

# Non-invasive sampling of wild boar and the role of carcasses in the epidemiology of ASF



EFSA, 23-25 Nov 2015



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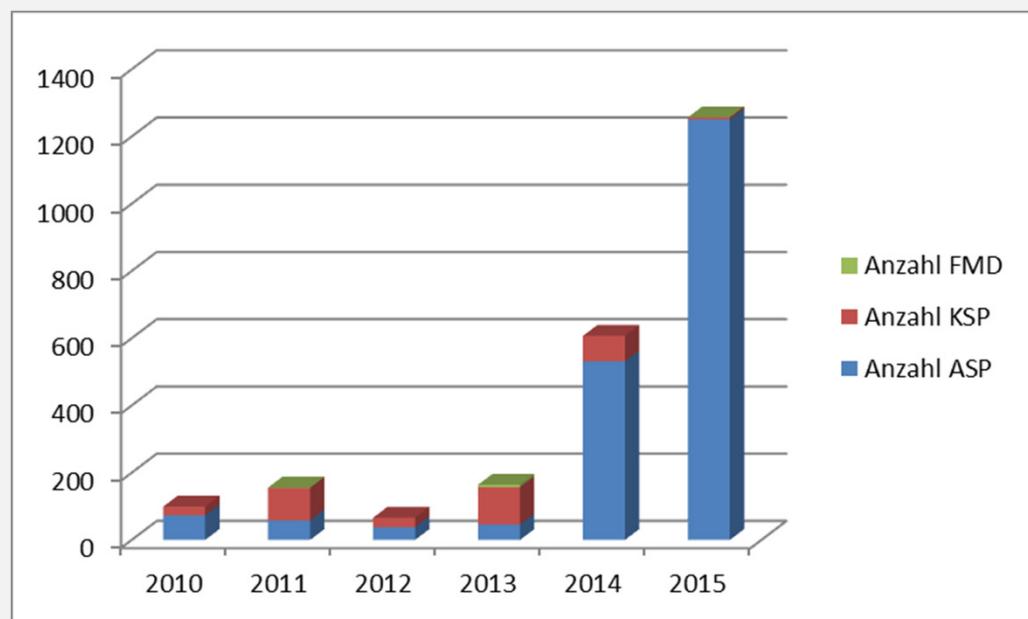
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# Role of wild boar in the transmission of ASF, CSF and FMD in Eurasia

- ❖ FMD incursion to Bulgaria (2011)
- ❖ CSF outbreaks in Latvia and Lithuania and other European countries (2011-2013)
- ❖ ASF epidemic in the Caucasus, Russian Federation, Lithuania, Latvia, Poland and Estonia (2007-2015) - still ongoing
- ❖ remarkable increase of wild boar populations in Europe



Datenquelle: empres- i Grafik: FLI



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# Wildlife surveillance

- ❖ Wildlife surveillance mostly linked to hunting
- ❖ Sampling rather irregular
- ❖ Hunting seasonally limited
- ❖ Rarely adequate number of samples collected
- ❖ Mostly serological tests performed - only retrospective analysis

**Solution:** Non-invasive sampling of wild boar?

**Advantage of non-invasive sampling:**

- Independent of hunting (access to areas where hunting is limited/prohibited)
- Collective sample: Disease detection on basis of herds
- No disturbance of natural behaviour



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# pSWAB material

Cotton ropes, native (without treatment), 10 cm length, 8 mm

*Matrix: Cereal based with „marzipan flavor“ (CSFV vaccine bait, IDT Dessau)*



All pictures: Susan Mouchantat



CSF vaccine baits for wild boar



## Toxicity test:

- Native cotton rope no impact on virus stability (Mouchantat et al., unpublished)

## Saliva volume capacity:

Varies by size of animal, 1-2,65 ml

**p**athogen

**S**ampling of

**W**ild

**A**nimals with

**B**aits

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# pSWAB - The Work Flow



Susan Mouchantat



Jan Forth



Jan Forth



Jan Forth

tested in four animal trials  
with wild boar, two in  
domestic pigs  
(ASF, CSF, FMD)



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# Experimental infection of wild boar with ASF

Arch Virol (2015) 160:1657–1667  
DOI 10.1007/s00705-015-2430-2



ORIGINAL ARTICLE

## Course and transmission characteristics of oral low-dose infection of domestic pigs and European wild boar with a Caucasian African swine fever virus isolate

Jana Pietschmann<sup>1</sup> · Claire Guinat<sup>5</sup> · Martin Beer<sup>3</sup> · Valery Pronin<sup>2</sup> ·  
Kerstin Tauscher<sup>3</sup> · Anja Petrov<sup>1</sup> · Günther Keil<sup>4</sup> · Sandra Blome<sup>1</sup>



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Experimental study: 17 days

<i>Animals</i>	6 wild boar (8 Mo, ≈ 15 kg)
<i>Inoculation</i>	oronasal 100 HAU / ml (Minimum dosis) Armenia/08
<i>pSWABs</i>	max. 10 distributed (DPI 3-8 and DPI 16+17) collected next days
<i>Collection blood/oropharyngeal sample</i>	every 2 days under sedation
<i>Diagnostic</i>	qPCR, serology



