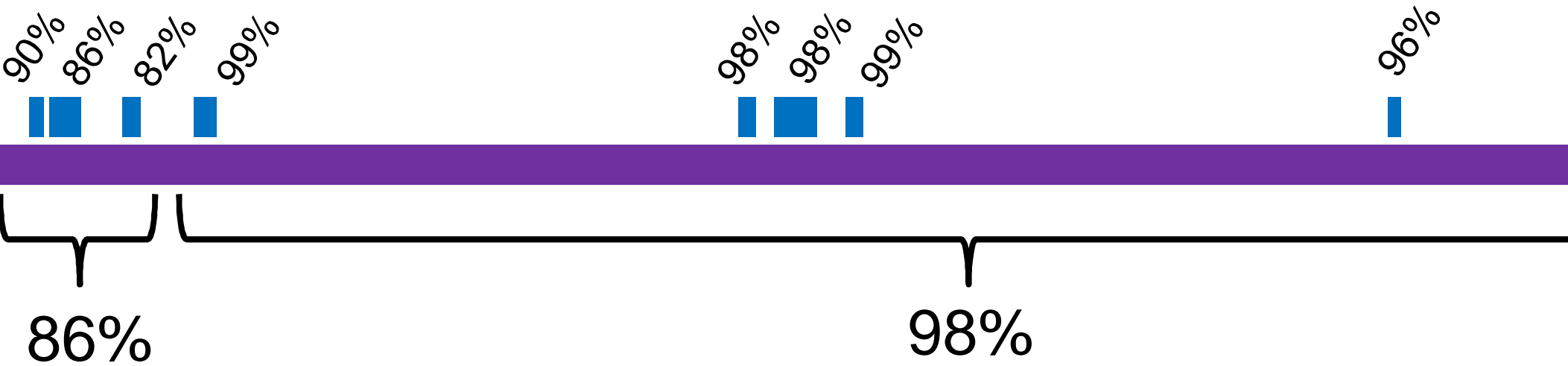
Second PRRSV outbreak: whole genome sequence

ORF5 comparison: 98.5% similar

Partial genome comparison: 94.2% similar



CASE 2: PRRSV re-break in a sow farm



CASE 2: PRRSV re-break in a sow farm

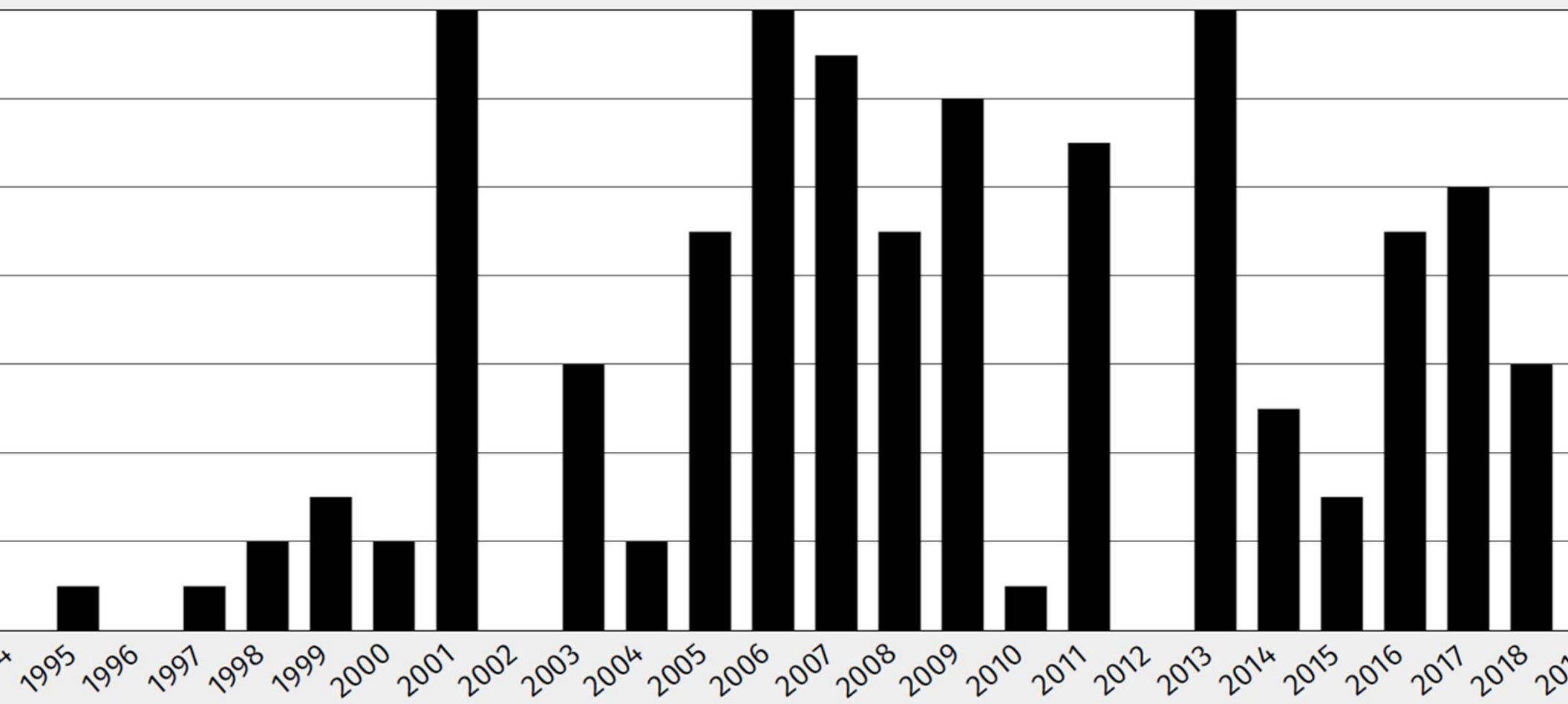
The first and second outbreaks were caused by different strains of PRRSV

