

Swine Influenza in Europe

Epidemiology – Diagnostics – Prevention

Agenda

1. Epidemiology
2. Swine Influenza on Farm
3. Diagnostics
4. Prevention

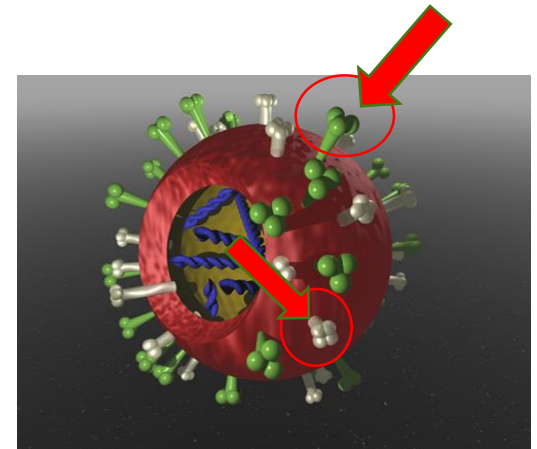
Epidemiology



IDT Biologika

Influenza Virus

- Family: *Orthomyxoviridae*
- Enveloped virus with segmented genome
- 8 segments code for 11 proteins
- Classification by type: matrix (M) and nucleoprotein (NP)
 - A, B, C or D
- Subtypes defined by hemagglutinin (HA or H) and neuraminidase (NA or N) > currently 18H and 11N
- New subtypes can develop by
 - Shift
 - Drift
 - Reassortment



Morphology, Source: Fields Virology

Genetic variability

Antigenic drift

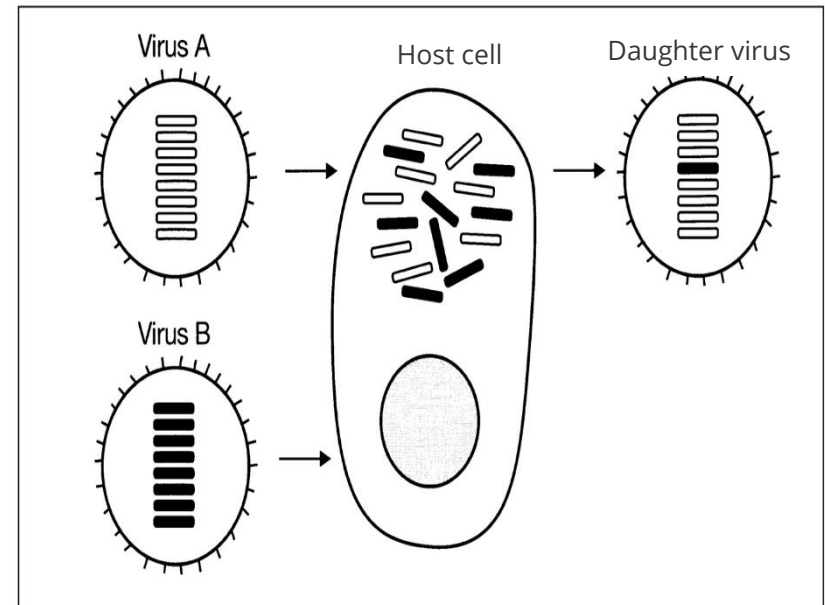
point mutations = variation within a subtype (aa)

variations that accumulate over time

Antigenic shift

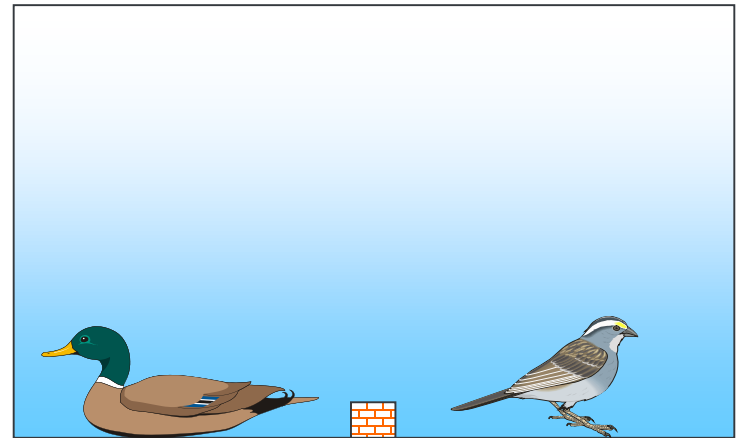
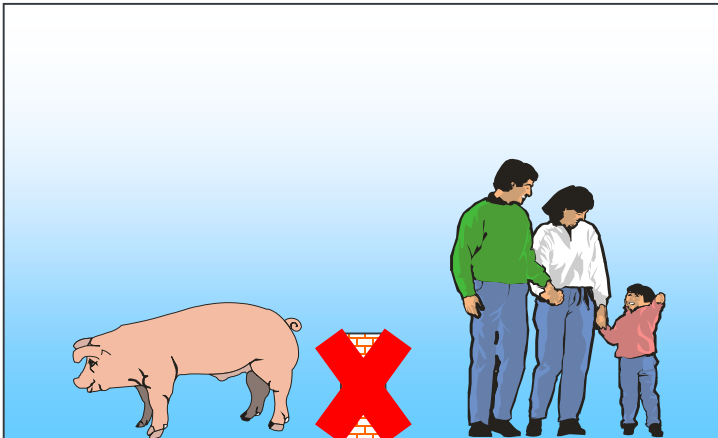
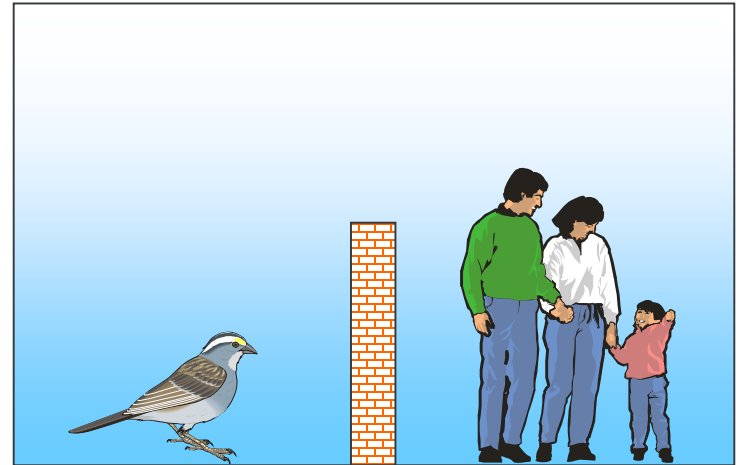
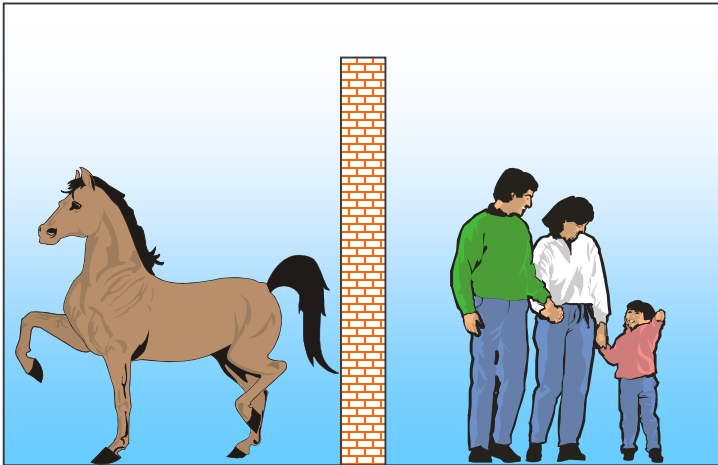
Reassortment

