OFFLU Swine Influenza Virus technical meeting
27 – 28 February 2019
OIE Headquarters, Paris, France

Gaëlle Simon
Swine Virology Immunology Unit
National Reference Laboratory for Swine Influenza
ANSES, Ploufragan-Plouzané-Niort Laboratory
France

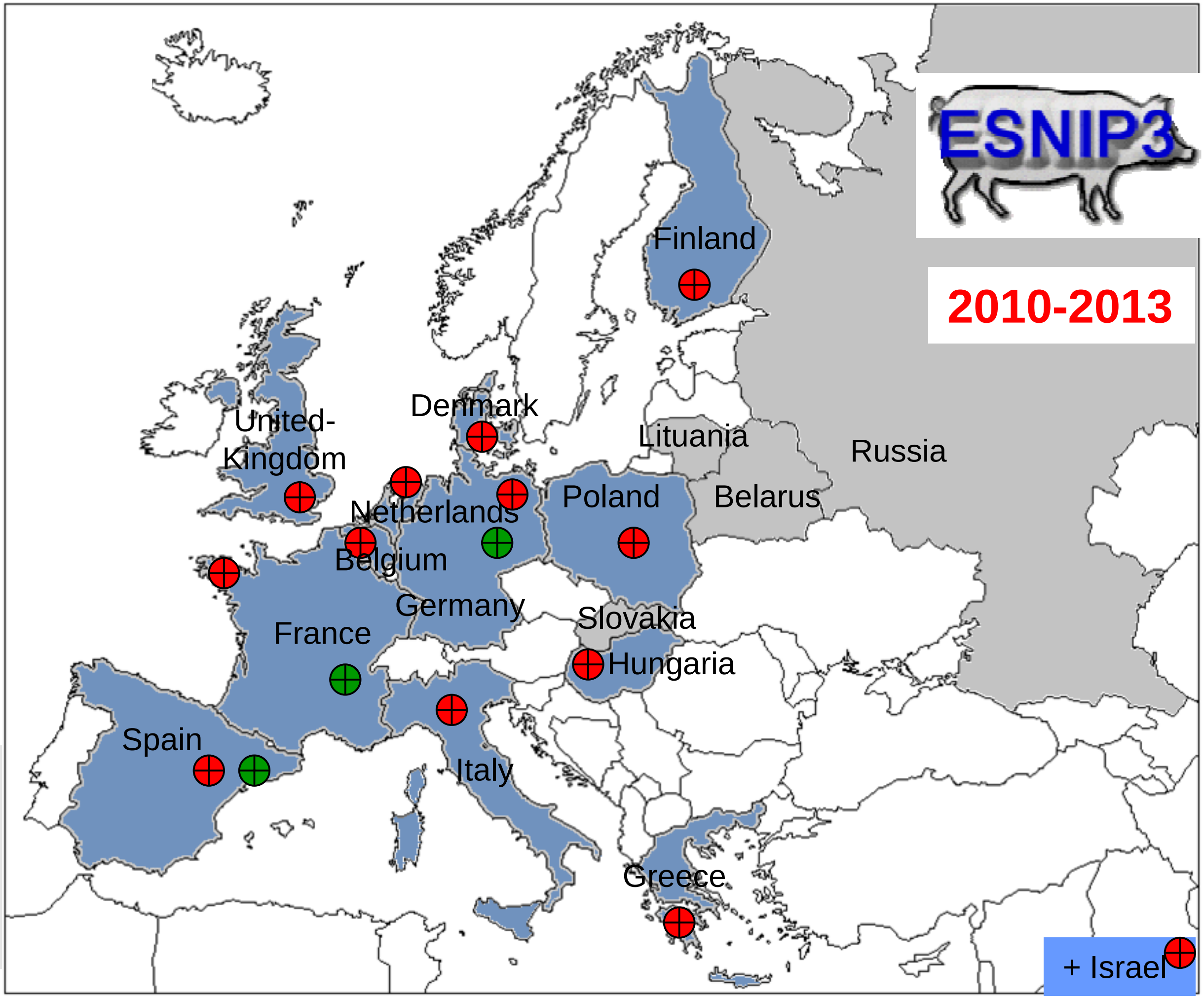
Surveillance in France and in Europe

European Surveillance Network for Influenza in Pigs

- ✓ 13 institutional partners: AHVLA, Anses, CVI, DTU, EVIRA, FLI, IZSLER, KVI, LCV, NVRI, UGhent, UTH, VDD
- ✓ 3 medical companies: IDT Biologika, Merial, Hipra
- ✓ sampling: 17 countries

Passive surveillance
Molecular subtyping (N=1887)
WGS (N=290)
Antigenic subtyping

- ➢ regional variations
- ➢ 23 genetic constellations
- ➢ 10 genotypes = 88% of isolates

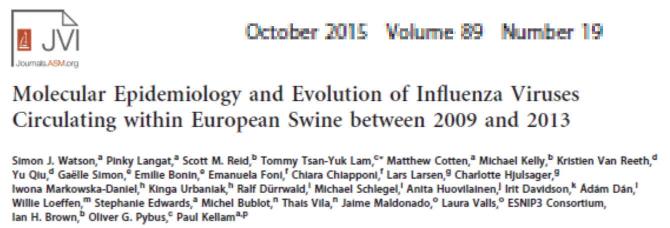



PLOS ONE | DOI:10.1371/journal.pone.0115815 December 26, 2014

RESEARCH ARTICLE

European Surveillance Network for Influenza in Pigs: Surveillance Programs, Diagnostic Tools and Swine Influenza Virus Subtypes Identified in 14 European Countries from 2010 to 2013

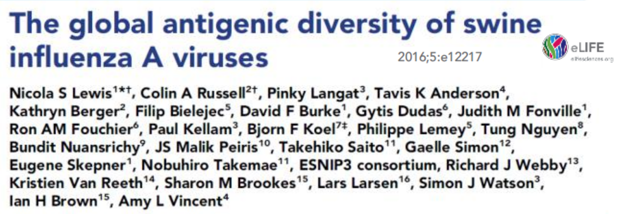
Gaëlle Simon[1*], Lars E. Larsen[2], Ralf Dürrwald[3], Emanuela Foni[4], Timm Harder[5], Kristien Van Reeth[6], Iwona Markowska-Daniel[7], Scott M. Reid[8], Adam Dan[9], Jaime Maldonado[10], Anita Huovilainen[11], Charalambos Billinis[12], Irit Davidson[13], Montserrat Agüero[14], Thaïs Vila[15], Séverine Hervé[1], Solvej Østergaard Breum[2], Chiara Chiapponi[4], Kinga Urbaniak[7], Constantinos S. Kyriakis[12], ESNIP3 consortium[¶], Ian H. Brown[8], Willie Loeffen[16]


JVI Journals.ASM.org

October 2015 Volume 89 Number 19

Molecular Epidemiology and Evolution of Influenza Viruses Circulating within European Swine between 2009 and 2013

Simon J. Watson,[a] Pinky Langat,[a] Scott M. Reid,[b] Tommy Tsan-Yuk Lam,[c*] Matthew Cotten,[a] Michael Kelly,[b] Kristien Van Reeth,[d] Yu Qiu,[d] Gaëlle Simon,[e] Emilie Bonin,[e] Emanuela Foni,[f] Chiara Chiapponi,[f] Lars Larsen,[g] Charlotte Hjulsager,[g] Iwona Markowska-Daniel,[h] Kinga Urbaniak,[h] Ralf Dürrwald,[i] Michael Schlegel,[i] Anita Huovilainen,[j] Irit Davidson,[k] Ádám Dán,[l] Willie Loeffen,[m] Stephanie Edwards,[a] Michel Bublot,[n] Thais Vila,[n] Jaime Maldonado,[o] Laura Valls,[o] ESNIP3 Consortium, Ian H. Brown,[b] Oliver G. Pybus,[c] Paul Kellam[a,p]