Where did the second strain come from? Lateral introduction?
Recombination?

Orf5 vs Whole Genome

	ORF5	WHOLE GENOME
Genome coverage	4%	100%
Turnaround time	2 days	2 weeks
Cost	\$100	\$350
Success rate	High	Medium
Databases	Excellent	Poor

New porcine molecular procedures introduced per year. University of Minnesota Veterinary Diagnostic Laboratory



- Increase Data Analysis from lab results
- Artificial Intelligence
- PRECISION LIVESTOCK FARMING
- POINT OF CARE=need



Data Analytics in Animal health

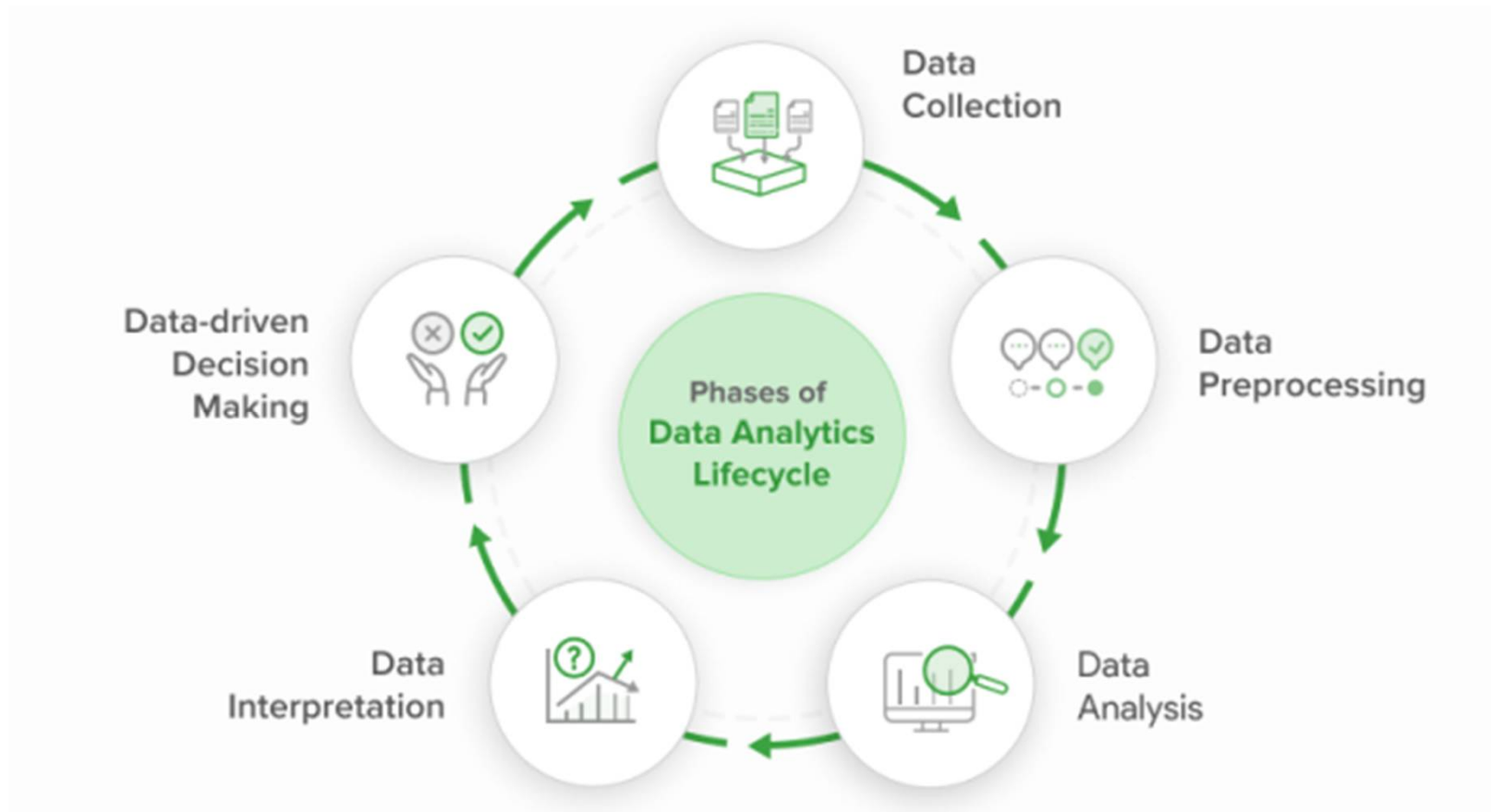


About ▾

Data-driven control and prioritisation of non-EU-regulated contagious animal disease



Data Analytics in Animal health





UNIVERSITY OF MINNESOTA

Scoring of swine lung lesion images by an artificial intelligence algorithm: A comparison to human expert scorers

Author: Robert Valeris-Chacin

Corresponding Author: Maria Pieters, University of Minnesota, St. Paul, MN, USA

Presenters: Robert Valeris-Chacin, Beatriz Garcia, Marina Sibila, Albert Canturri, Isaac Ballarà Rodriguez, Ignacio Ballester, Ramon Jordà Casadevall, Maria Pieters



IPVS2022

26th international pig veterinary society
congress - rio de janeiro - brazil

**Artificial intelligence as a new method of
assessing enzootic pneumonia and
atrophic rhinitis lesions**



Advantages of using AI in lung scoring:

Automated: it only requires taking photos and upload them in the system, so we do not need to send specialized technicians to the slaughters.

Reliable: it is a fully objective process, where the subjectivity of the evaluators is eliminated, and the images are always evaluated according to the same set of criteria and levels of accuracy, so the system is standardizing the process of lung scoring and snout lesions assessment.

Simple: it is a very user-friendly system, where only the farm data has to be completed and the images of the lungs or nasal turbinates' must be added for the system to perform the evaluation automatically. The system generates a report automatically to make interpretation easier for the user.



POINT OF CARE

POC diagnostics are analytical devices and other tests that provide rapid diagnostic capabilities, without the need for core laboratory facilities

The emergence of novel pathogens, the modern farming systems, and the complexity of globalized supply chains and trade networks make animal production susceptible to disease outbreaks.

Rapid, low-cost, and reliable field diagnosis is gradually becoming indispensable to support evidence-based disease-control strategies in veterinary medical practice.