Exchange of gene segments

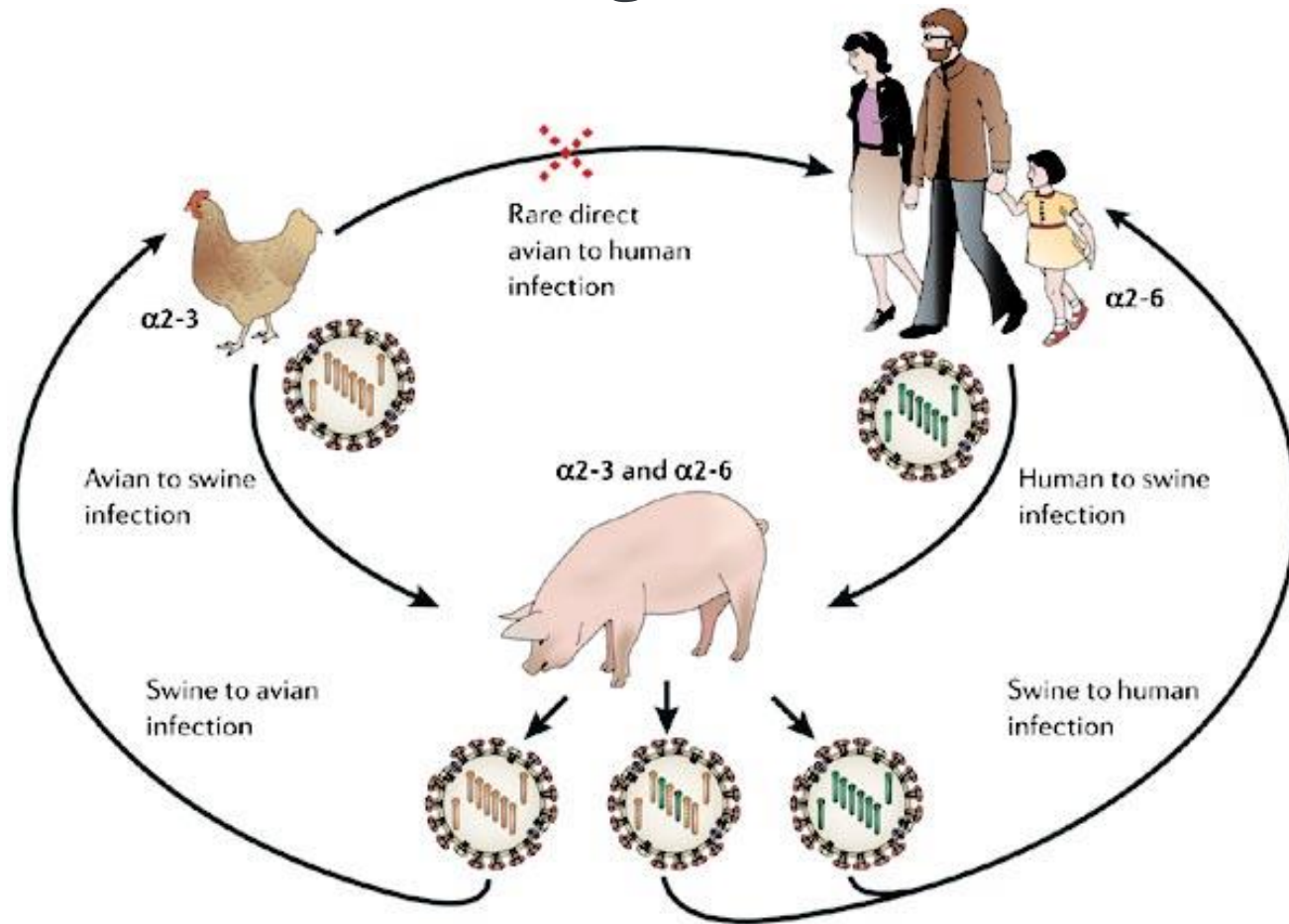


Shift - Reassortment, Source: R. Rott, Gießen

Influenza – species barrier

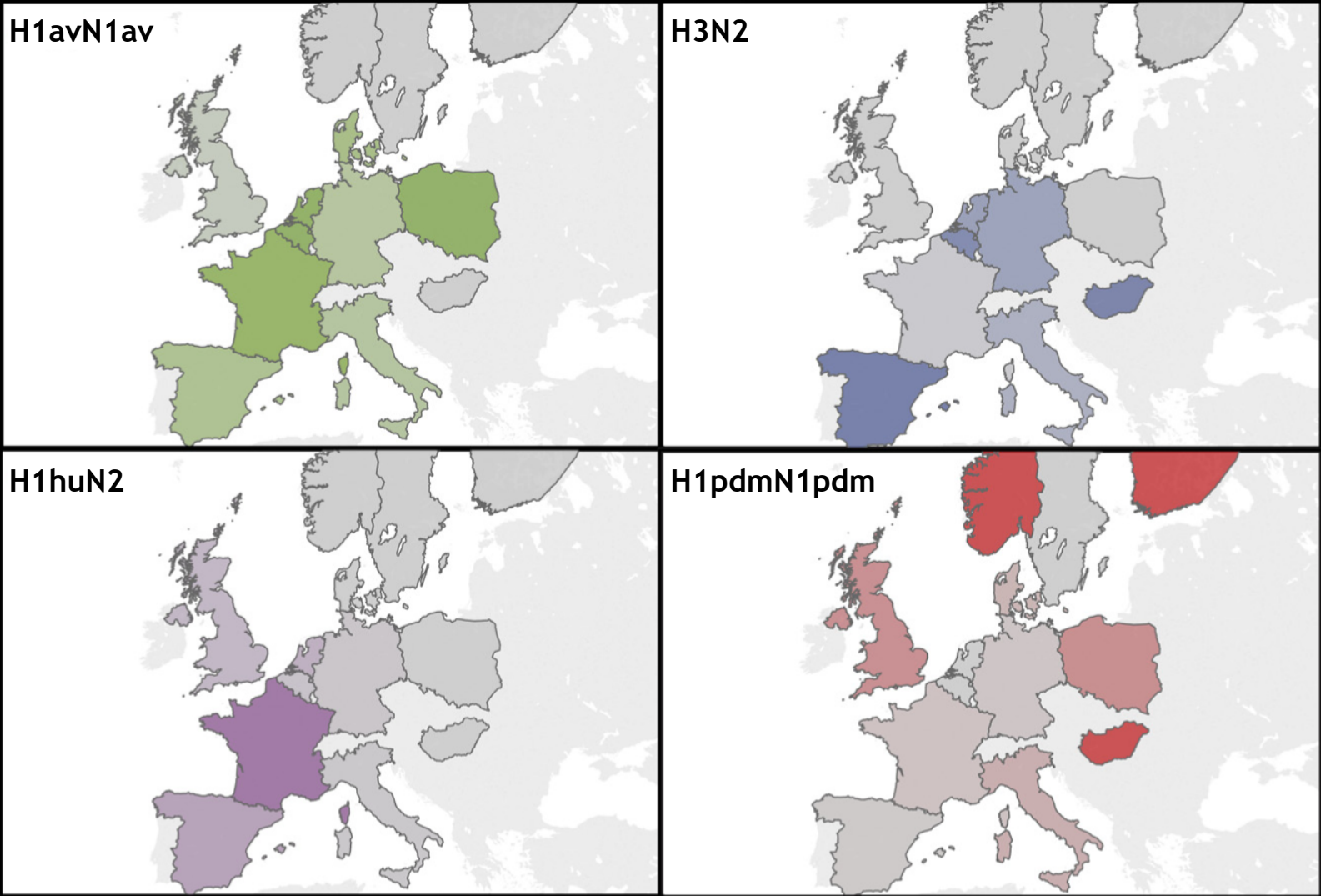


The swine as a mixing vessel



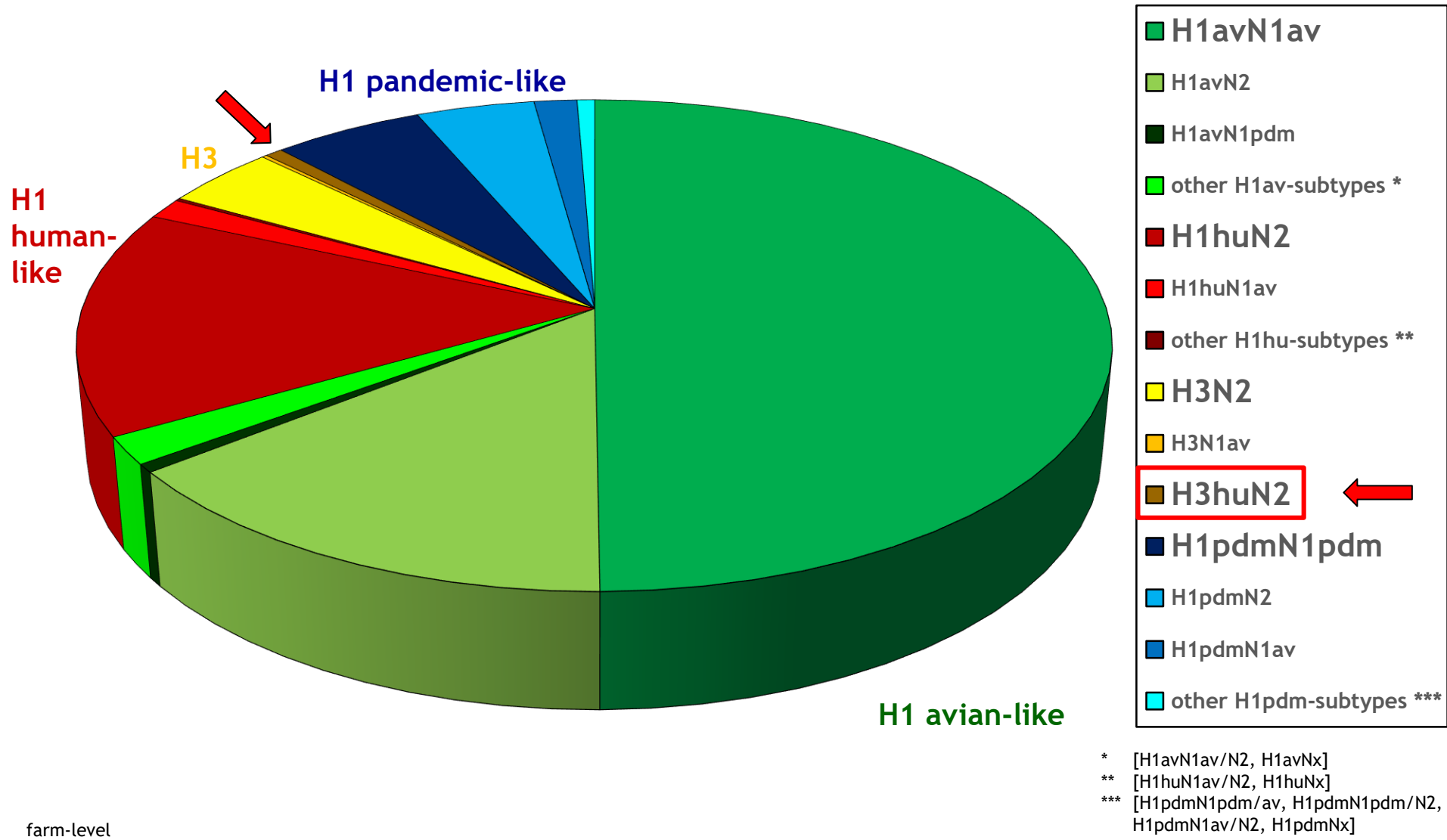
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Nature Reviews | Microbiology