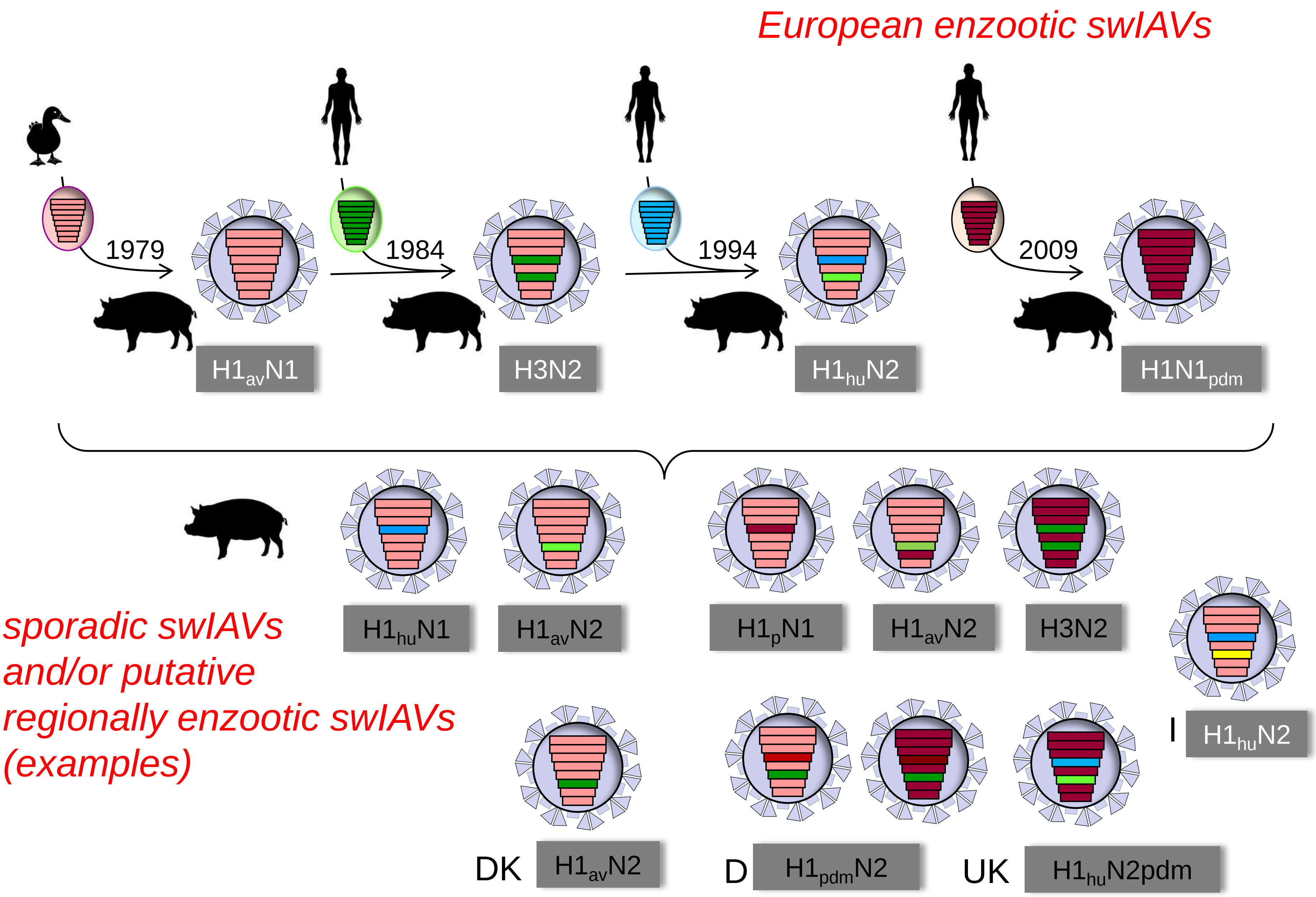

The global antigenic diversity of swine influenza A viruses

2016;5:e12217 eLIFE

Nicola S Lewis[1*†], Colin A Russell[2†], Pinky Langat[3], Tavis K Anderson[4], Kathryn Berger[2], Filip Bielejec[5], David F Burke[1], Gytis Dudas[6], Judith M Fonville[1], Ron AM Fouchier[6], Paul Kellam[3], Bjorn F Koel[7‡], Philippe Lemey[5], Tung Nguyen[8], Bundit Nuansrichy[9], JS Malik Peiris[10], Takehiko Saito[11], Gaelle Simon[12], Eugene Skepner[1], Nobuhiro Takemae[11], ESNIP3 consortium, Richard J Webby[13], Kristien Van Reeth[14], Sharon M Brookes[15], Lars Larsen[16], Simon J Watson[3], Ian H Brown[15], Amy L Vincent[4]

swIAVs in Europe (2010-2013)

Simon et al., 2014 ; Watson et al., 2015



European enzootic swIAVs

1979 $H1_{av}N1$

1984 H3N2

1994 $H1_{hu}N2$

2009 $H1N1_{pdm}$

sporadic swIAVs and/or putative regionally enzootic swIAVs (examples)