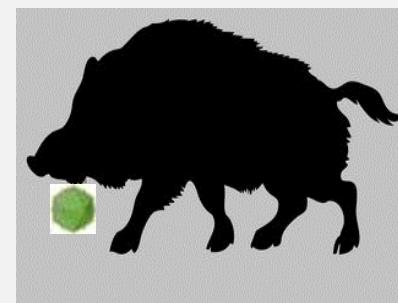
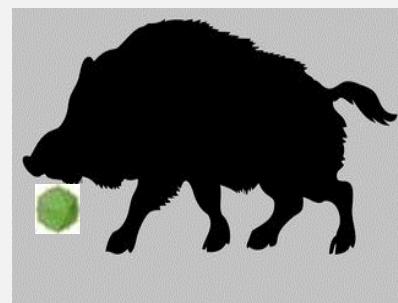
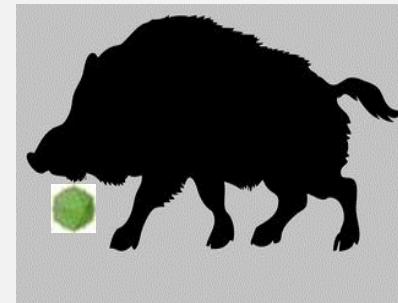
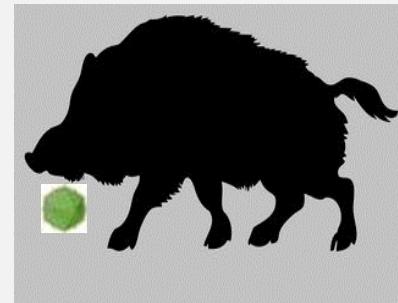
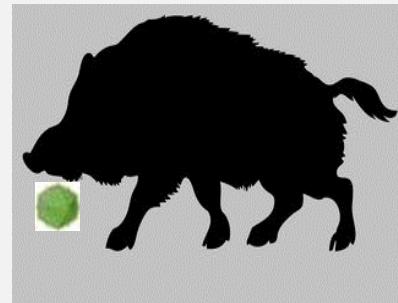
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# Experimental infection ASF



D0= Infection 100 HAU/ml orally ASF Virus Armenia/08



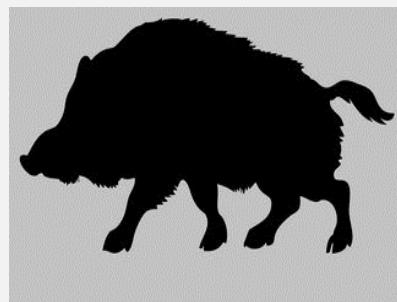
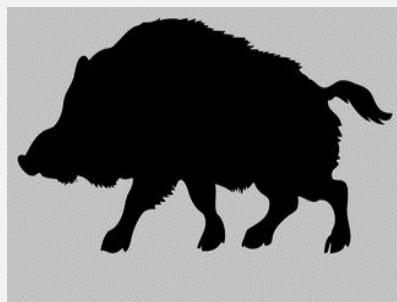
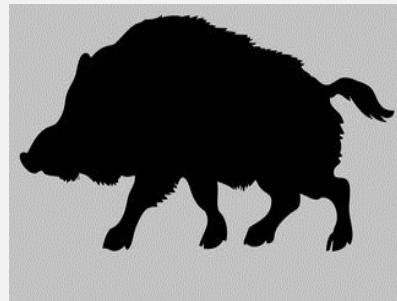
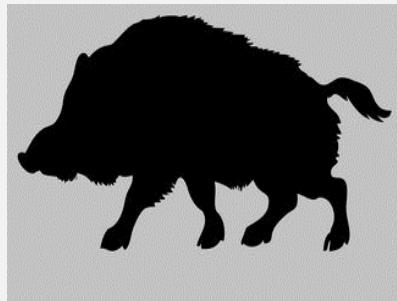
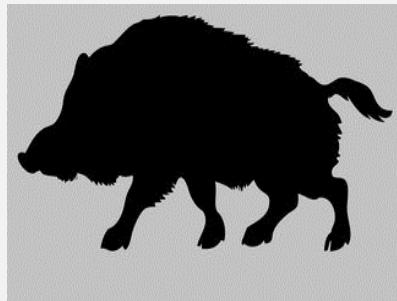
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# Experimental infection ASF