Lineage-specific geographical restrictions of SIV in Europe before 2015



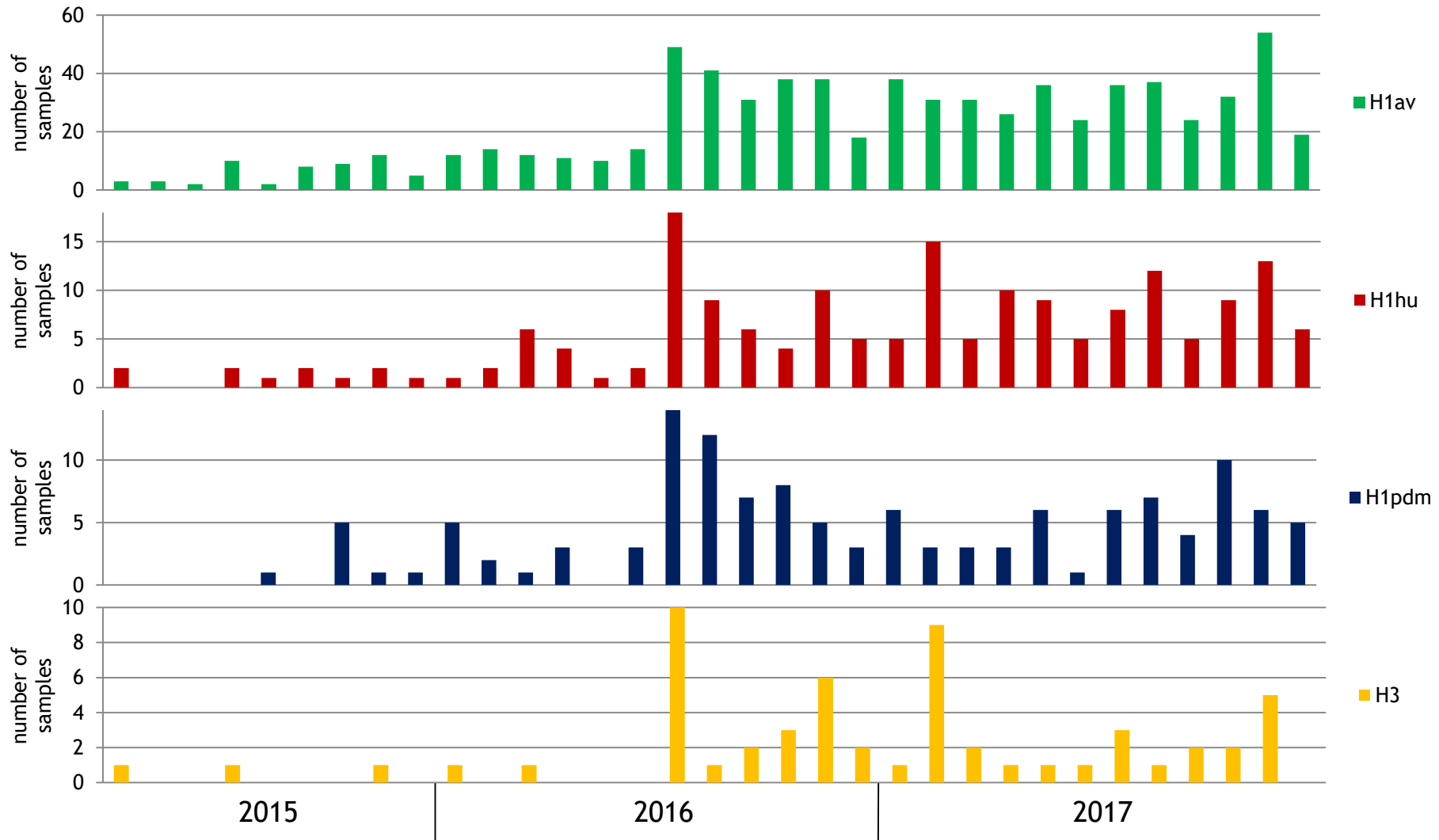
Watson et al. (2015) (modified)

IAV-subtyping





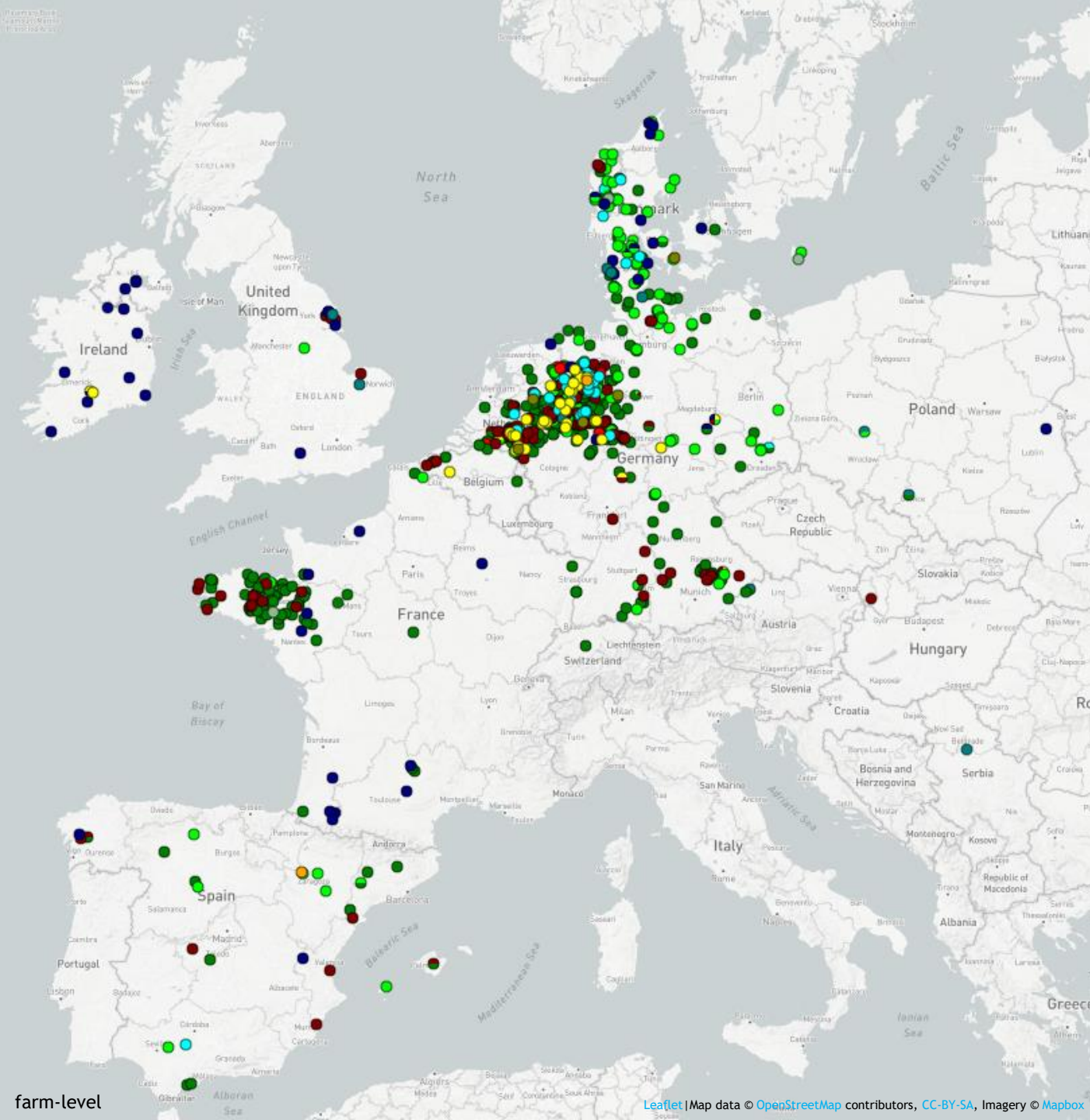
Distribution of HA subtypes per month



farm-level

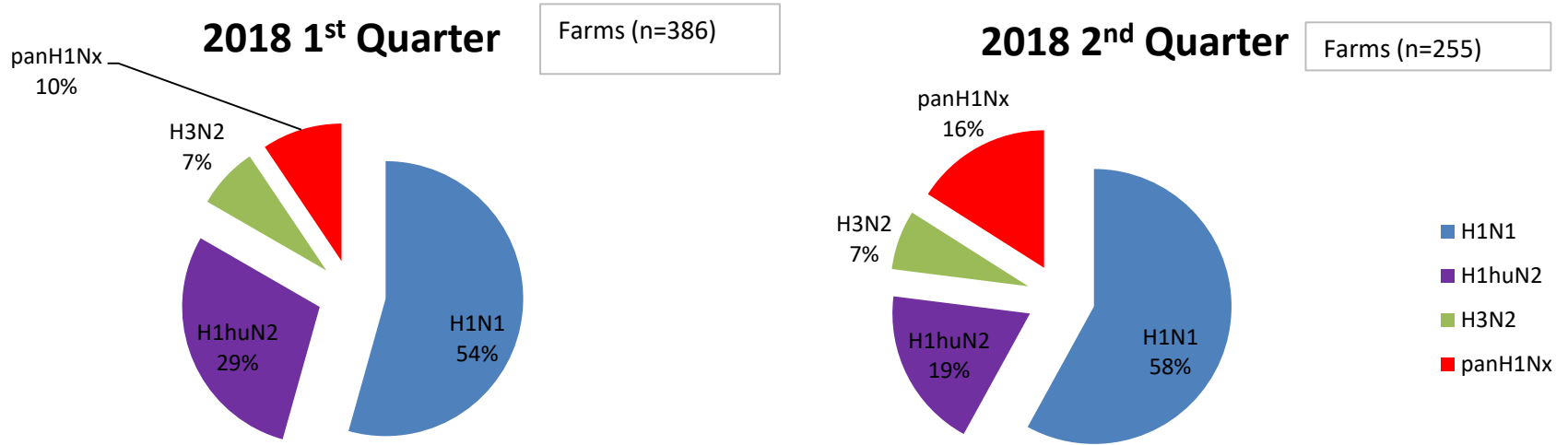
Geographic restriction of IAV subtypes

- H1avN1av
- H1avN1pdm
- H1avN2
- H1huN1av
- H1huN2
- H1pdmN1av
- H1pdmN1pdm
- H1pdmN2
- H3N1av
- H3N2
- H3huN2



ISIVR, Brighton 2018

Results from Germany (Farms 2018)