$H1_{hu}N1$

$H1_{av}N2$

$H1_{p}N1$

$H1_{av}N2$

H3N2

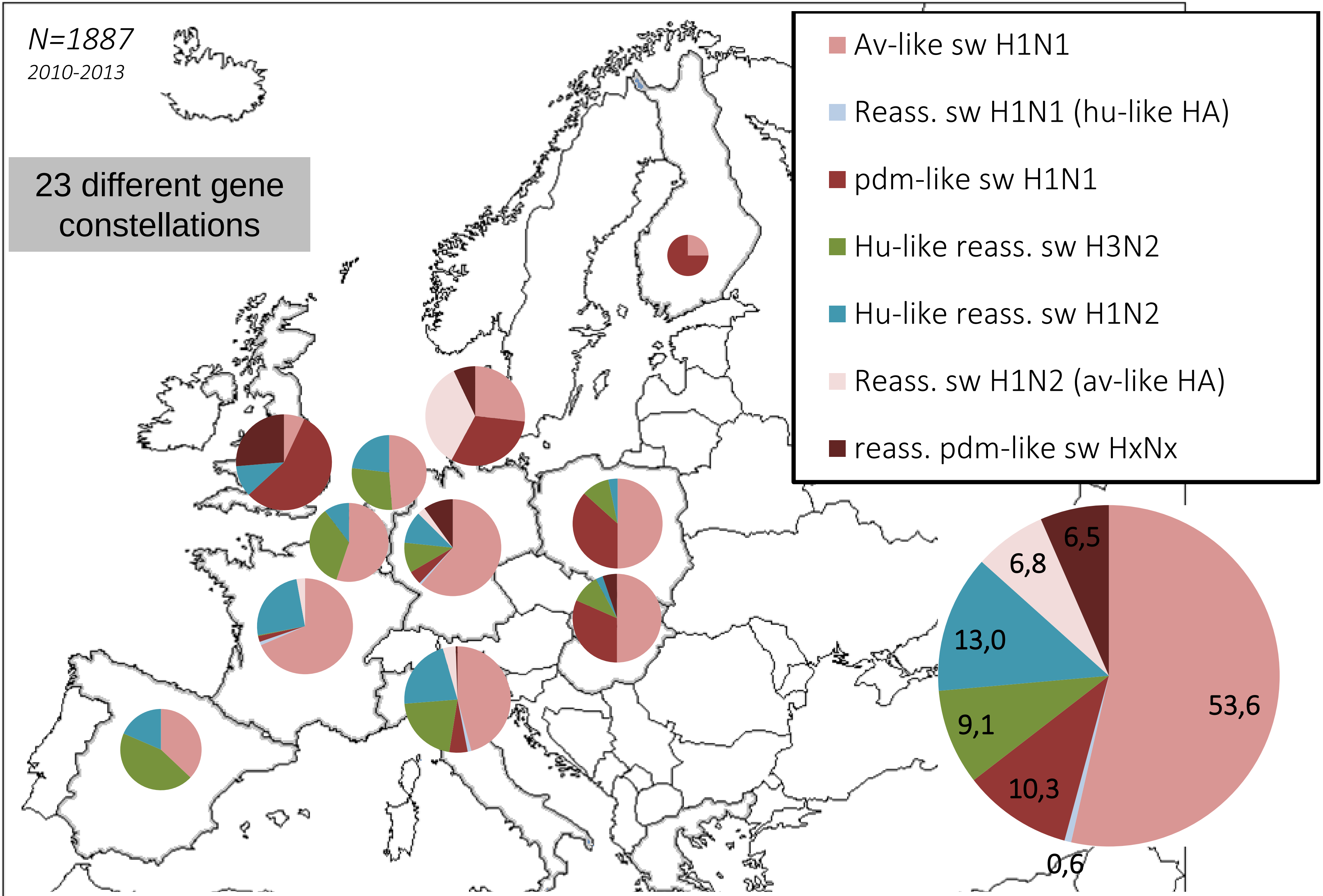
I $H1_{hu}N2$

DK $H1_{av}N2$

D $H1_{pdm}N2$

UK $H1_{hu}N2pdm$

Regional variation in the epidemiology of European swIAVs



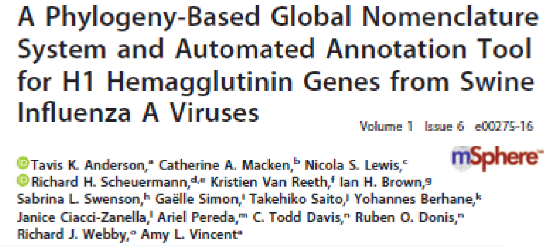
N=1887

2010-2013

23 different gene constellations

Simon *et al.*, 2014

H1 viruses identified in Europe

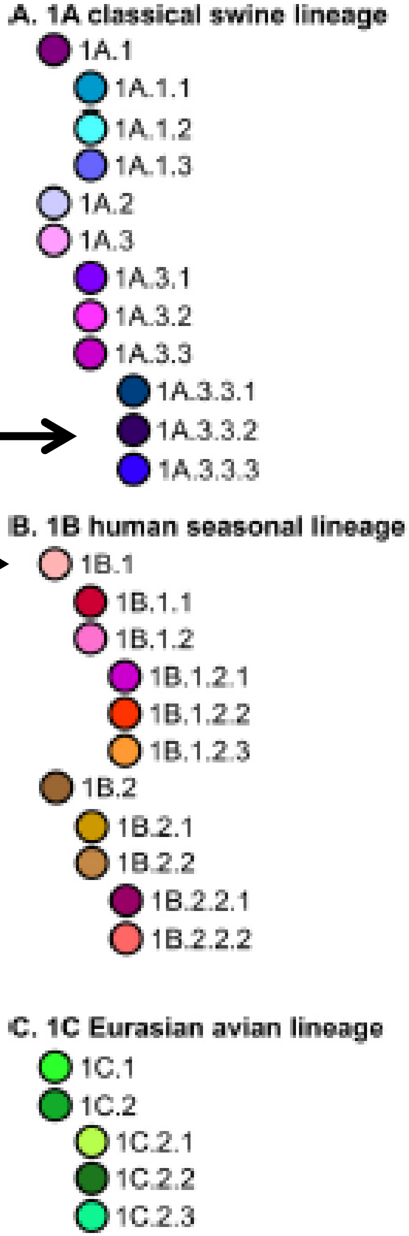


A Phylogeny-Based Global Nomenclature System and Automated Annotation Tool for H1 Hemagglutinin Genes from Swine Influenza A Viruses

Volume 1 Issue 6 e00275-16

mSphere™

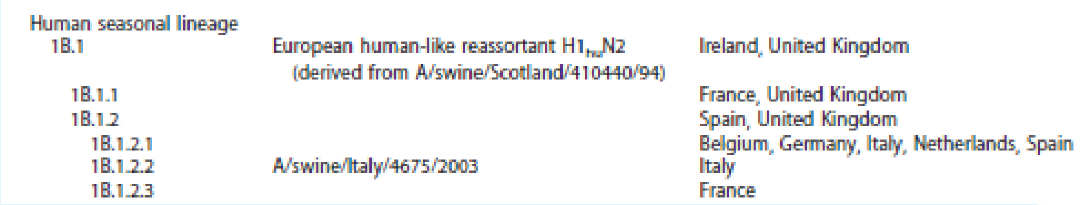
Tavis K. Anderson,[a] Catherine A. Macken,[b] Nicola S. Lewis,[c] Richard H. Scheuermann,[d,e] Kristien Van Reeth,[f] Ian H. Brown,[g] Sabrina L. Swenson,[h] Gaëlle Simon,[i] Takehiko Saito,[j] Yohannes Berhane,[k] Janice Ciacci-Zanella,[l] Ariel Pereda,[m] C. Todd Davis,[n] Ruben O. Donis,[n] Richard J. Webby,[o] Amy L. Vincent[a]



H1pdm → 1A.3.3.2

$H1_{hu}$ → 1B.1

$H1_{av}$ → C. 1C Eurasian avian lineage

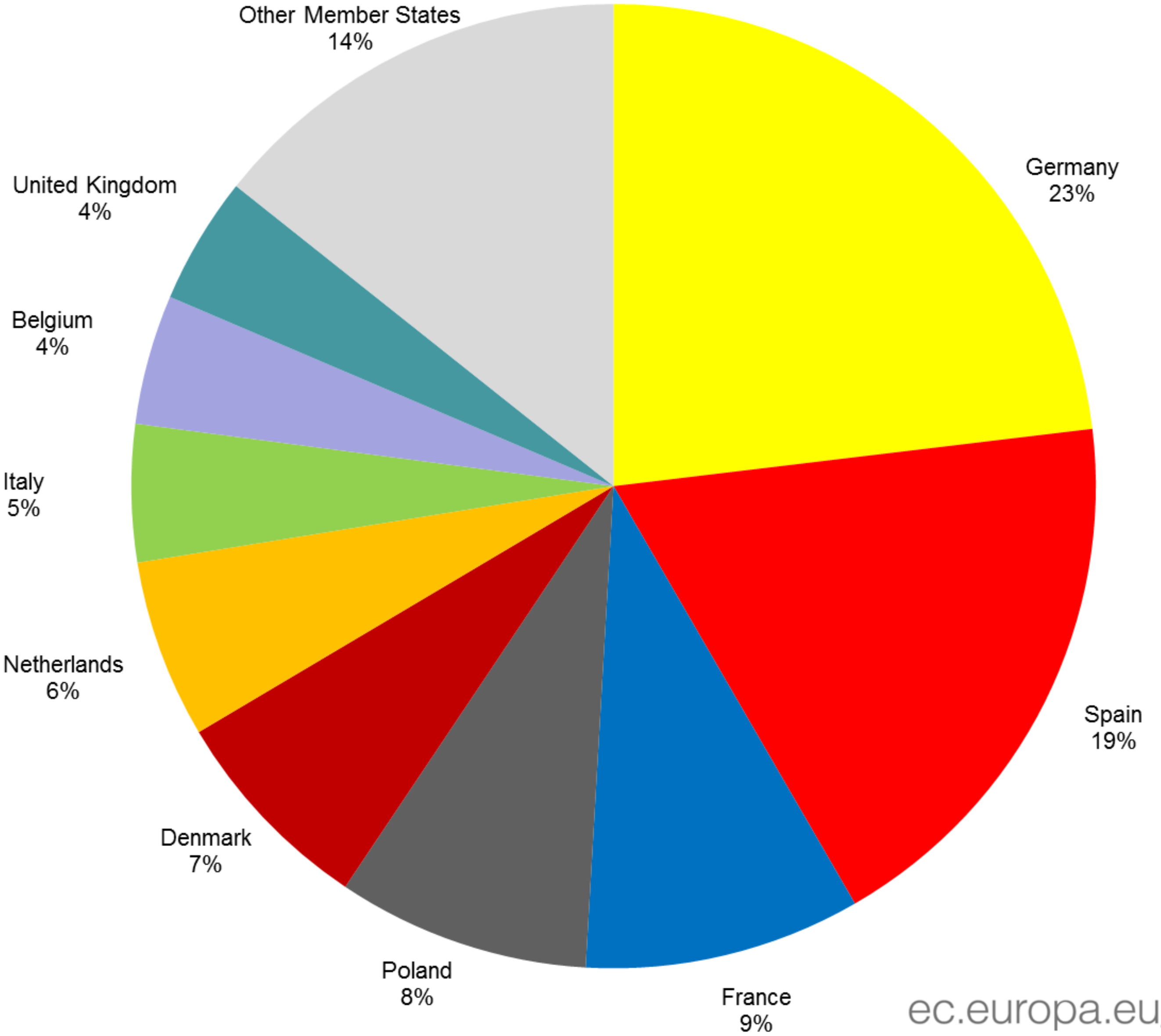


Human seasonal lineage		
1B.1	European human-like reassortant $H1_{hu}N2$ (derived from A/swine/Scotland/410440/94)	Ireland, United Kingdom
1B.1.1		France, United Kingdom
1B.1.2		Spain, United Kingdom
1B.1.2.1		Belgium, Germany, Italy, Netherlands, Spain
1B.1.2.2	A/swine/Italy/4675/2003	Italy
1B.1.2.3		France