D12



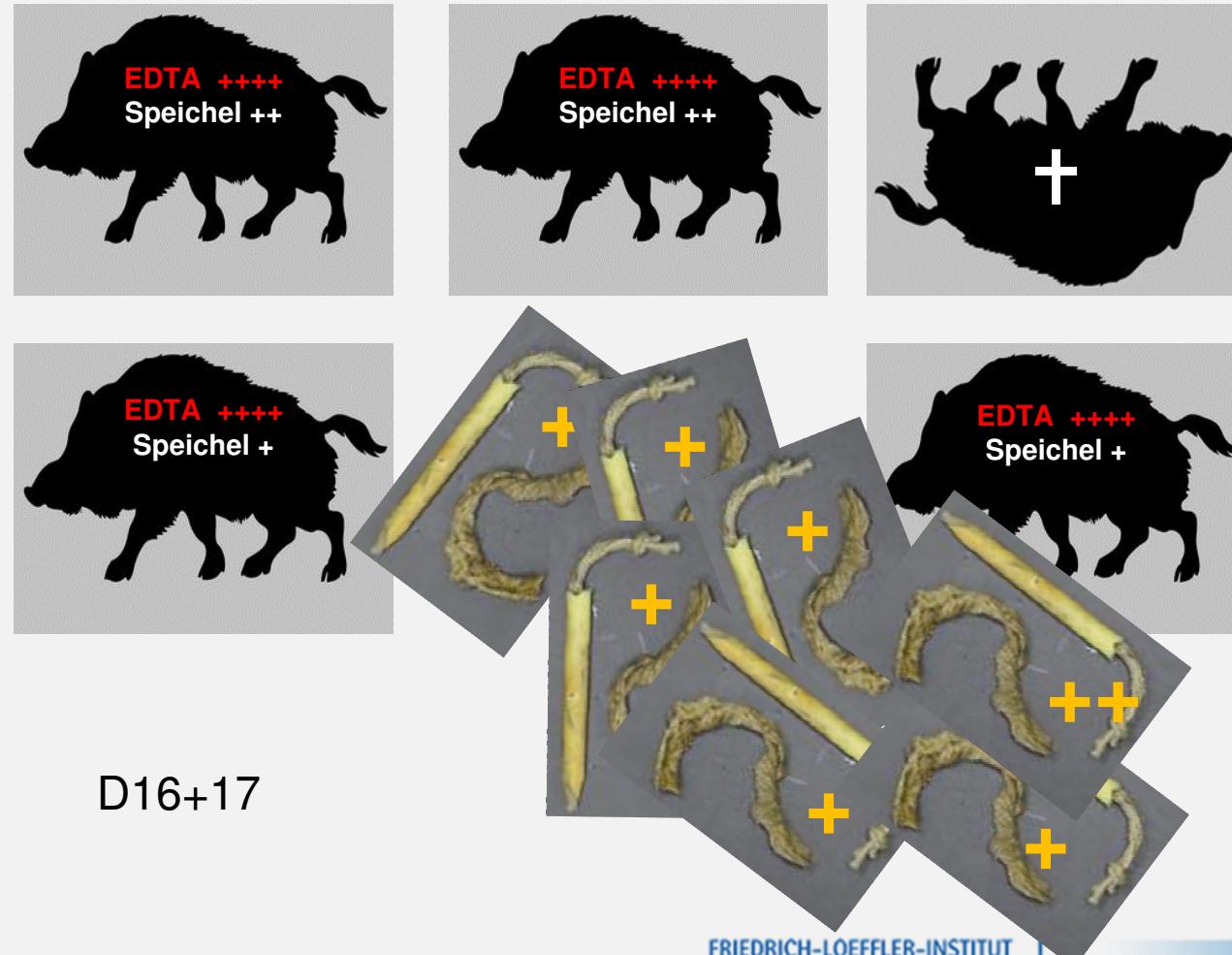
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# Experimental infection ASF



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# Experimental infection ASF



D18



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# Experimental infection CSF and FMD

Veterinary Microbiology 170 (2014) 425–429

Contents lists available at ScienceDirect

**Veterinary Microbiology**

journal homepage: [www.elsevier.com/locate/vetmic](http://www.elsevier.com/locate/vetmic)

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Short Communication

Novel rope-based sampling of classical swine fever shedding in a group of wild boar showing low contagiousity upon experimental infection with a classical swine fever field strain of genotype 2.3

Susan Mouchantat\*, Anja Globig, Wolfgang Böhle, Anja Petrov, Heinz-Günther Strebler, Thomas C. Mettenleiter, Klaus Depner

Friedrich-Loeffler-Institut, Federal Research Institute for Animal Health, Südufer 10, D-17483 Greifswald-Insel Riems, Germany



Wolfgang Böhle

Veterinary Microbiology 172 (2014) 329–333

Contents lists available at ScienceDirect

**Veterinary Microbiology**

journal homepage: [www.elsevier.com/locate/vetmic](http://www.elsevier.com/locate/vetmic)

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Short Communication

Proof of principle: Non-invasive sampling for early detection of foot-and-mouth disease virus infection in wild boar using a rope-in-a-bait sampling technique

Susan Mouchantat\*, Bernd Haas, Wolfgang Böhle, Anja Globig, Elke Lange, Thomas C. Mettenleiter, Klaus Depner

Friedrich-Loeffler-Institut, Federal Research Institute for Animal Health, Südufer 10, D-17483 Greifswald-Insel Riems, Germany