Analyses performed at: IVD GmbH Seelze

Swine Influenza on farm

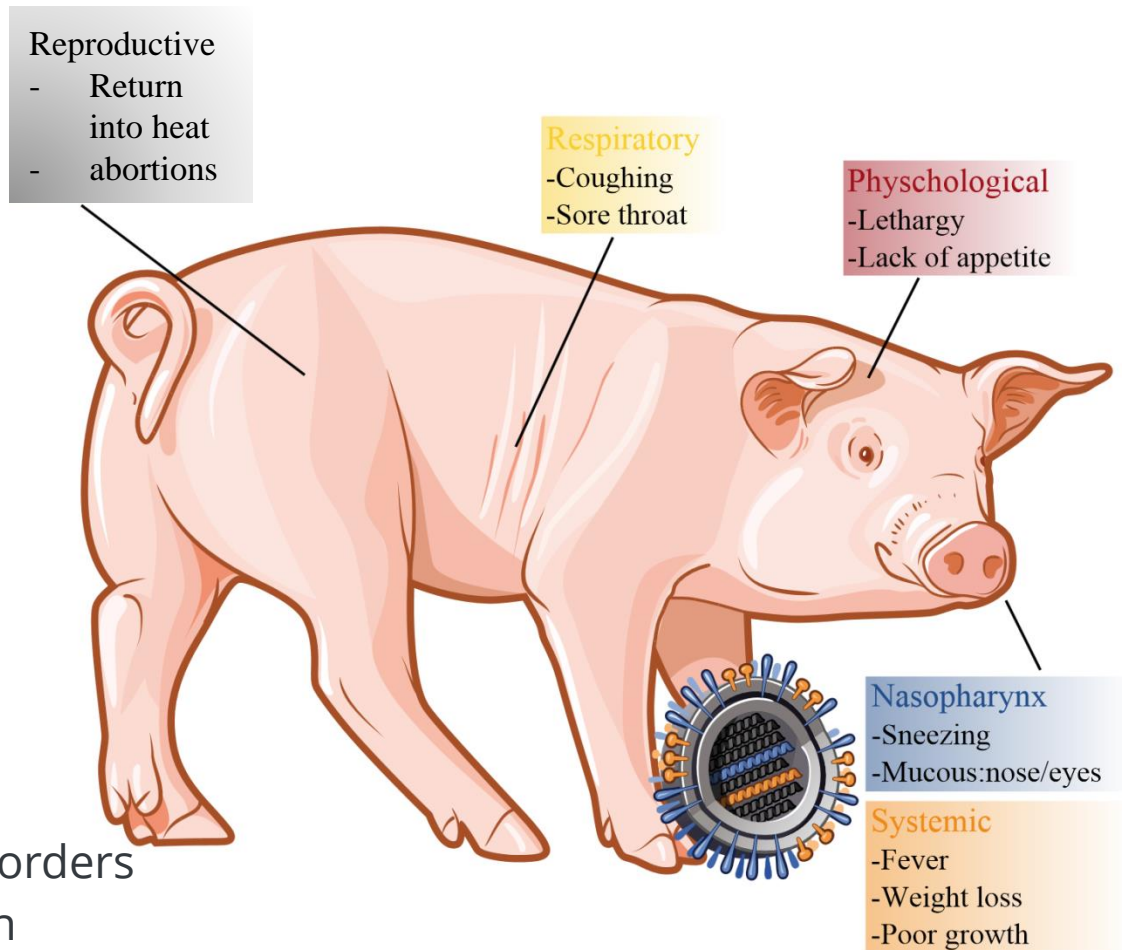
Influenza in pigs

Classical form

- All stages might be affected
- High fever
- Lethargy, coughing
- Periodic „every autumn“

Endemic („recurrent“) form

- No clear clinical signs
- Fever yes/no
- sporadic coughing
- Increase of reproductive disorders
- Severe respiratory disease in piglets (6-7 weeks of age)

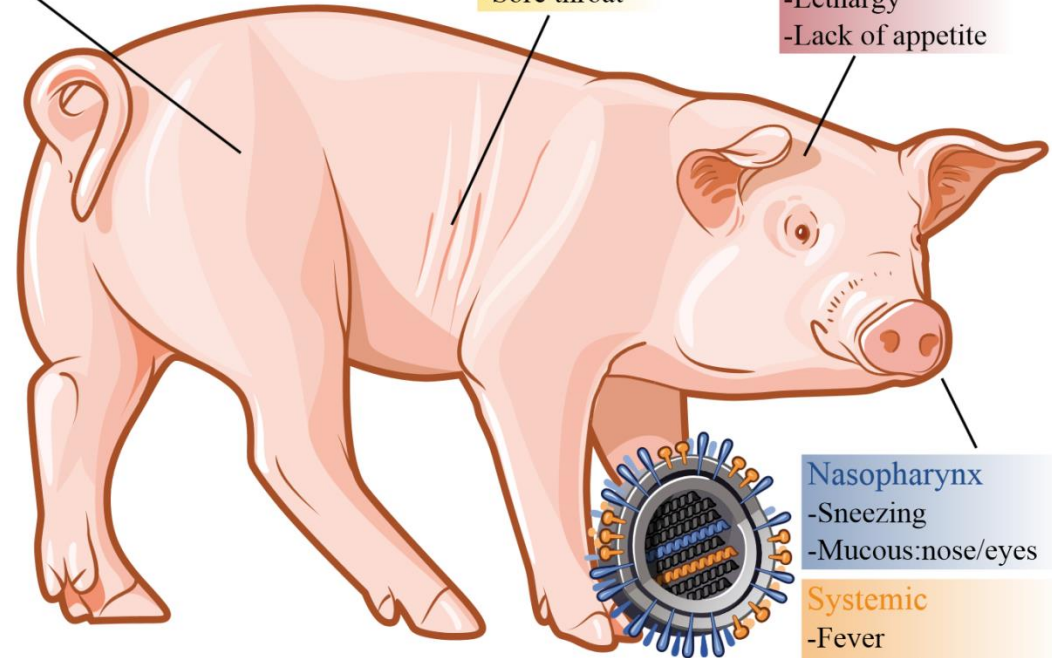


Influenza in pigs

Reproductive
- Return into heat
- abortions

Respiratory
-Coughing
-Sore throat

Physchological
-Lethargy
-Lack of appetite



Nasopharynx
-Sneezing
-Mucous:nose/eyes

Systemic
-Fever
-Weight loss
-Poor growth

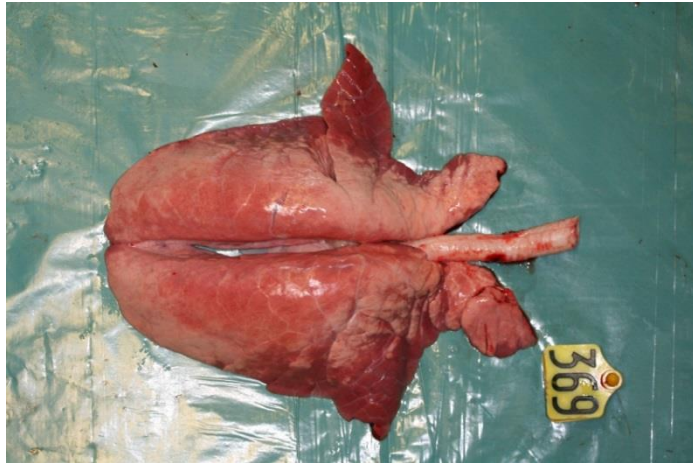
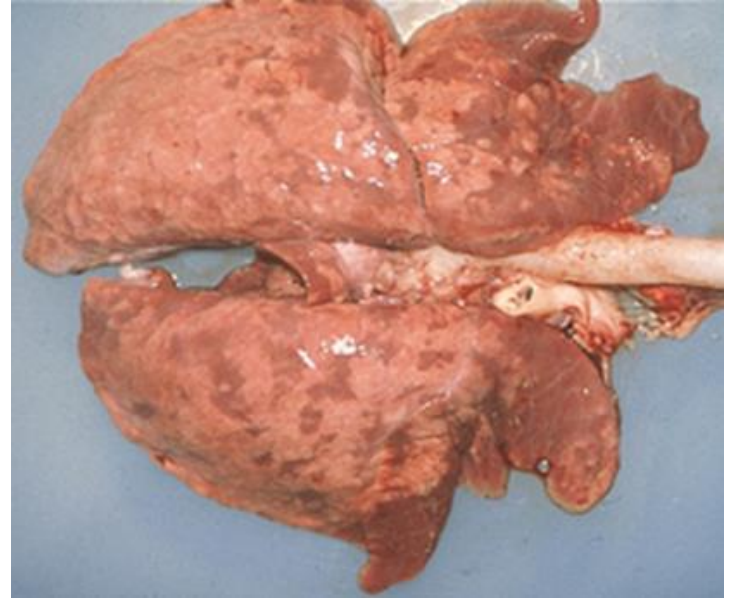
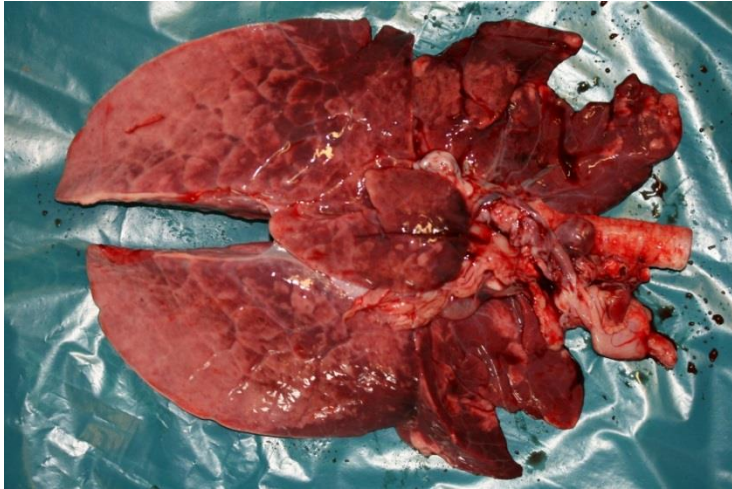
“Untypical” form