Eurasian avian lineage		
1C.1	Avian-like swine $H1_{av}N1$ (derived from A/swine/Amsberg/6554/1979 and A/swine/Belgium/WVL1/1979)	Belgium, Canada, France, Germany, Hong Kong, Ireland, Italy, Netherlands, Spain, United Kingdom
1C.2	Avian-like swine $H1_{av}N1$ (derived from A/swine/Ille et Vilaine/1455/1999)	Belgium, Denmark, Finland, Germany, Italy, Mexico, Netherlands, Poland, Sweden
1C.2.1		Belgium, Denmark, France, Germany, Hungary, Italy, Netherlands, Poland, Russia, Spain
1C.2.2		France, Germany, Italy, Luxembourg, Netherlands, Poland, Spain
1C.2.3		China, Czech Republic, France, Hong Kong, Italy, Poland, South Korea

Pigmeat: slaughterings in the EU Member States, 2016

(% of EU total, based on number of animals)



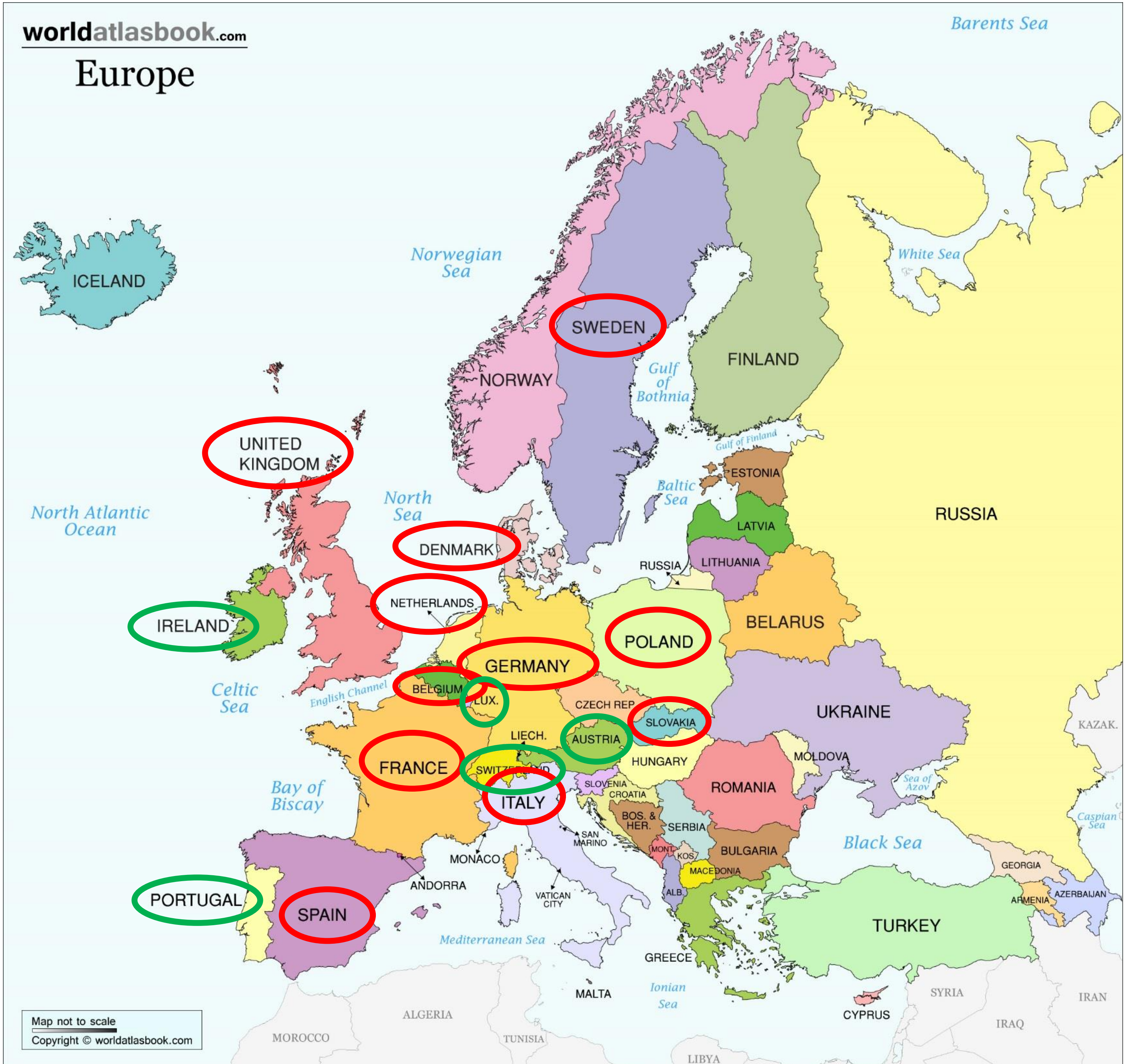
ec.europa.eu/**eurostat**

2014-2018

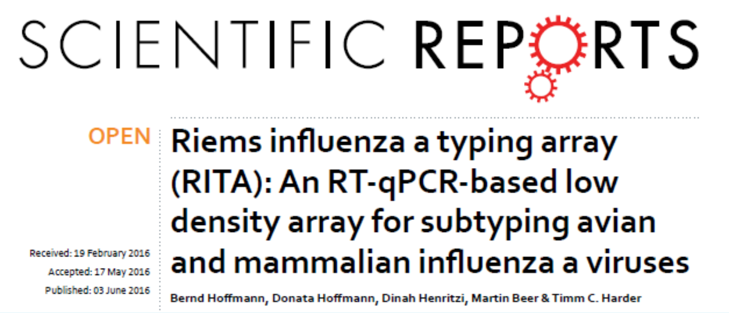
Summary of data provided by volunteer collaborators

ANSES
APHA
DTU
IZSLER
IZSVE
NVRI
UAB
Ughent
SVA
IDT
Biologika/FLI

+ Peer-reviewed papers



Diagnostic tools



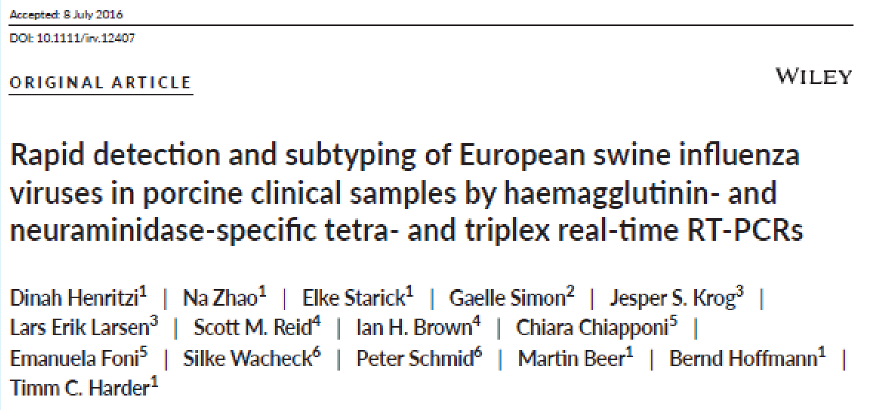
SCIENTIFIC REPORTS

OPEN

Riems influenza a typing array (RITA): An RT-qPCR-based low density array for subtyping avian and mammalian influenza a viruses

Received: 19 February 2016
Accepted: 17 May 2016
Published: 03 June 2016

Bernd Hoffmann, Donata Hoffmann, Dinah Henritzi, Martin Beer & Timm C. Harder



Accepted: 8 July 2016
DOI: 10.1111/irv.12407

ORIGINAL ARTICLE

WILEY

Rapid detection and subtyping of European swine influenza viruses in porcine clinical samples by haemagglutinin- and neuraminidase-specific tetra- and triplex real-time RT-PCRs