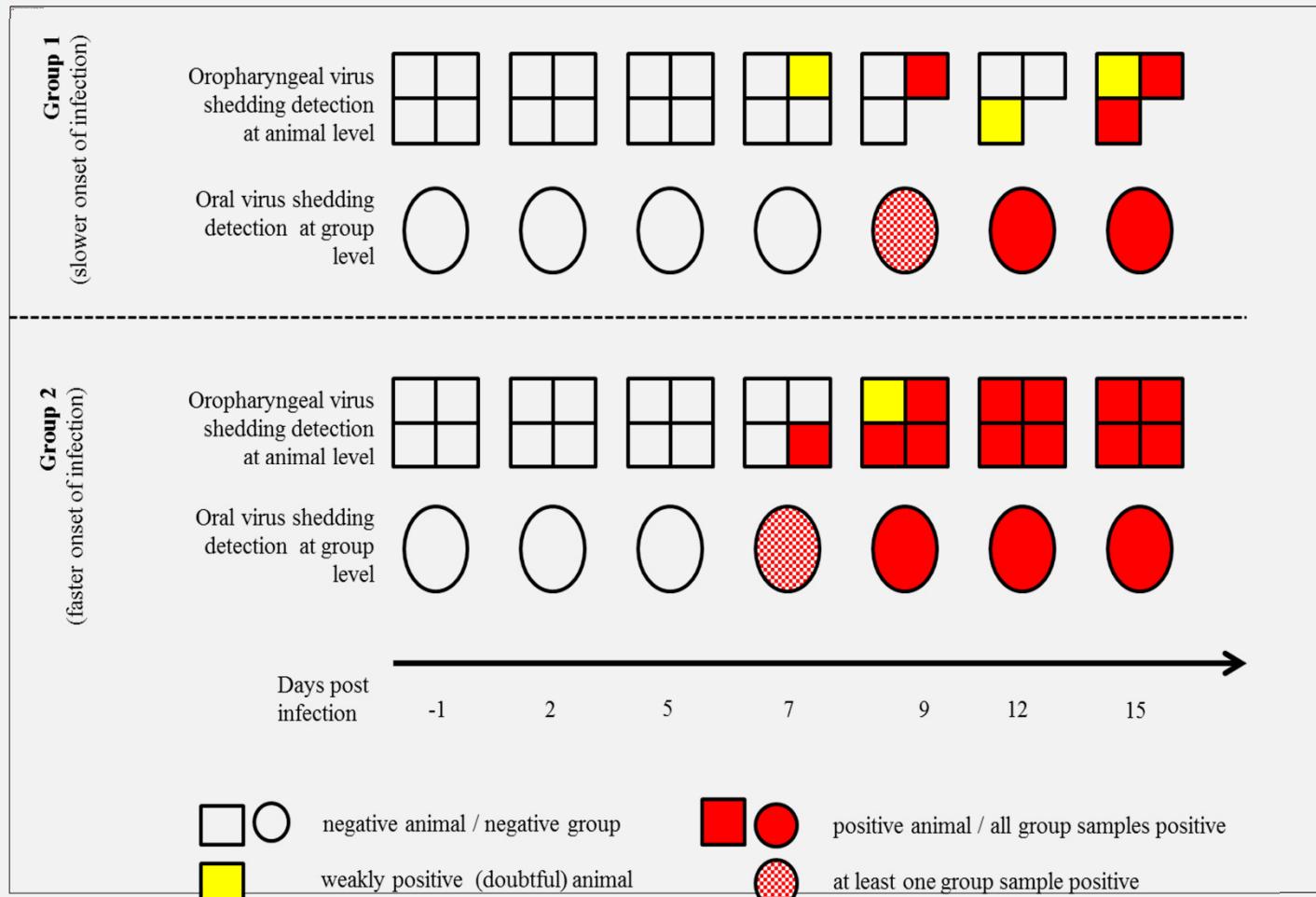


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# CSF experimental infection in domestic pigs



## In the field - non infected area

- Distribution of pSWABs at two locations
  - Recovery rate depending on day of collection:
    - Up to 100% when collection following day
    - decreasing recovery rate with every day (<50% after 4 days)
  - Ropes sometimes on the paths of wild boar far from distribution site: needs systematic search



# Who is chewing?



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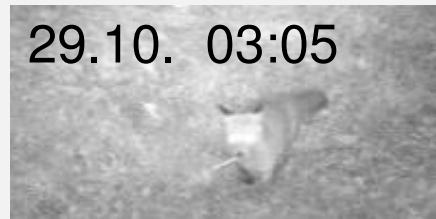
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29.10. 18:19



29.10. 03:05



30.10. 01:51

30.10. 2:57



31.10. 01:54



01.11. 2:28



05.11. 19:44

# They all love it



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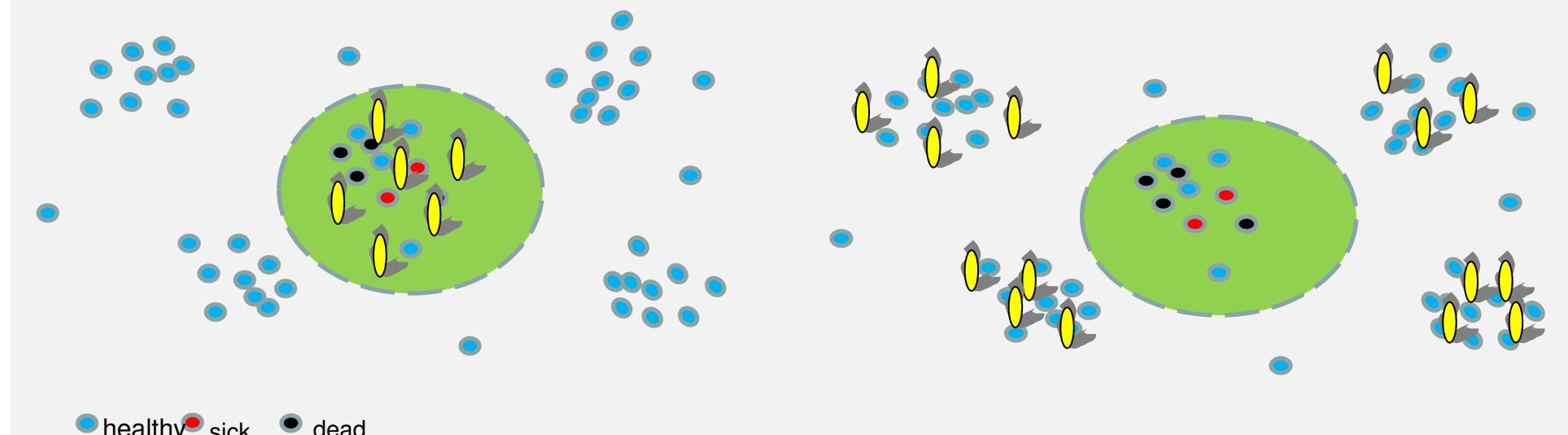
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# Development of a Triplex qPCR for simultaneous detection of swine-specific and ASF genome combined with internal control