- “creeps through the farm”
- Reproductive disorders:
 - Increased abortion rate
 - Decreased live born
 - Increase of non-vital piglets
 - Apathetic piglets due to decrease in milk production of sows
- Re-occurring respiratory disease in all stages of production
- Often associated with pandemic strains

Clinical signs on farm



Pictures from necropsy

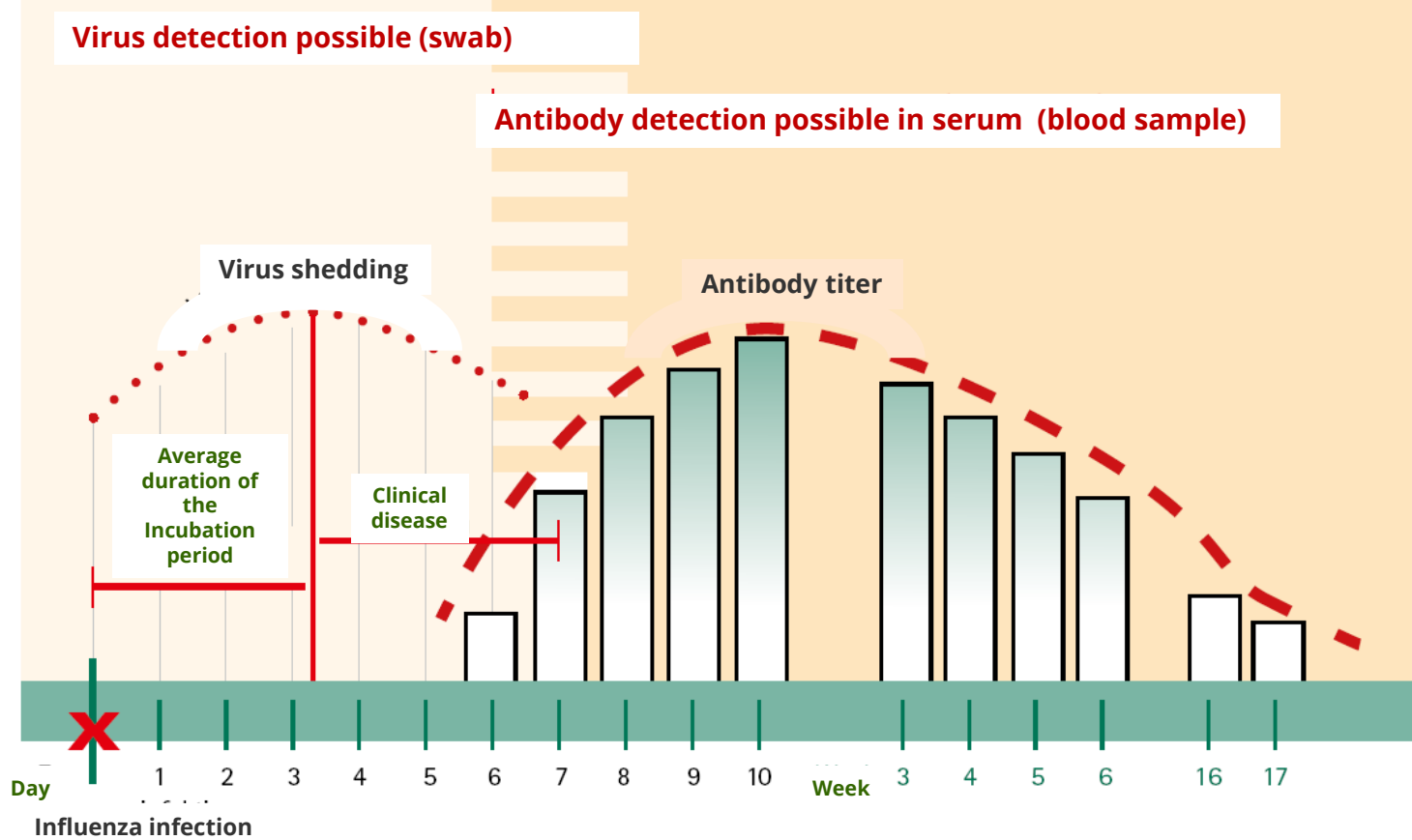


Diagnostics



IDT Biologika

The course of Influenza disease



The course of Influenza disease

Time	Activity of the virus	Clinical signs	Analyses	Recommended samples
0 h	Infection	---	---	---
24 h	Replication	Nasal discharge	Virus detection via PCR, qPCR (Virus isolation)	Nasal swabs
48 h	Shedding	Lethargy		BALF
1 – 5 days		Anorexia		Oral fluids
		Sneezing		(lung tissue)
		Coughing		
	Dyspnea			
6 – 8 days	Clearance	Recovery		
10 – 14 days	Seroconversion		Antibody detection via ELISA or HI	Serum

What does this mean?

Influenza can be difficult to diagnose because:

- The shedding of the virus is only for a short time
- The prevalence within the batch/affected groups may be low

Thus:

Different clinical signs
in **different** age groups
call for **different** sampling strategies

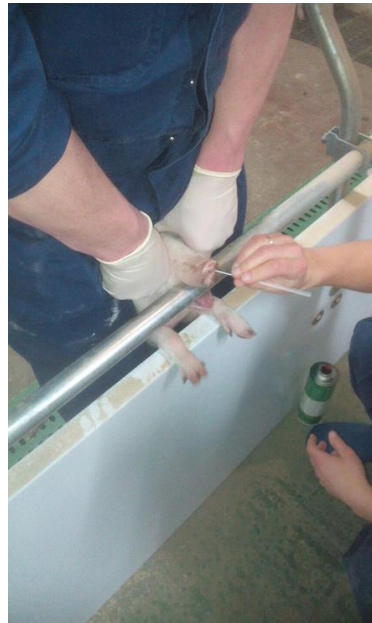
Detection methods

1. Virus detection

- I. **Nasal swabs**
- II. **Bronchial swabs**
- III. **BALF**
- IV. **Oral fluids**

2. Antibody detection

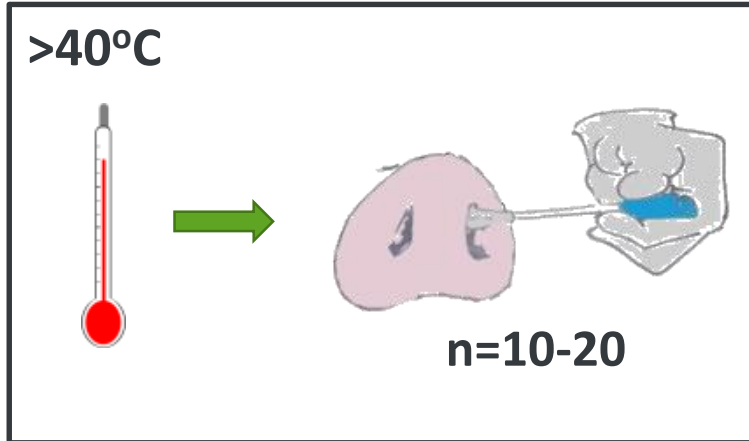
- I. **HI testing**
- II. **ELISA**



Clear clinical signs

Features:

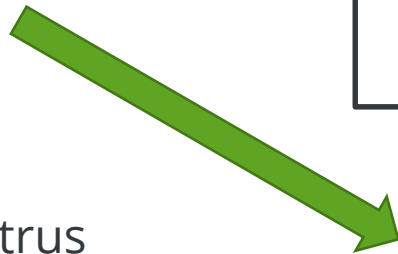
- High Incidence
- High Fever
- Dyspnea
- Mortality



or BALF

In sows

- Return to estrus
- Abortions



Bronchial swabs are better than lung tissue!

Clear clinical signs

Features:

- High Incidence
- High Fever
- Dyspnea
- Mortality

In sows

- Return to estrus
- Abortions

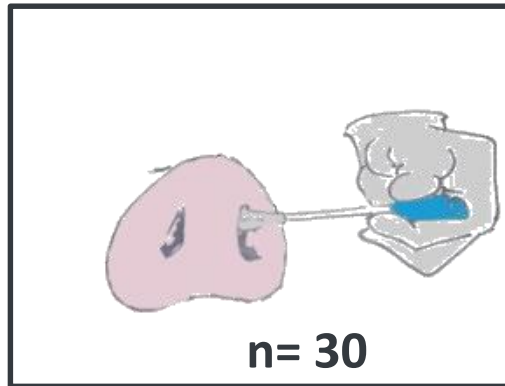
Serological Analyses:

- Difficult to interpret in (vaccinated) sows, 90% of farms und 75% of sows seropositive (DE)
- Can give a hint to the involvement of pandemic subtypes
- Useful in nursery and fattening, particularly when paired samples are taken (same animals 2-3 weeks later)
- Important: sample size is crucial!