Lateral Flow

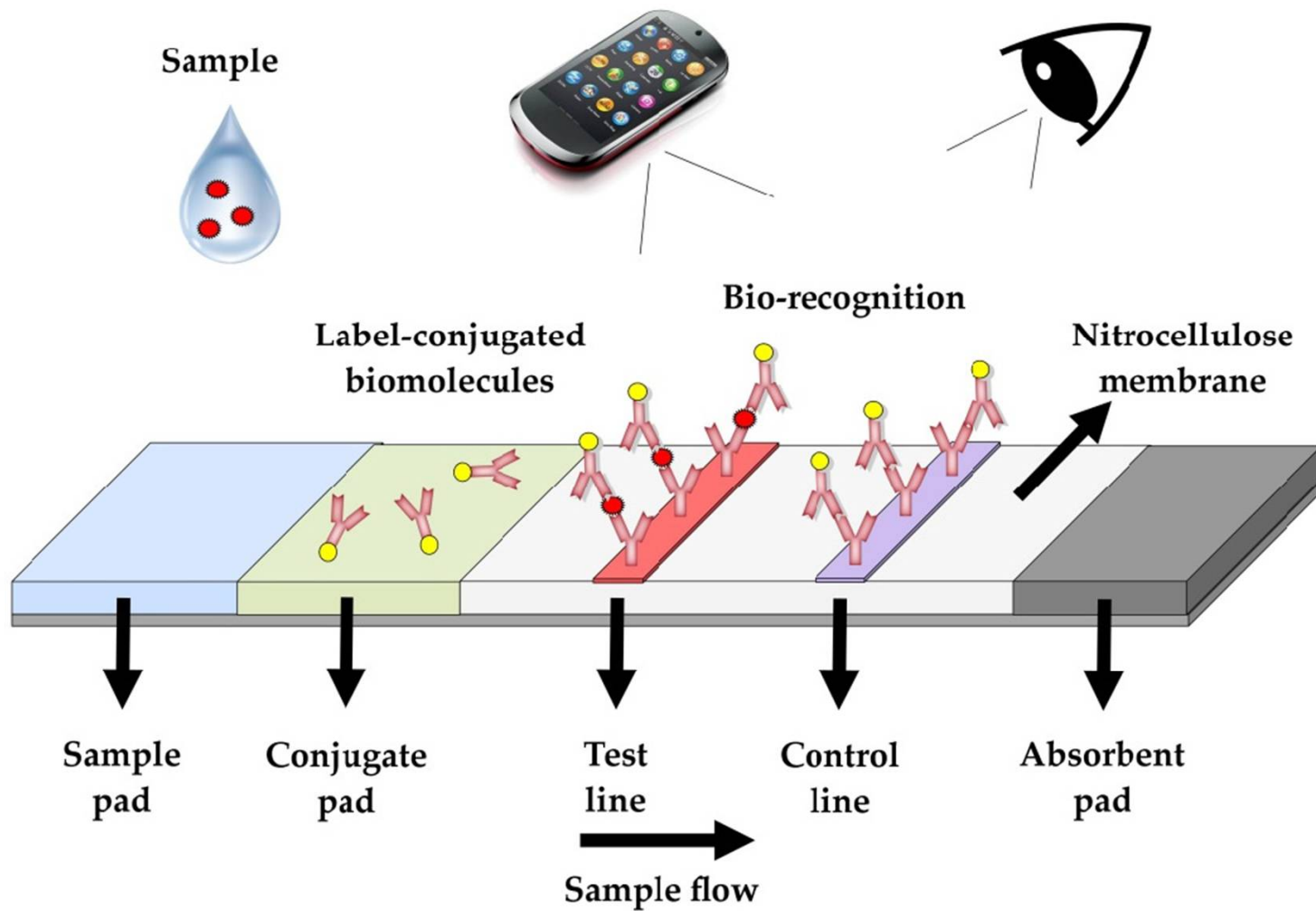