Dinah Henritzi[1] | Na Zhao[1] | Elke Starick[1] | Gaelle Simon[2] | Jesper S. Krog[3] | Lars Erik Larsen[3] | Scott M. Reid[4] | Ian H. Brown[4] | Chiara Chiapponi[5] | Emanuela Foni[5] | Silke Wacheck[6] | Peter Schmid[6] | Martin Beer[1] | Bernd Hoffmann[1] | Timm C. Harder[1]

Bonin *et al. Virology Journal* (2018) 15:7
DOI 10.1186/s12985-018-0920-z

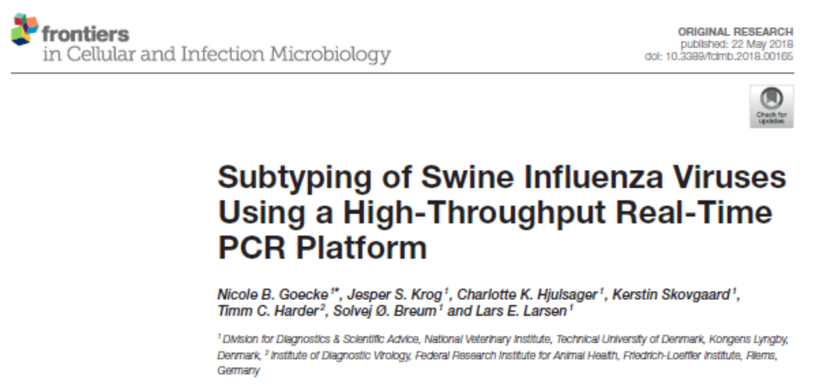
Virology Journal

METHODOLOGY **Open Access**

CrossMark

Molecular subtyping of European swine influenza viruses and scaling to high-throughput analysis

Emilie Bonin[1,2,5], Stéphane Quéguiner[1,2], Cédric Woudstra[3], Stéphane Gorin[1,2], Nicolas Barbier[1,2], Timm C. Harder[4], Patrick Fach[3], Séverine Hervé[1,2] and Gaëlle Simon[1,2*]



frontiers in Cellular and Infection Microbiology

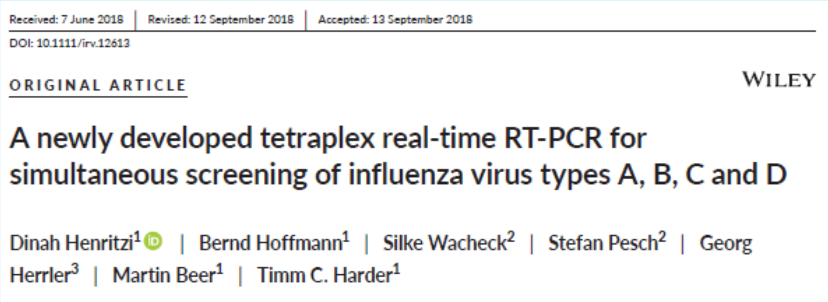
ORIGINAL RESEARCH
published: 22 May 2018
doi: 10.3389/fcimb.2018.00165

Check for updates

Subtyping of Swine Influenza Viruses Using a High-Throughput Real-Time PCR Platform

Nicole B. Goecke[1], Jesper S. Krog[1], Charlotte K. Hjulsager[1], Kerstin Skovgaard[1], Timm C. Harder[2], Solvej Ø. Breum[1] and Lars E. Larsen[1]*

[1] Division for Diagnostics & Scientific Advice, National Veterinary Institute, Technical University of Denmark, Kongens Lyngby, Denmark, [2] Institute of Diagnostic Virology, Federal Research Institute for Animal Health, Friedrich-Loeffler Institute, Riems, Germany



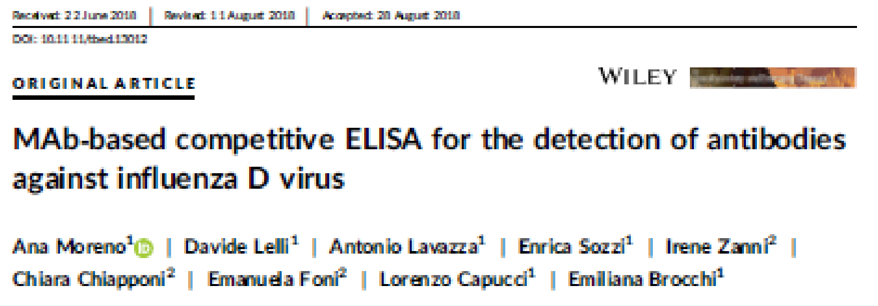
Received: 7 June 2018 | Revised: 12 September 2018 | Accepted: 13 September 2018
DOI: 10.1111/irv.12613

ORIGINAL ARTICLE

WILEY

A newly developed tetraplex real-time RT-PCR for simultaneous screening of influenza virus types A, B, C and D

Dinah Henritzi[1] | Bernd Hoffmann[1] | Silke Wacheck[2] | Stefan Pesch[2] | Georg Herrler[3] | Martin Beer[1] | Timm C. Harder[1]



Received: 22 June 2018 | Revised: 11 August 2018 | Accepted: 20 August 2018
DOI: 10.1111/tbed.13012

ORIGINAL ARTICLE

WILEY

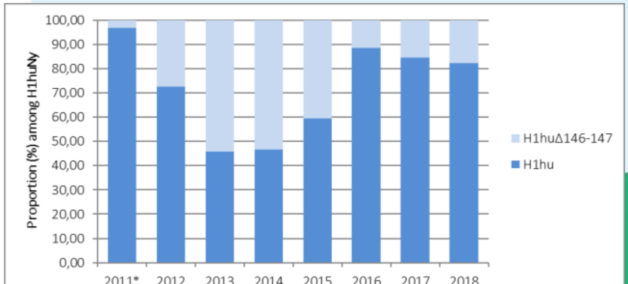
MAb-based competitive ELISA for the detection of antibodies against influenza D virus

Ana Moreno[1] | Davide Lelli[1] | Antonio Lavazza[1] | Enrica Sozzi[1] | Irene Zanni[2] | Chiara Chiapponi[2] | Emanuela Foni[2] | Lorenzo Capucci[1] | Emiliana Brocchi[1]

France - Anses UVIP
NRL for Swine Influenza

					Number of strains per lineage					
					H1N1			H3N2	H1N2	
Period	Number of herds investigated	Number of Influenza A positive cases	Frequency of positive cases (%)	Number of subtyped viruses*	$H1_{av}N1$	H1N1pdm	$H1_{pdm}N1$	H3N2	$H1_{hu}N2$	$H1_{av}N2$
2014	273	129	47,25	**103**	**75**	**3**		**2**	**22**	**1**
2015	378	193	51,06	**147**	**101**	**8**		**1**	**32**	**5**
2016	459	218	47,49	**153**	**107**	**19**		**1**	**23**	**3**
2017	342	146	42,69	**110**	**82**	**2**		**0**	**26**	**0**
2018	349	166	47,56	**133**	**89**	**10**	**1**	**0**	**29**	**5**
2014-2018	**1801**	**852**	**47,31**	**646**	**454**	**42**	**1**	**4**	**132**	**14**

- ✓ **Regionalization of the different subtypes – 13 different gene constellations**
- ✓ Last detection of a $H1_{hu}N1$ reassortant in 2012
- ✓ Emergence of a **$H1_{hu}\Delta146$-147 variant** (drift) in 2011 > **$H1_{hu}N2v$ *versus* $H1_{hu}N2$**
- ✓ **Several sub-populations of $H1_{av}N2$**: $H1_{av}N1xH1_{hu}N2$ or $H1_{av}N1xH3N2$ or $H1_{av}N2$ closest to the DK virus or $H1_{av}N1x$ seasonal H3N2
- ✓ **First reassortants with 1 or more gene(s) from H1N1pdm detected in 2016**
- ✓ H3N2 with IGC from H1N1pdm
- ✓ $H1_{av}N2$ (with seasonal N2) **with Mpdm gene**
- ✓ **Novel swine-divergent (swD) genogroup** among H1N1pdm isolates



Serological survey in Corsica
(extensive breeding)
45,8% of pig farms H1N1pdm positive
20.8% of pig farms $H1_{av}N1$ positive
1.4% of wild boars IAV positive (same subtypes)