No clear clinical signs:

Features:

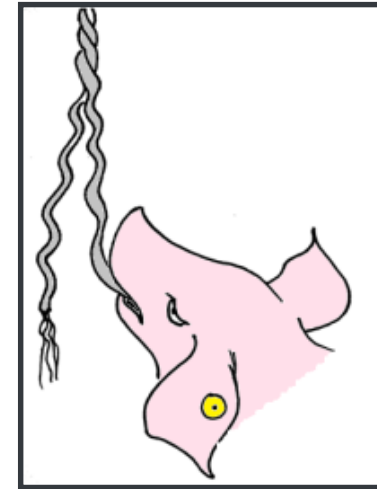
- Low Incidence of disease
- Low performance
- Sneezing, occasional coughing 2 – 4. week of life
- Increased feed conversion rate
- Increased number of secondary infections
- Increased mortality due to secondary infections
- Occasionally acute cases



Analytic method:

Virus detection via PCR/qPCR

Alternative:



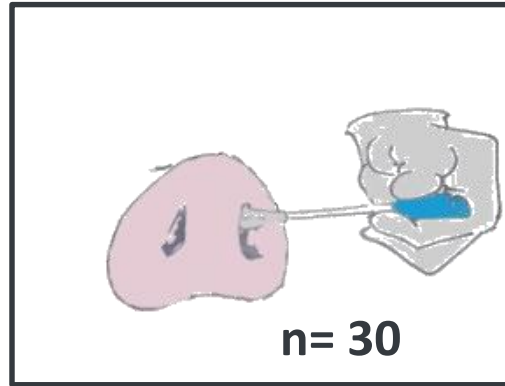
plus Serum profiles
n= 20-30



No clear clinical signs: suckling piglets

Features:

- Low Incidence of disease
- Sneezing
- Occasional coughing 2 - 4. week of life
- Occasional animals with fever



- If there are piglets with fever, sample these
- In litters where some of the piglets are coughing, sample the entire litter
- If clinical signs are completely unclear, sample litters of gilts

Analytic method:

Virus detection via PCR/qPCR

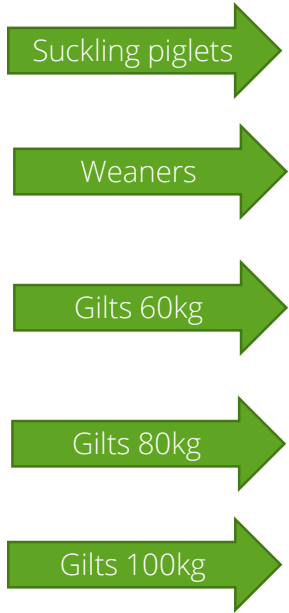


Results of diagnostics: what can you expect?

Nasal swabs



Nasal swab result



Unter-suchungs-nummer	Identifikation	SIV	SIV ct	Kommentar
18/10113-1	1 Pool aus 4 Tupfern in einem Gefäß SF	neg		
18/10113-2	2 Pool aus 5 Tupfern in einem Gefäß AF	POS	20	
18/10113-3	3 JS-Stall 1 Pool aus 3 Tupfern in einem Gefäß JS	POS	36	
18/10113-4	4 JS-Stall 2 Pool aus 3 Tupfern in einem Gefäß JS	(pos) ¹⁾	39	
18/10113-5	5 JS-Stall Pool aus 3 Tupfern in einem Gefäß JS	POS	36	

Unter-suchungs-nummer	Identifikation	HA-Typ	NA-Typ	Subtyp	Kommentar	Sonstiges
18/10113-2	2 Pool aus 5 Tupfern in einem Gefäß AF	H1pdm	N1all	Reassortante		



Antibody detection I

ELISA: Enzyme-Linked Immunoabsorbent Assay

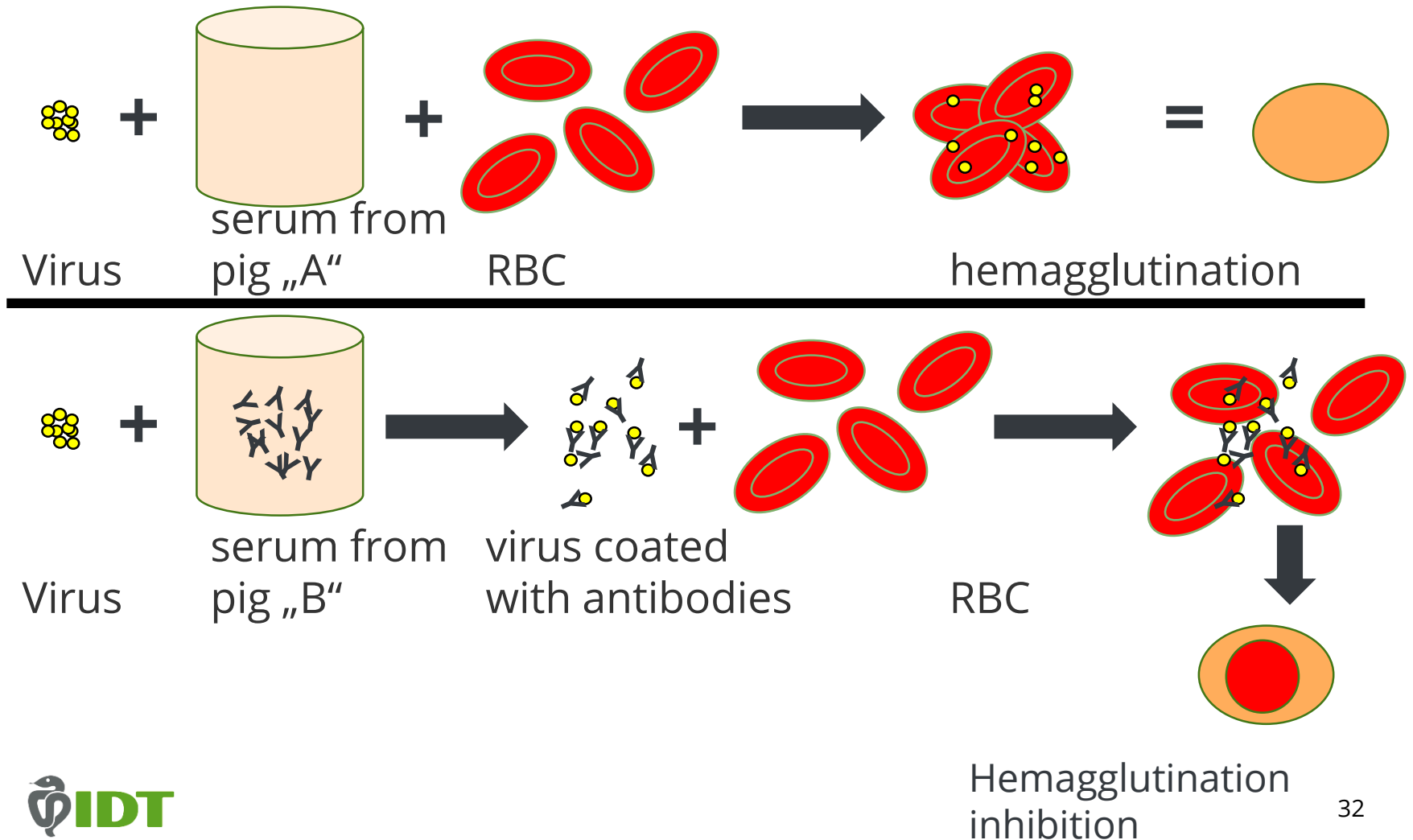
- A method for herd screening
- identifies species-independent influenza A
- Qualitative result only → positive or negative
- Interpretation of results: > 80% positive samples indicate active disease
- Reaction often seen “earlier” than in the Hemagglutination inhibition test

Antibody detection II

Hemagglutination inhibition test (HI):

- specific test for specific subtypes
- Options: detection of antibody-titers
- Interpretation of results:
 - Increase in titer from 10 days post infection
 - Experience in interpreting the results is needed
 - Cross-reactions possible
 - No differentiation between vaccinated or infected animals possible
 - Paired serum samples can help to monitor the situation

Principle HI test

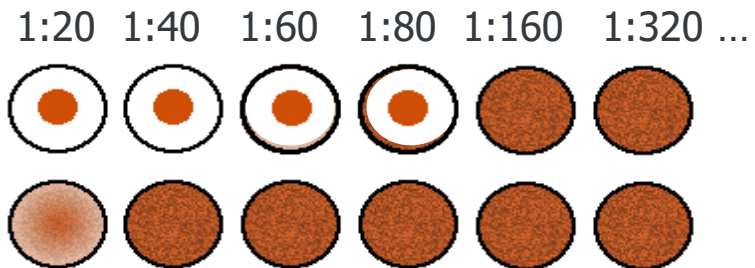


An example

H1N2 – infection unvaccinated animals

BU Animal Health VF- CV/HAH Influenza virus

no	Animal ID	Reciprocal titer			
		H1N1	H1N2	H3N2	H1N1 pan.
45	SV 125 -1	0	320	0	20
46	-2	0	320	0	0
47	-3	0	640	0	20
48	-4	0	320	0	0
49	-5	0	640	0	0
50	-6	0	320	20	20
51	-7	40	2560	0	20
52	-8	80	2560	0	40
53	-9	40	1280	0	20
54	SV125 -10	20	640	0	40



Results of diagnostics: what can you expect?

Serology

A typical result:

Untersuchungsnummer	Identifikation	avH1N1 Antigentyp 2	avH1N1 Antigentyp 1	huH1N2	huH3N2	panH1N1	panH1N2
17/18400-1	SL17A1201/1	≥2560	160	160	160	<20	20
17/18400-5	SL17A1201/5	160	40	<20	40	80	20
17/18400-6	SL17A1201/6	640	640	640	320	80	40
17/18400-7	SL17A1201/7	20	20	40	80	<20	20
17/18400-8	SL17A1201/8	20	80	80	160	<20	<20
17/18400-9	SL17A1201/9	40	40	160	160	<20	<20
17/18400-10	SL17A1201/10	80	160	320	320	40	40
17/18400-11	SL17A1201/11	20	160	160	160	<20	20
17/18400-12	SL17A1201/12	160	320	160	160	40	20
17/18400-13	SL17A1201/13	40	320	80	160	40	40
17/18400-14	SL17A1201/14	80	160	320	160	20	40
17/18400-15	SL17A1201/15	40	160	160	160	<20	80
17/18400-16	SL17A1201/16	80	160	80	320	<20	80
17/18400-17	SL17A1201/17	80	80	80	160	<20	80
17/18400-18	SL17A1201/18	80	80	160	160	40	80
17/18400-19	SL17A1201/19	40	80	80	80	<20	20
17/18400-20	SL17A1201/20	160	320	80	160	20	20
17/18400-21	SL17A1201/21	80	160	160	320	<20	80

Cross reactions can occur between the H-Antigens in the test. Serology in a vaccinated herd is almost impossible to interpret!