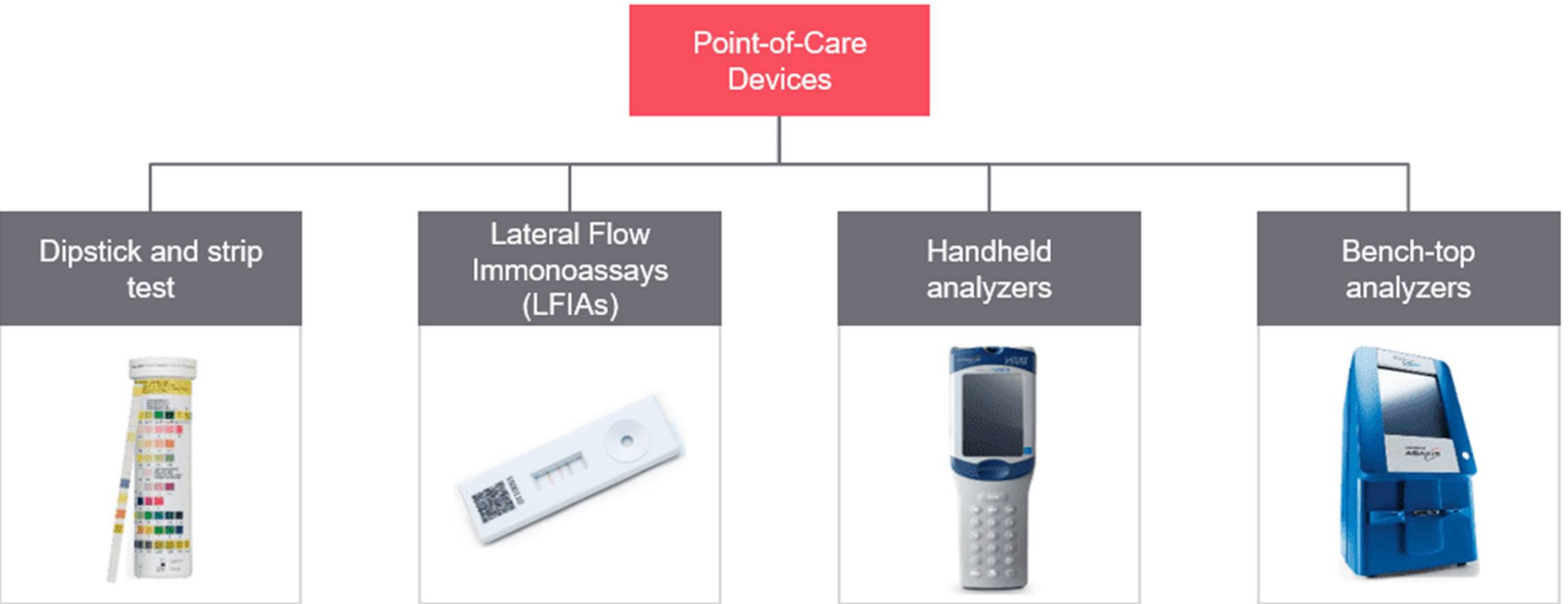