Primer design for *Sus scrofa* specific target gene regions of CEACAM18 and mtCytB with ActB as control (Mouchantat, Forth, Kammerer et al., in preparation)

# In the field - ASF-infected area

- Collaboration with Estonia and Latvia
- Project ongoing: Distribution of pSWABs in Estonia, collection and investigation in the laboratory
- Applicable where/when hunting is not possible
- Suitable for ASF-antibody detection? (In process)



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# Summary

## Material

- pSWAB well accepted by wild boar -> collective sample of a wild boar herd
- The rope is robust, has a high capacity to absorb saliva, is easily produced, cost-effective and logically simple to integrate into wildlife management practices
- pSWAB is virus stable and is generally suitable as a rope-in-a-bait

## Suitability for virus detection

- Four experimental infections of wild boar with ASF (2), CSF and FMD proved the suitability of the pSWABs *in vivo*
  - Detection on herd basis as long as there is virus shedding by saliva even when shedding period is very short, viral load in saliva is low and animals do not exhibit any clinical signs.

## Suitability in the field

- pSWAB is chewed on by wild boars at feeding or wellness places (disadvantage: also other animals take them (away))
- In infected areas: first results from Estonia
- Can be useful in areas with no or prohibited hunting

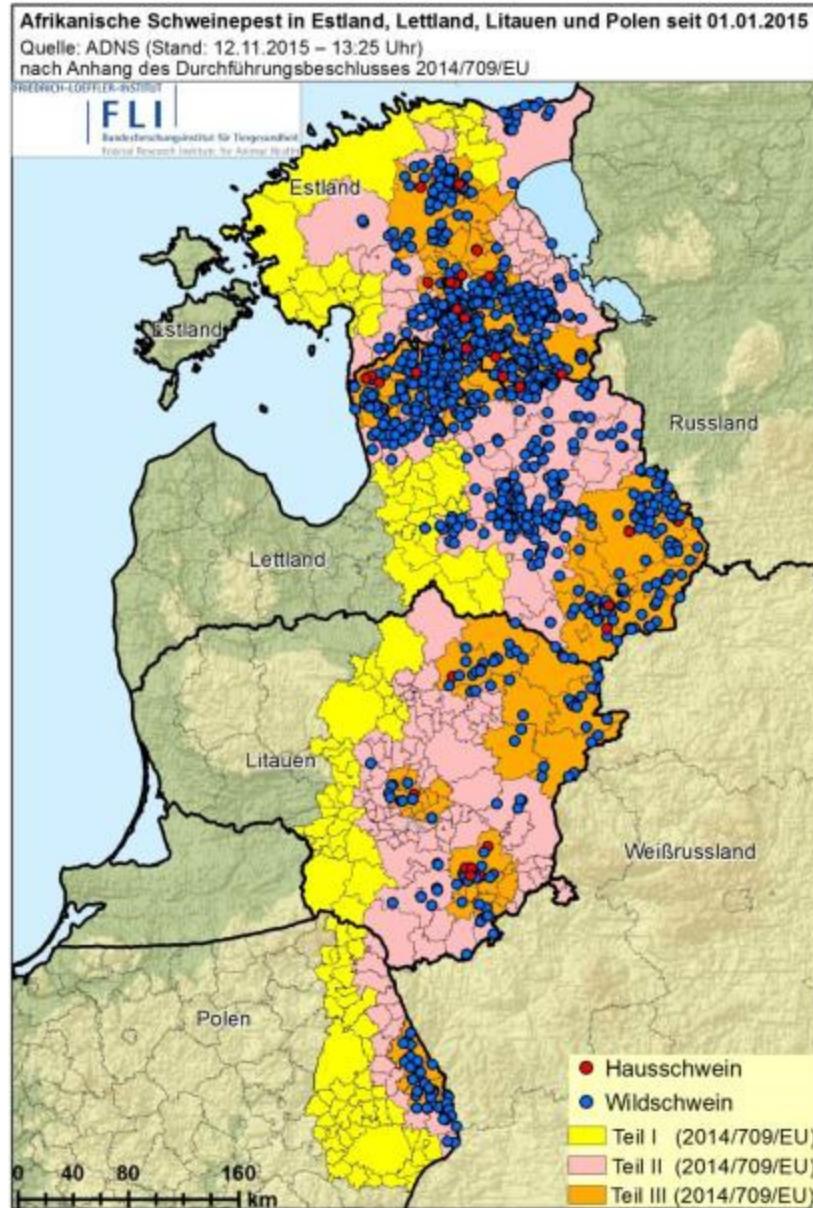


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## Hypotheses (2014):

1. ASF will fade out
2. ASF moves rapidly westwards

One year later.....