Preventive Veterinary Medicine 157 (2018) 94-98

Contents lists available at ScienceDirect

Preventive Veterinary Medicine

journal homepage: www.elsevier.com/locate/prevetmed

Serological survey of influenza A viruses in domestic and wild *Suidae* in Corsica (France), a Mediterranean island environment

Sébastien Grech-Angelini[a,*], Séverine Hervé[b,d], Nicolas Rose[c,d], Nicolas Barbier[b,d], François Casabianca[a], Oscar Maestrini[a], Alessandra Falchi[e], Gaëlle Simon[b,d]

Journal of Virology® December 2018 Volume 92 Issue 24 e00988-18

Spatiotemporal Distribution and Evolution of the A/H1N1 2009 Pandemic Influenza Virus in Pigs in France from 2009 to 2017: Identification of a Potential Swine-Specific Lineage

Amélie Chastagner,[a,c] Séverine Hervé,[a,c] Emilie Bonin,[a,c*] Stéphane Quéguiner,[a,c] Edouard Hirchaud,[b,c] Dinah Henritzi,[d]
Véronique Béven,[b,c] Stéphane Gorin,[a,c] Nicolas Barbier,[a,c] Yannick Blanchard,[b,c] Gaëlle Simon[a,c]

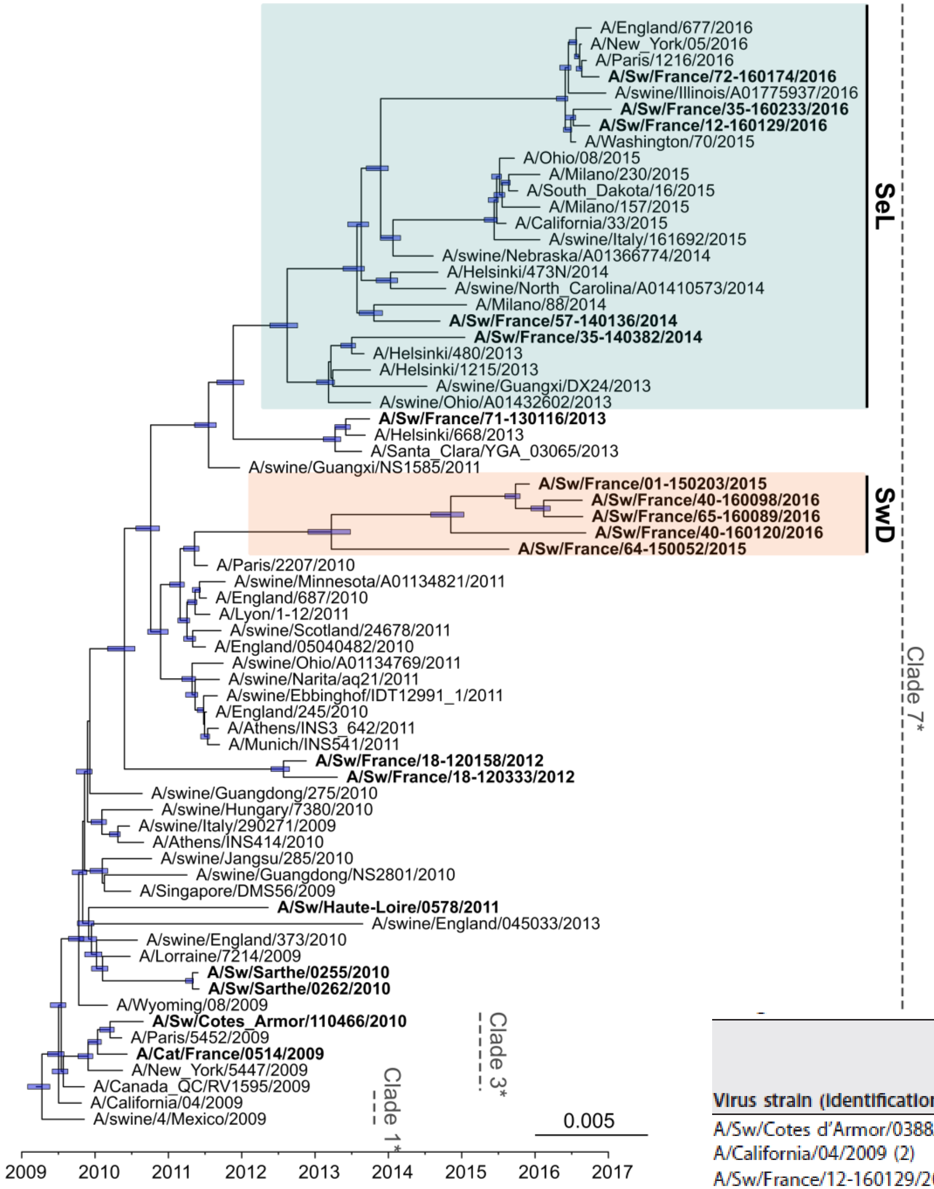
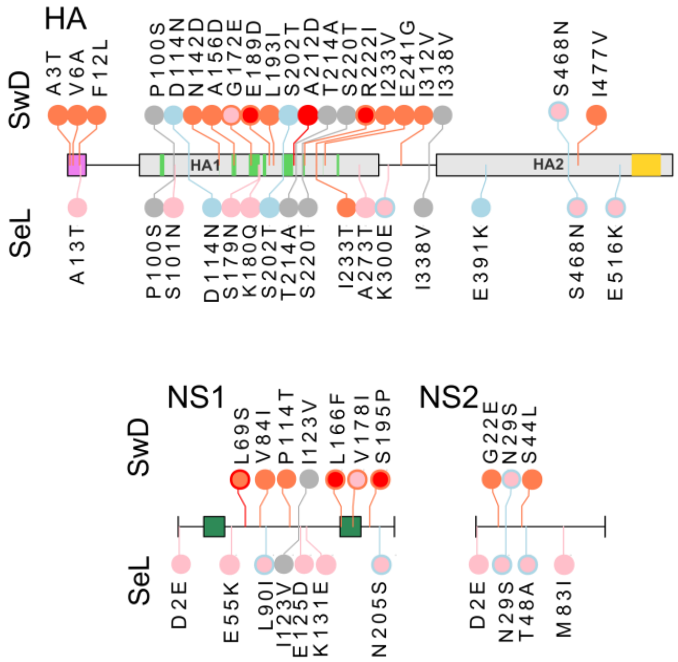


FIG 3 Bayesian inference tree of H1N1pdm virus strains from whole-genome sequences. Strains isolated from pigs (and a cat) in France are indicated in boldface type. Nodes supported by more than 50% of sampled trees are indicated by a blue bar displaying the 95% highest posterior density (HPD) intervals of the node heights. The SeL and SwD genogroups were defined in this study. *, clades 1, 3, and 7 correspond to the classification of Nelson et al. (33).



		Hemagglutination inhibition titer with sera against:		
Virus strain (identification no.)[b]	Lineage (clade)	$H1_{av}N1$ A/Sw/CA/0388/2009	H1N1pdm A/California/04/2009	H1N1pdm A/Sw/Sarthe /0255/2010
A/Sw/Cotes d'Armor/0388/2009 (1)	$H1_{av}N1$	**640**	<10	<10
A/California/04/2009 (2)	H1N1pdm (3)	40	**640**	640
A/Sw/France/12-160129/2016 (15)	H1N1pdm (SeL)	20	160	320
A/Sw/France/35-160233/2016 (16)	H1N1pdm (SeL)	10	80	80
A/Sw/France/72-160174/2016 (17)	H1N1pdm (SeL)	40	160	320
A/Sw/France/64-150052/2015 (18)	H1N1pdm (SwD)	10	80	160
A/Sw/France/65-160089/2016 (19)	H1N1pdm (SwD)	10	80	320
A/Sw/France/40-160098/2016 (20)	H1N1pdm (SwD)	20	160	320