Summary: Diagnostics I

- The type of sample and the type of analytic method depends on the clinical signs and which age group is affected
- Sample size is crucial!
 - Always take at least 20 samples per farm
 - Always take at least 10 samples per affected group
- Nasal swabs are the sample of choice to detect the virus
 - Alternatives are: bronchial swabs, BALF or oral fluids

Summary: Diagnostics II

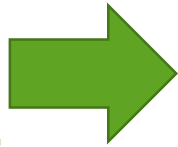
- Serology has its uses, but also its limitations:
 - Cross-reactions make interpretation difficult
 - Should not be used in vaccinated sows
 - In cases of the pandemic subtype, not the height of the titer is important, but how many animals have a titer
- Paired serum samples can offset some of the limitations
 - 2-3 weeks apart & from the same animals
 - Have the samples analyzed together in order to reduce inter-test problems

Case report

- Three piglet producers (150 sows, 800 sows, 300 sows) wean into 1 rearing unit (6kg – 30 kg)
- All sow herds are vaccinated with Respiporc FLU3
- In May 2015
 - ~ 3 weeks after weaning, the piglets started coughing
 - state laboratory detected Influenza A
 - Suspicion of pandemic Influenza; farm was re-sampled

Sampling results

Pool-No.	Date	Age	Material	Influenza A	Type
1	15.06.15	3 Wo weaned	NT-Pool	pos	H1N1
2	15.06.15	4 Wo weaned	NT-Pool	pos	H1N1
3	15.06.15	3 Wo weaned	NT-Pool	pos	H1N1
4	15.06.15	3 Wo weaned	NT-Pool	neg	neg
5	15.06.15	4 Wo weaned	NT-Pool	neg	neg
6	15.06.15	3 Wo weaned	NT-Pool	pos	H1N1
7	15.06.15	3 Wo weaned	NT-Pool	neg	neg
8	15.06.15	4 Wo weaned	NT-Pool	neg	neg
9	15.06.15	3 Wo weaned	NT-Pool	neg	neg
10	15.06.15	4 Wo weaned	NT-Pool	pos	H?N2
11	15.06.15	4 Wo weaned	NT-Pool	pos	panH1N2
12	15.06.15	3 Wo weaned	NT-Pool	neg	neg
13	15.06.15	4 Wo weaned	NT-Pool	neg	neg
14	15.06.15	4 Wo weaned	NT-Pool	neg	neg
15	15.06.15	3 Wo weaned	NT-Pool	neg	neg



Increased sample size necessary to detect pandemic subtypes

Prevention



IDT Biologika

Respiporc FLU3

Specific Product Characteristics (SPC):

Active immunization of pigs (*i.m.*)

- from the age of 56. days of life onwards
- Subtypes: H1N1, H1N2 and H3N2
- to reduce clinical signs
- reduce the viral load of the lungs
- Also for use in pregnant sows!



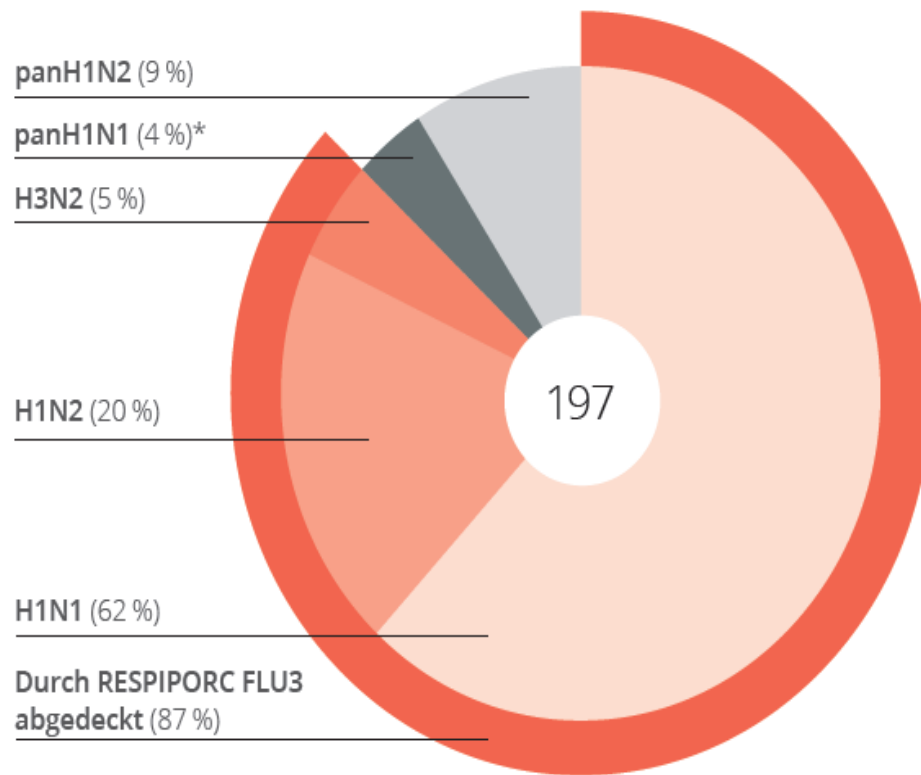
Dose: 2 ml *i.m.*

Onset of immunity: 7 days after basic immunization

Duration of immunity: 4-6 months (depending on age at the first vaccination)



Results of the analyses of Influenza in Europe



Virus analyses of
the FLI in 2016 :

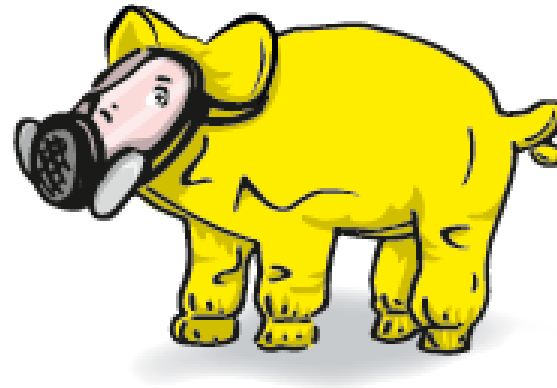


87 % of the isolates
are covered by
RespiPorc FLU 3

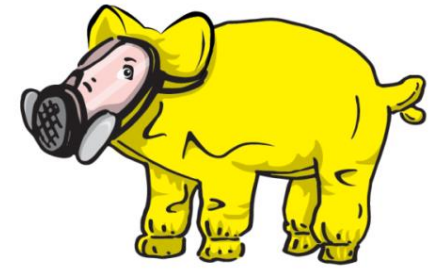


Do you really want
to influenza me?

Respiportc FLUpan H1N1



Respiporc FLUpan H1N1



Active Substance

Inactivated influenza A virus/human
Strain: A/Jena/VI5257/2009(H1N1)pdm09 16-64 HU*

*HU – haemagglutinating units in the vaccine

Adjuvant

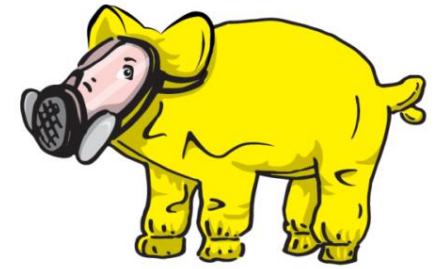
Carbomer 971 P NF 2 mg

Excipient

Thiomersal 0.1 mg



Respiporc FLUpan H1N1

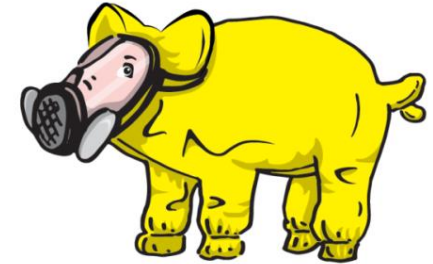


- Vaccination schedule -



- Vaccination of pigs possible from the age of 8 weeks onwards -

Case report



Farrow to finish farm with 120 sows

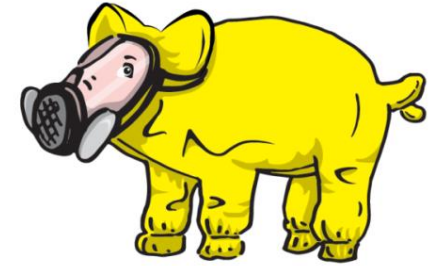
- two-week batch production with 3 week suckling period
- Vaccination of sows: Parvovirus, PRRSV, Respiporc FLU3,
- Vaccination of piglets : *Mycoplasma hyopneumoniae*, PCV2, PRRSV

Problem on- farm

In the rearing unit and in fattening:

- Wave-like occurrence of coughing, dyspnea, fever (40-41°C)
- Increased number of runts, groups became inhomogeneous and there was a reduced feed intake
- Antimicrobials did not help even though the overall use of antimicrobials increased considerably

Case report



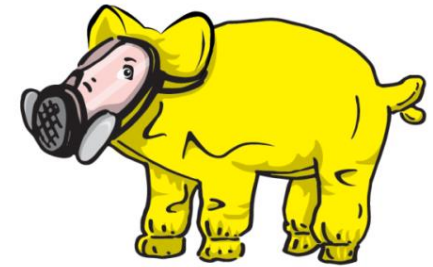
Diagnostics of 12 week old piglets

Animal ID	reciprocal titre			
	H1N1	H1N2	H3N2	H1N1 pan.
1	∅	∅	∅	40
2	∅	∅	∅	20
3	∅	∅	∅	20
4	∅	∅	∅	20
5	∅	∅	∅	80
6	∅	∅	∅	40
7	∅	20	∅	20
8	∅	∅	∅	80
9	∅	∅	∅	40
10	∅	∅	∅	40