Both hypotheses proved to be more or less wrong

- No fading out
- No rapid westward movement

=> Established in the region

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# What we know

Lethality is high (>90%)

*(number of deaths / number of infected x 100)*

Mortality is low (<5%)

*(number of deaths / population x 100)*

Prevalence low (<5%)

*(number of infected / population x 100)*

**Contagiosity is low**

*(slow spreading.... Takes time)*

-> **direct contact** with infected material (blood/meat) or sick/dead animals

A low dose may not lead to an infection of healthy wild boar



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Klaus Depner

## ASF VIRUS IS VERY STABLE

- 140 days in Iberian and Serrano hams
- 399 days in Parma ham
- 112 days in Iberian pork loins.
- 18 months in pig blood at 4°C
- 11 days in faeces at 20°C
- Possibly long time in carcasses (dead animals) which decompose

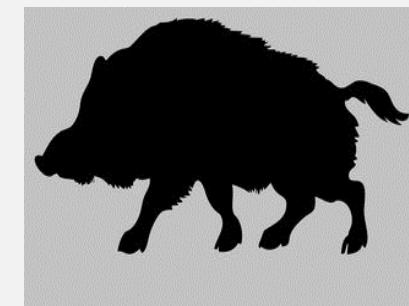
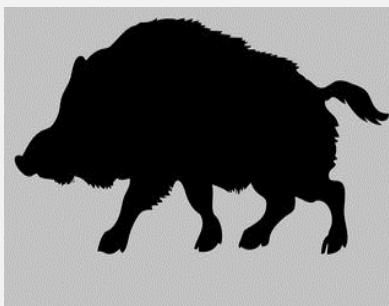
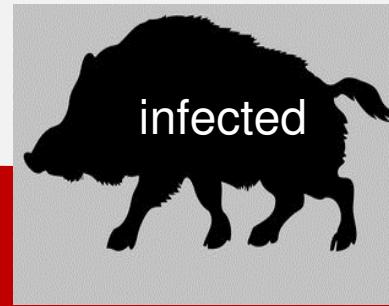
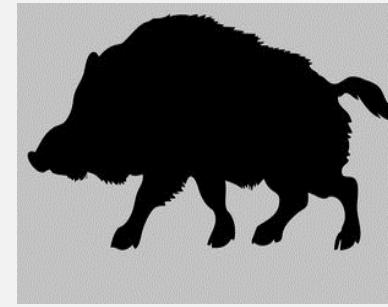
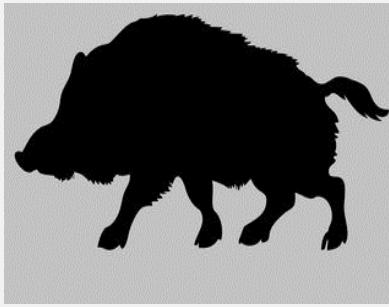


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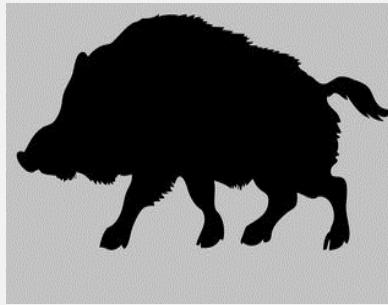
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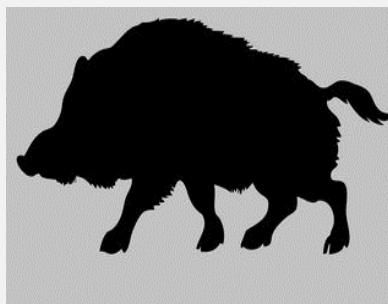
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- Do wild boar get into contact with dead wild boar and after how long time does this happen?
- Contact randomly or targeted?
- Which animals get into contact (age group)?



+ 6/7 days

- What kind of contact (nibbling, tearing, eating)?
- Real cannibalism between wild boar?
- How long is a carcass interesting for wild boar? Do they eat larvae?



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# Carcass experiment in the field



Wild boar, 100 kg, 2 years old, male, shot 27 October 2015  
and transferred in forest at a wellness and feeding place of wild boar



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# Fixing a wild life camera



# Week 1



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## Week 2: Carcass entirely intact



pSWABs distributed to attract wild boar



No direct contact, only taking pSWABs



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# Week 3: Carcass almost entirely intact



M

18/11/2015 01:30:57



Buzzard picking on nose



Little plants are growing  
Larvae all over

# Three weeks later (16 November 2015): First close contact



SEISSIGER Wildkamera

M

16/11/2015 22:46:19

010°C P0



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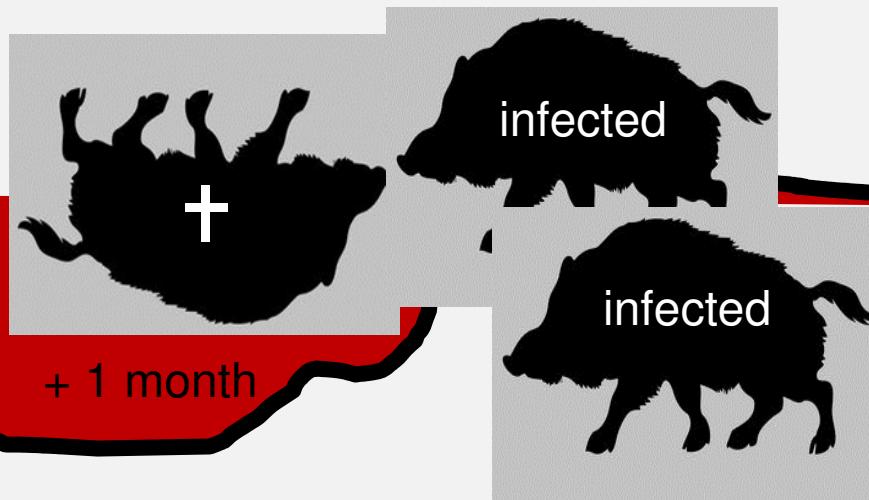
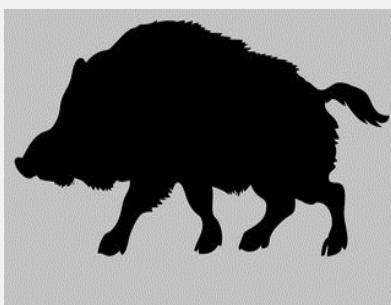
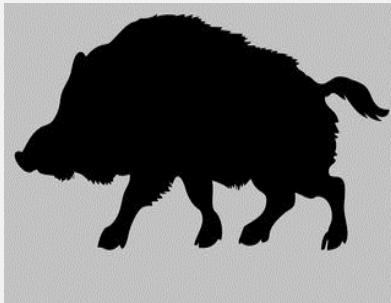
# Deer carcass, same time, other location