United-Kingdom - APHA

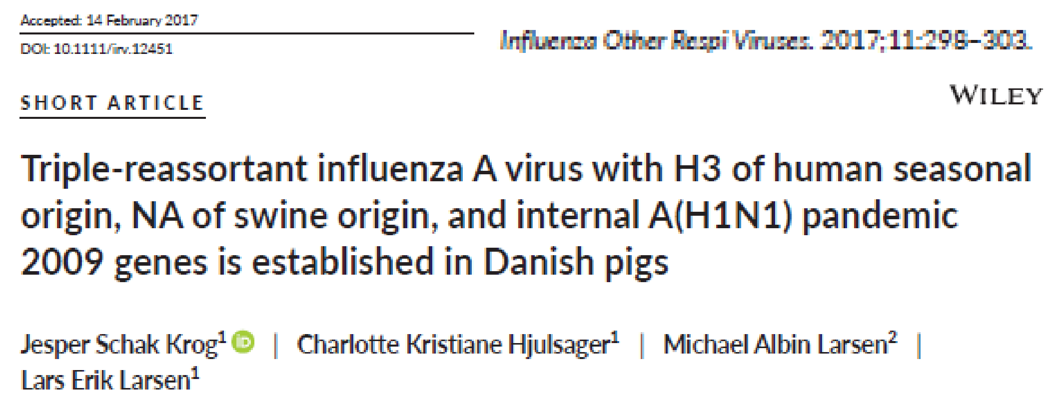
					Number of strains per lineage			
					H1N1		H3N2	H1N2
Period	Number of herds investigated	Number of Influenza A positive cases	Frequency of positive cases (%)	Number of subtyped viruses	$H1_{av}N1$	H1N1pdm	H3N2	$H1_{hu}N2$*
2014	184	73	39.7	**27**	**3**	**11**		**13**
2015	171	44	25.7	**16**	**1**	**8**		**7**
2016	174	58	33.3	**18**	**3**	**4**		**11**
2017	133	23	17.3	**10**	**0**	**2**		**8**
2018	140	70	50.0	**17**	**3**	**0**		**14**
2014-2018	**802**	**268**	**33.4**	**88**	**10**	**25**	**0**	**53**

- *$H1_{hu}N2$ include reassortant viruses with IGC from H1N1pdm
- Several partially subtyped viruses (H?N1 or H?N2): questions about a possible drift in H1pdm – to be confirmed

Denmark - DTU

Period	Number of herds investigated	Number of Influenza A positive cases	Frequency of positive cases (%)	Number of subtyped viruses*	Number of strains per lineage (frequencies)					
					H1N1		H3N2	H1N2		Others
					$H1_{av}N1$	H1N1pdm	H3N2	$H1_{hu}N2$	$H1_{av}N2$	
2014	422	199	47.15	80		17.5%	2.5%		68.75%	11.25%
2015	488	256	52.45	176		25.6%	2.8%		64.8%	6.8%
2016	388	204	52.57	123		18.7%	2.4%		70.8%	8.1%
2017	362	195	53.87	147		15.6%	0%		78.2%	6.2%
2018	410	246	60.0	176		14.8%	0.56 %		74.4%	10.24%
2014-2018	**2070**	**1100**	**53.14**	**702**	**0%**	**18.4 %**	**1.65 %**	**0%**	**71.4 %**	**8.52%**

- ✓ $H1_{av}N2$ enzootic in Danish pigs since 2003 *(Trebbien et al., 2013)*
- ✓ **2012**: sporadic reassortants? 1 with 7 segments from $H1_{av}N1$ and a N2 from a seasonal H3N2 from mid-90' *(Breum et al., 2013)* + 1 with 7 segments from H1N1pdm and a N2 gene from H3N2 (H1pdmN2sw)
- ✓ **2015**: Triple reassortant H3N2 : H3 from a seasonal H3N2 2004-2005 + N2sw + IGC from H1N1pdm



Accepted: 14 February 2017
DOI: 10.1111/irv.12451
Influenza Other Respi Viruses. 2017;11:298–303.

SHORT ARTICLE

WILEY

Triple-reassortant influenza A virus with H3 of human seasonal origin, NA of swine origin, and internal A(H1N1) pandemic 2009 genes is established in Danish pigs

Jesper Schak Krog[1] | Charlotte Kristiane Hjulsager[1] | Michael Albin Larsen[2] | Lars Erik Larsen[1]

Sweden - National Veterinary Institute (SVA)

						Number of swIAV strains					
						H1N1		**H3N2**	**H1N2**		
Period	**Surveillance program**	**Number of herds investigated**	**Number of Influenza A positive cases**	**Frequency of positive cases (%)**	**Number of subtyped viruses***	**$H1_{av}N1$**	**H1N1pdm**	**H3N2**	**$H1_{hu}N2$**	**$H1_{av}N2$**	**$H1_{pdm}N2$**
2014	**Passive**	**18**	**7 (40 pigs)**	**38 (27)**	**40**		**19**			**14**	**7**
	*Active**	10	5 (79 pigs)	50 (9)	**79**		**60**			**5**	**14**
2015	**Passive**	**8**	**2 (6 pigs)**	**25 (22)**	**6**		**3**			**3**	**0**
	*Active**	10	4 (20 pigs)	40 (2)	**20**		**12**			**6**	**2**
2016	**Passive**	**7**	**2**	**1 pig/herd**	**2**		**1**			**1**	**0**
2017	**Passive**	**9**	**3**	**1 pig/herd**	**3**		**2**			**1**	**0**
2018	**Passive**	**9**	**0**	**0**	**0**		**0**			**0**	**0**
Total (passive)		**51**	**14**	**27,45**	**150**	**0**	**97**	**0**	**0**	**30**	**23**

*RT-qPCR subtyping and WGS on selected numbers of representative strains

**In coll. with farmer's association - 10 vets who selected 10 pig farms representative of the pig production systems in Sweden. From first week of Feb. 2014 or 2015, the farms were visited every second week for 6 consecutive visits. 15 nasal swabs/farm/visit.

2009: $H1_{av}N2$ exclusively

2013: clinical outbreak in southern part of Sweden, due to H1N1pdm

2014: detection of a novel H1N2 reassortant, derived from H1N1pdm (HA+ IGC) and $H1_{av}N2$ (NA) + transmissions of $H1_{pdm}N2$ to 2 swine farmers (asymptomatic cases) without further human infections (Who, 2014)

Belgium and Netherlands Virology, Ghent University

					Number of strains per lineage				
					H1N1		H3N2	H1N2	Others
Period	**Number of herds investigated**	**Number of Influenza A positive cases**	**Frequency of positive cases (%)**	**Number of subtyped viruses**[1]	**$H1_{av}N1$**	**H1N1pdm**	**H3N2**	**$H1_{hu}N2$**	Co-infections H3N2 x $H1_{av}N1$
2014	**51**	**23**	**45**	**23**	**10**		**8**	**5**	
2015	**11**	**3**	**27**	**3**	**3**		**0**	**0**	
2016	**3**	**2**		**2**	**1**		**0**	**1**	
2017	**24**	**8**	**33**	**8**	**4**		**2**	**2**	
2018	**58**	**21**	**36**	**24**[2]	**10**		**0**	**8**	**3**
2014-2018	**147**	**57**	**39**	**60**	**28**	**0?**	**10**	**16**	**3**
2019	18	6	33	6	5		0	1	0

[1] Isolates were subtyped by HI assay with 7 distinct hyperimmune sera, NI assays are pending. For some isolates, comparable HI titers were obtained with sera against $H1_{av}$ viruses and H1pdm. For now these isolates are included in $H1_{av}$ column, final confirmation of HA lineage is needed.

Remark: 43 isolates were also subtyped by multiplex PCR at APHA - except for one isolate, HA lineage was consistent with that in HI assay.

[2] Three samples yielded two virus isolates (co-infections with H3N2 and $H1_{av}N1$).