Figure 1. Principle of LFA sandwich format.

FIGURE 1: Classification of PoC Devices



Source: Secondary Research

Challenges of POC

POC diagnostics should focus on validation using complex clinical samples and large animal populations

POC diagnostics must be low-cost and simple.

POC devices should be portable and multiplexed.

Only a few POC devices targeting bacteria, protozoa, and parasites have been developed,

DEFINITION OF PRECISION LIVESTOCK FARMING (PLF)

PLF

Manage individual animals by *continuous* real-time monitoring of health, welfare, production/reproduction and environmental impact

Real-time measuring and analyzing which provides warnings and direction to the area of needed attention – actionable

A SET OF TOOLS THAT ALLOWS
FEWER PRODUCERS TO MONITOR
MORE ANIMALS.

CAN BE USED TO MONITOR ANIMAL
HEALTH AND WELFARE AND REDUCE
ENVIRONMENTAL IMPACT WHILE
ASSURING THE PRODUCTIVITY IN THE
PROCESS (PUBLIC PERCEPTION)

IS A MULTIDISCIPLINARY SCIENCE THAT
REQUIRES COLLABORATION AMONG
ANIMAL SCIENTIST, PHYSIOLOGISTS,
VETERINARIANS, ETC.



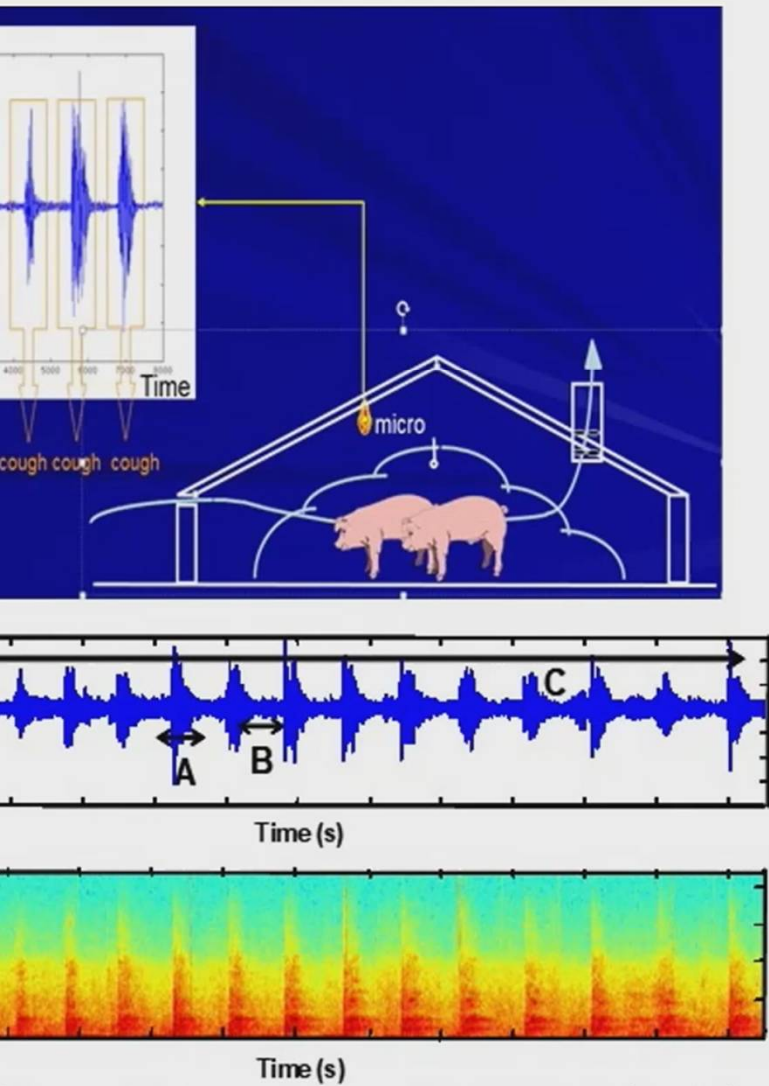


BOOST YOUR FARM BIOSECURITY

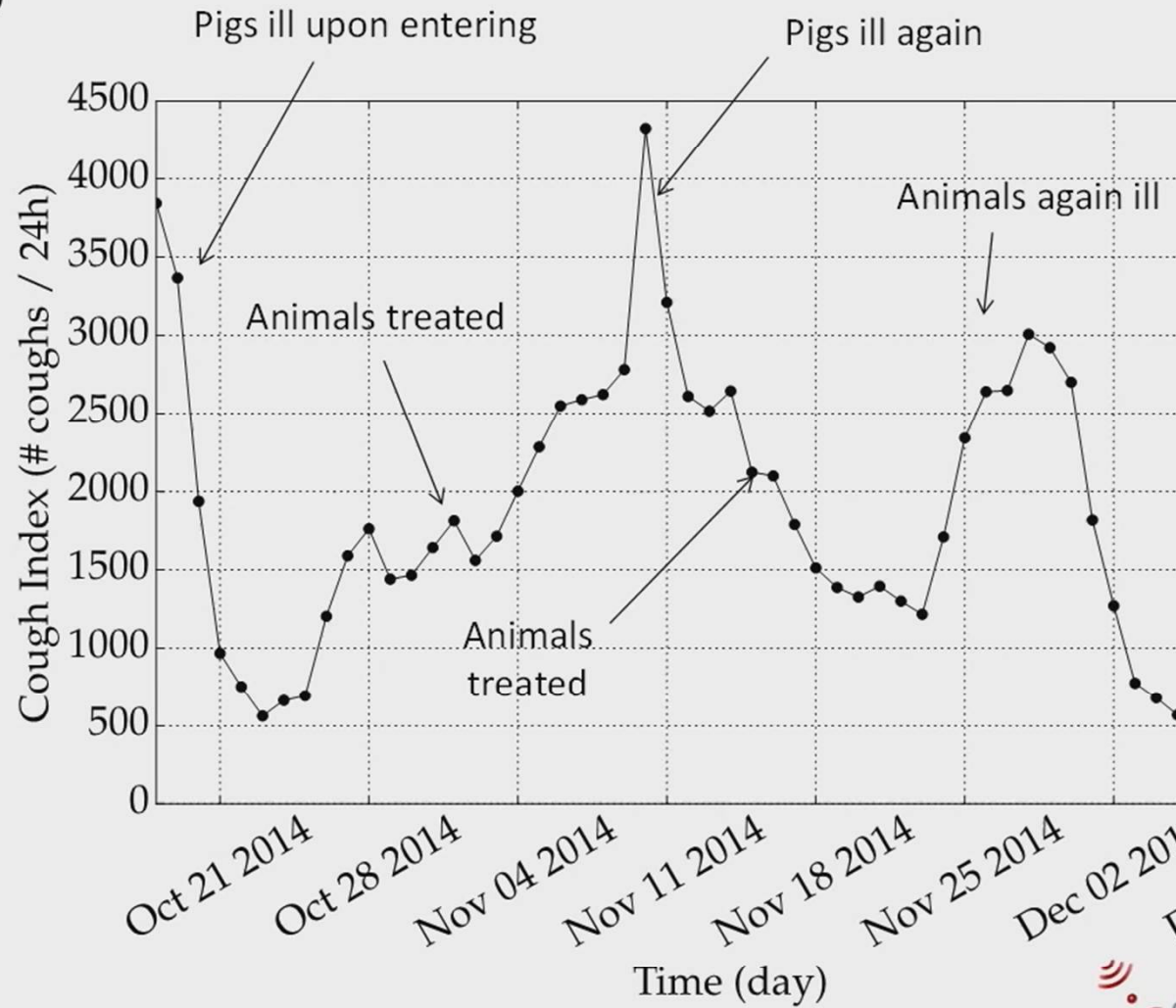
What are the benefits of using Farm Health Monitor?