KeepGuard

10-28-2015 04:17:37

© Bent Knoll

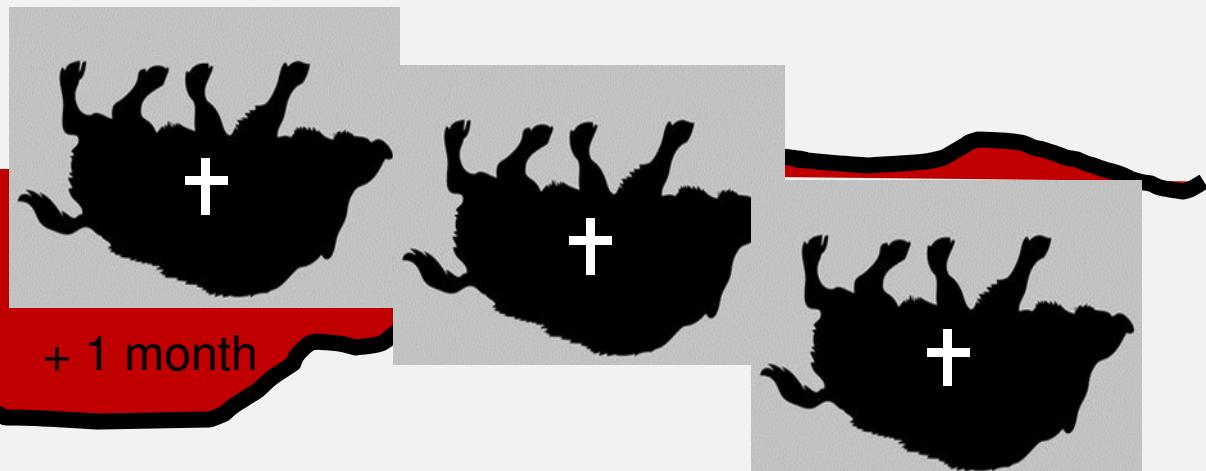
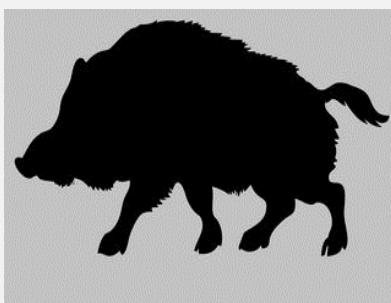
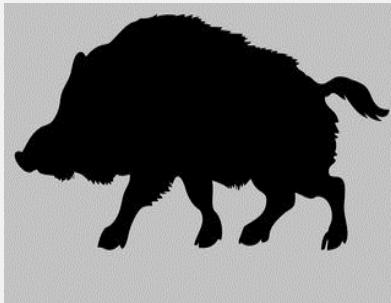


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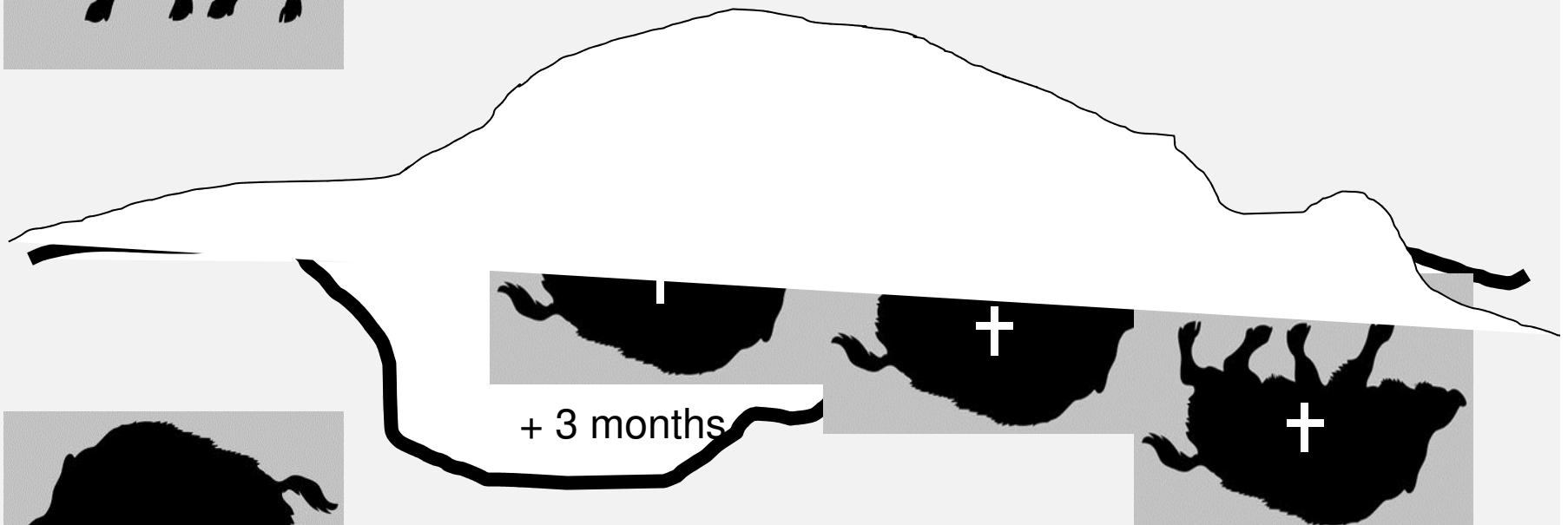
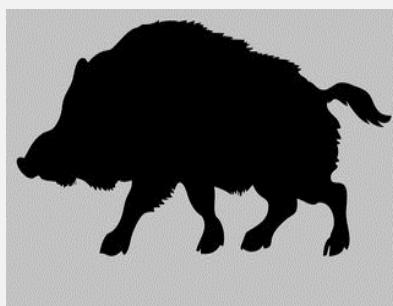
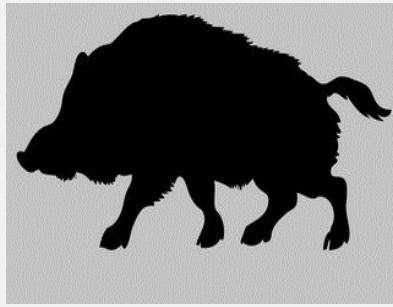