Begin of vaccination with Respiporc FLU panH1N1

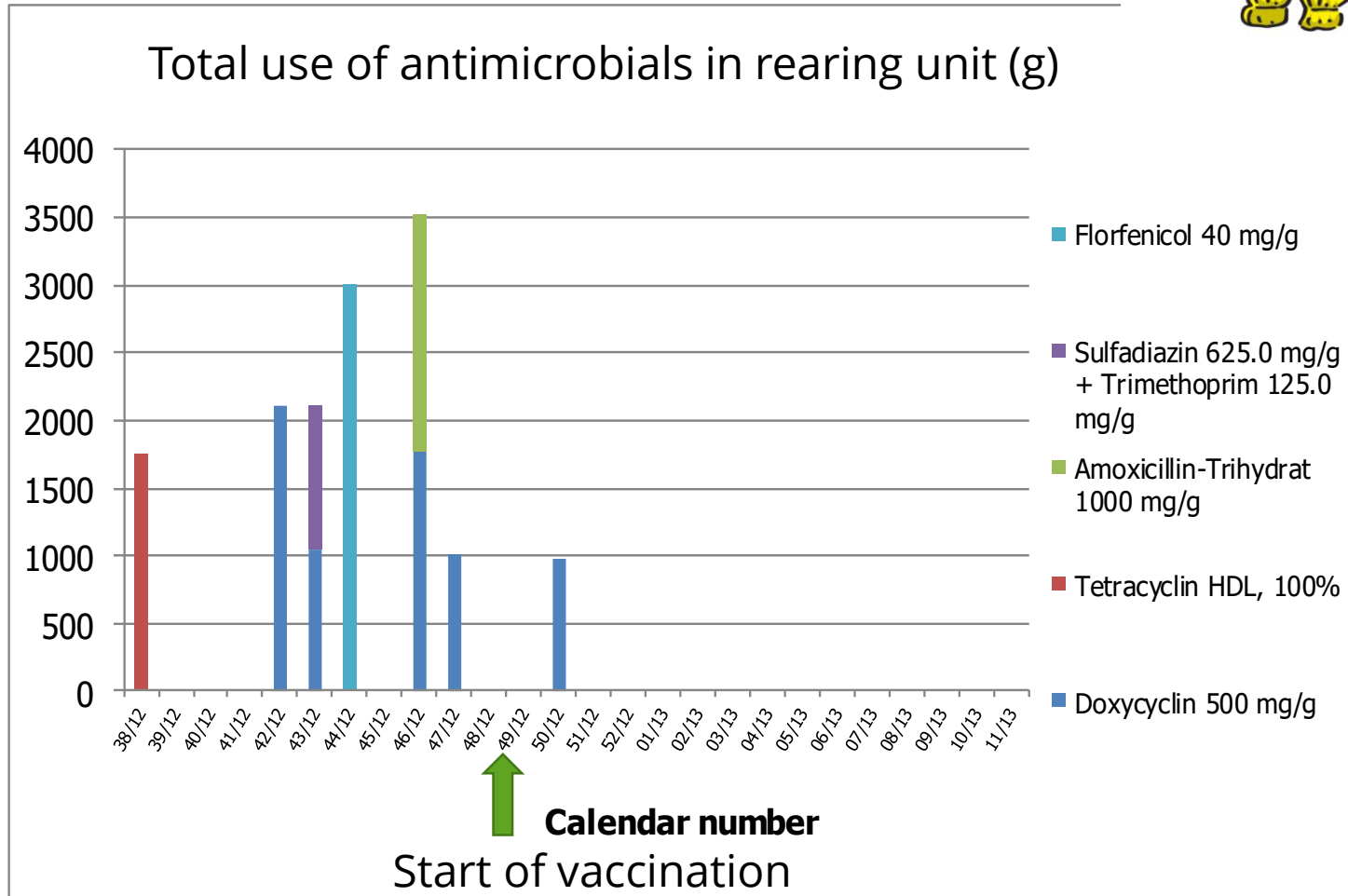
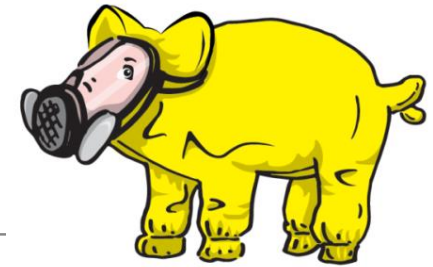
Development



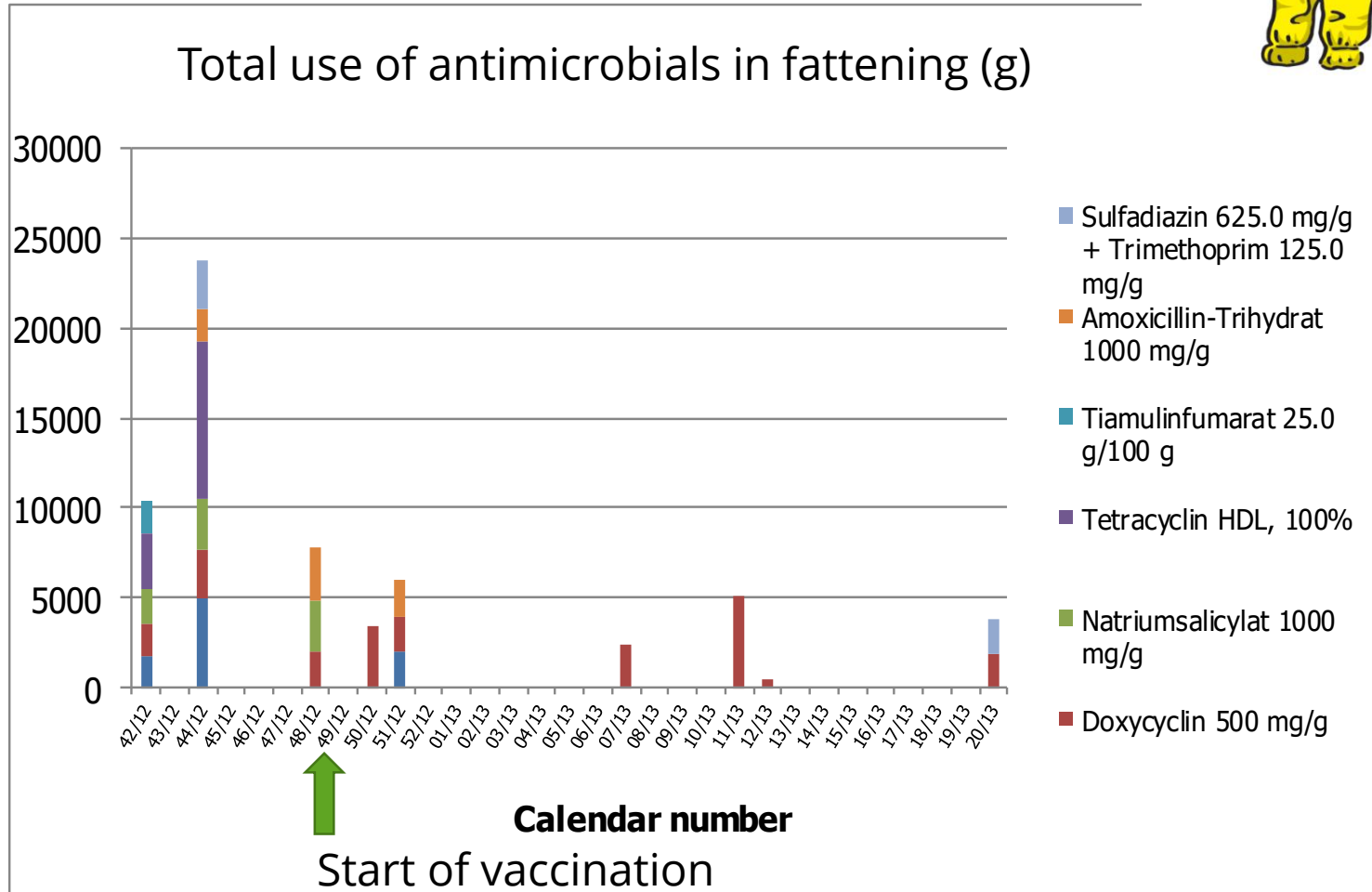
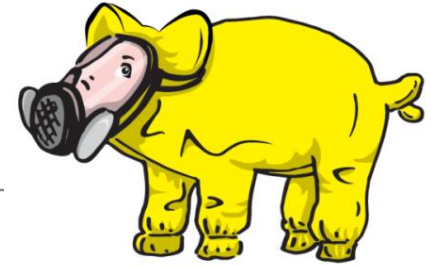
	Mortality (%)	
	6 months before vaccination	6 months after vaccination
Rearing unit	3.0	2.0
Fattening	5.0	3.0

	Average Daily Weight Gain (g)	
	6 months before vaccination	6 months after vaccination
Fattening	620	730

Development

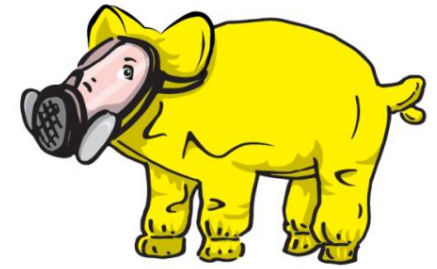


Development

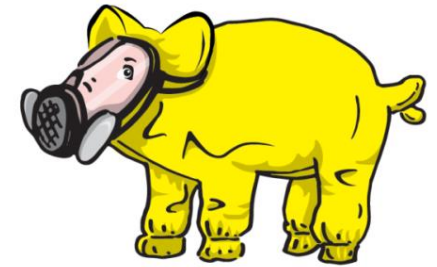


Respiporc FLUpan H1N1

- One Health is a holistic approach -



Overall summary



- Influenza is a serious problem for pigs all over Europe independent of season
- swIAV is becoming more complex
- Diagnostics are difficult, but Vetmarket (and IDT Biologika) is happy to help
 - Which samples to take when (free diagnostic kit)
 - High standard diagnostics at an independent laboratory
 - Help with the interpretation of results
- And when Influenza is diagnosed on your farm, IDT Biologika offers the comprehensive solution!

Respiporc FLU3 & Respiporc FLUpan H1N1

Thank you for your attention!