- ✔ **Early disease detection** with real-time input into cloud hosted health records and immediate and targeted communications
- ✔ **Accurate health and mortality tracking** with a digital recording and reporting health system
- ✔ **Increase efficiency** by connecting everyone on your animal health team and recording all information into one easily-accessible system

Cough Monitoring



) In collaboration with UNIMI (Italy)



Started by Van Hirtum and Berckmans (2000)



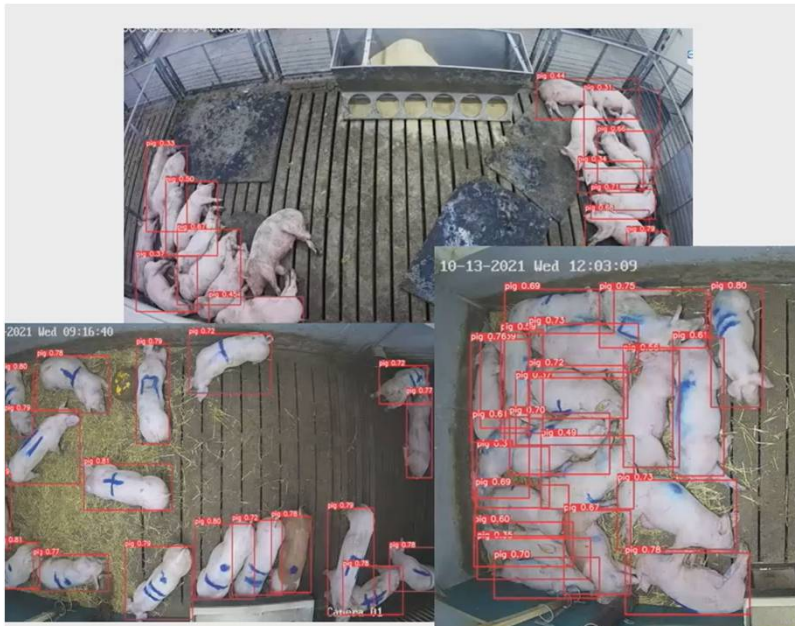
EU

FP7

The Future

Integrated pig production monitoring: Health/ welfare / productivity

Using Cameras → 1 device per pig pen



Conclusions

The ideal test does not exist, it is necessary to combine different tests to find solutions to the clinical cases.

No way!! Nowadays, swine vets must be trained to use and interpret all the diagnostics tools available in order to make the best treatment decisions.

On Farm diagnostics and increase of sensors will bring the possibility to have earlier diagnostics without the need to wait lab results.