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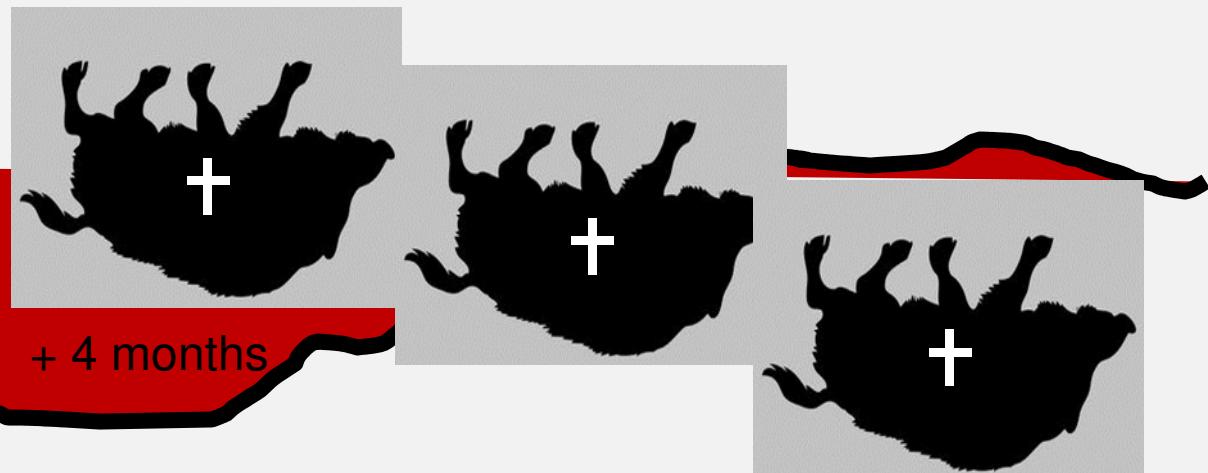
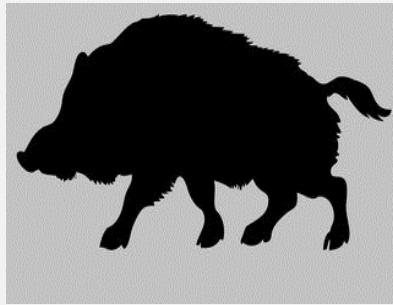
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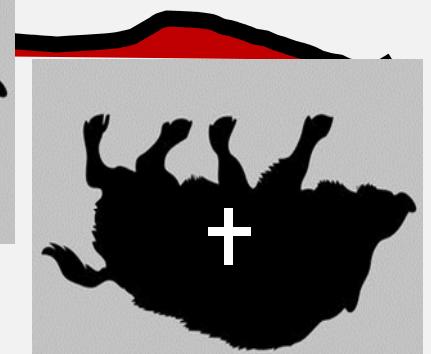
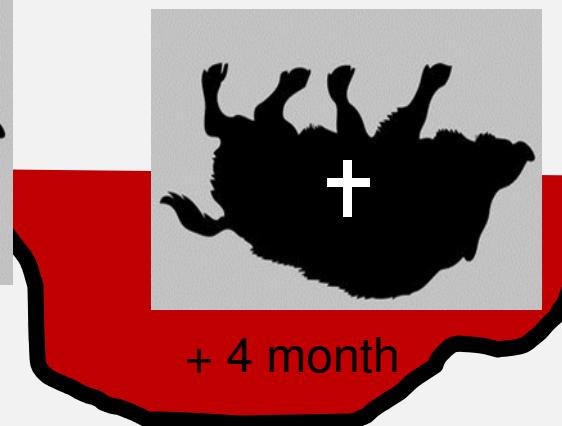
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Hypothetically: within 4 months 4 animals got infected at 1 wild boar



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# Hypothesis

ASF is a carcass-borne disease with infected carcasses as the virus reservoir

- May explain the virus persistence in geographical areas and the low prevalence (5%)
- If so: safe disposal of carcasses may be the key to success for the eradication of ASF (using sniffer dogs?)
- Investigation of wild boar populations and their behaviour to dead wild boar of different ages at different places and to different seasons will provide more insights on the epidemiology of ASF in wild boar populations



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- Ronald Abraham



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Thanks for your attention!



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