Poland and Slovakia - NVRI

						Number of strains per lineage					
						H1N1		H3N2	H1N2		
Country	Period	Number of herds investigated	Number of Influenza A positive cases	Frequency of positive cases (%)	Number of subtyped viruses	$H1_{av}N1$	H1N1pdm	H3N2	$H1_{hu}N2$	$H1_{av}N2$	$H1_{pdm}N2$
Poland	**2014**	50	14	28,00	6	2	3			1	0
Slovakia		10	1	10,00	1	0	1			0	0
Poland	**2015**	41	6	14,63	4	1	2			0	1
Slovakia		1	0	0,00	0	0	0			0	0
Poland	**2016**	31	10	32,26	0	0	0			0	0
Poland	**2017**	15	5	33,33	2	1	1			0	0
Poland	**2018**	32	11	34,38	1	0	0			1	0
Total		**180**	**47**	**26,11**	**14**	**4**	**7**	**0**	**0**	**2**	**1**

Further subtyping analyses ongoing

Animal species	Year	Number of sera	Number / % of positive sera (>20)				
			$H1_{av}$	$H1_{hu}$	H3	H1pdm	positive for more than 1 serotype*
swine	2015	923	9 / 0,97%	40 /4,33%	7 / 0,76%	61 / 6,61%	29 / 3,14%
	2016	2914	174 / 5,97%	5 / 0,17%	47 / 1,61%	22 / 0,75%	239 / 8,20%
	2017	1518	23 / 1,51%	5 / 0,33%	8 / 0,53%	44 / 2,90%	105 / 6,92%
	2018	4810	106 / 2,20%	12 / 0,25%	94 / 1,95%	67 / 1,39%	71 / 1,48%
wild boar	2017	742	6 / 0,81%	0 / 0,00%	2 / 0,27%	11 / 1,48%	26 /3,50%

*not included in the results for individual serotypes

Italy (1) - IZSLER

*NGS

					Numbers of strains per lineage					
					H1N1		H3N2	H1N2		
Period	Number of herds investigated	Number of Influenza A positive cases	Frequency of positive cases (%)	Number of subtyped viruses*	$H1_{av}N1$	H1N1pdm	H3N2	$H1_{hu}N2$	$H1_{av}N2$	$H1_{pdm}N2$
2014	444	110	24,77	48	13	2	10	14	6	3
2015	498	121	24,30	47	10	5	7	16	9	0
2016	568	129	22,71	55	13	8	16	14	4	0
2017	648	194	29,94	86	42	1	15	13	15	0
2018	646	193	29,88	91	38	1	9	26	17	0
2014-2018	2804	747	26,64	327	116	17	57	83	51	3

RT_PCR

		H1N1		H3N2	H1N2
Period	Number of subtyped viruses	H1N1	H1N1pdm	H3N2	H1N2
2014	102	36	2	13	51
2015	108	40	12	11	45
2016	123	45	13	36	29
2017	175	75	2	26	72
2018	158	56	2	19	81
2014-2018	666	252	31	105	278

- **Extremely high genomic heterogeneity in Italy in the last two decades**
- 27 different gene constellations
- **3 genotypes with the Mpdm gene** in old enzootic swIAVs background

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ORIGINAL ARTICLE

WILEY

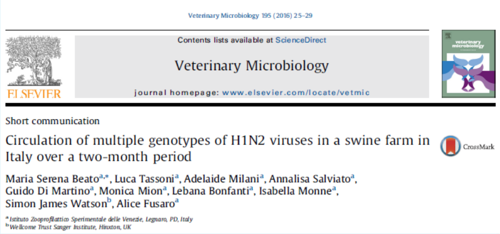
Genetic analysis of human and swine influenza A viruses isolated in Northern Italy during 2010–2015

C. Chiapponi[1,2] | E. Ebranati[3] | E. Pariani[4] | S. Faccini[2] | A. Luppi[2] | L. Baioni[1,2] | R. Manfredi[1,2] | V. Carta[3] | M. Merenda[1,2] | P. Affanni[5] | M. E. Colucci[5] | L. Veronesi[5] |

Italy (2) - IZSVE

					Number of strains per lineage					
					H1N1		H3N2	H1N2		Other
Period	Number of herds investigated	Number of Influenza A positive cases	Frequency of positive cases (%)	Number of subtyped viruses*	$H1_{av}N1$	H1N1pdm	H3N2	$H1_{hu}N2$	$H1_{av}N2$	H3N1
2014	0	0		9	2	1	1	5	0	0
2015	21	4	19,05	3	3	0	0	0	0	0
2016	26	6	23,08	2	1	1	0	0	0	0
2017	51	19	37,25	15	4	1	1	0	9	0
2018	65	15	23,08	19	2	7	0	3	6	1
2014-2018	163	44	26,99	48	12	10	2	8	15	1

RT-qPCRs (Heinritzi et al., 2016)
NGS : samples with M gene RT-PCR Cq<30
Infographics



Veterinary Microbiology 195 (2016) 25–29

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Veterinary Microbiology

journal homepage: www.elsevier.com/locate/vetmic

Short communication

Circulation of multiple genotypes of H1N2 viruses in a swine farm in Italy over a two-month period

Maria Serena Beato[a,*], Luca Tassoni[a], Adelaide Milani[a], Annalisa Salviato[a], Guido Di Martino[a], Monica Mion[a], Lebana Bonfanti[a], Isabella Monne[a], Simon James Watson[b], Alice Fusaro[a]

[a] Istituto Zooprofilattico Sperimentale delle Venezie, Legnaro, PD, Italy
[b] Wellcome Trust Sanger Institute, Hinxton, UK

Detection of novel reassortants with Mpdm gene

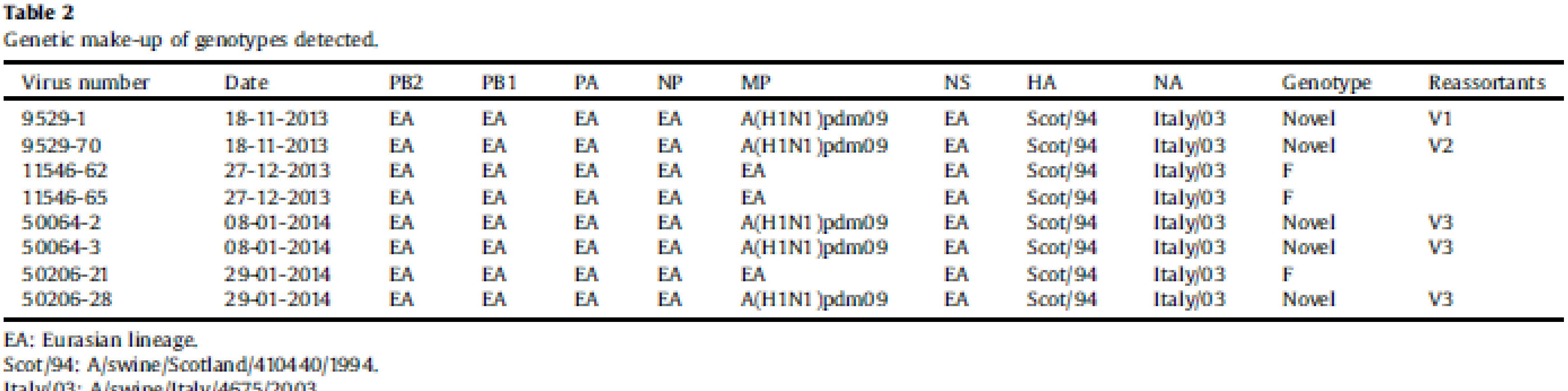


Table 2
Genetic make-up of genotypes detected.

Virus number	Date	PB2	PB1	PA	NP	MP	NS	HA	NA	Genotype	Reassortants
9529-1	18-11-2013	EA	EA	EA	EA	A(H1N1)pdm09	EA	Scot/94	Italy/03	Novel	V1
9529-70	18-11-2013	EA	EA	EA	EA	A(H1N1)pdm09	EA	Scot/94	Italy/03	Novel	V2
11546-62	27-12-2013	EA	EA	EA	EA	EA	EA	Scot/94	Italy/03	F	
11546-65	27-12-2013	EA	EA	EA	EA	EA	EA	Scot/94	Italy/03	F	
50064-2	08-01-2014	EA	EA	EA	EA	A(H1N1)pdm09	EA	Scot/94	Italy/03	Novel	V3
50064-3	08-01-2014	EA	EA	EA	EA	A(H1N1)pdm09	EA	Scot/94	Italy/03	Novel	V3
50206-21	29-01-2014	EA	EA	EA	EA	EA	EA	Scot/94	Italy/03	F	
50206-28	29-01-2014	EA	EA	EA	EA	A(H1N1)pdm09	EA	Scot/94	Italy/03	Novel	V3

EA: Eurasian lineage.
Scot/94: A/swine/Scotland/410440/1994.
Italy/03: A/swine/Italy/4675/2003.

Spain - UAB

2017-2018

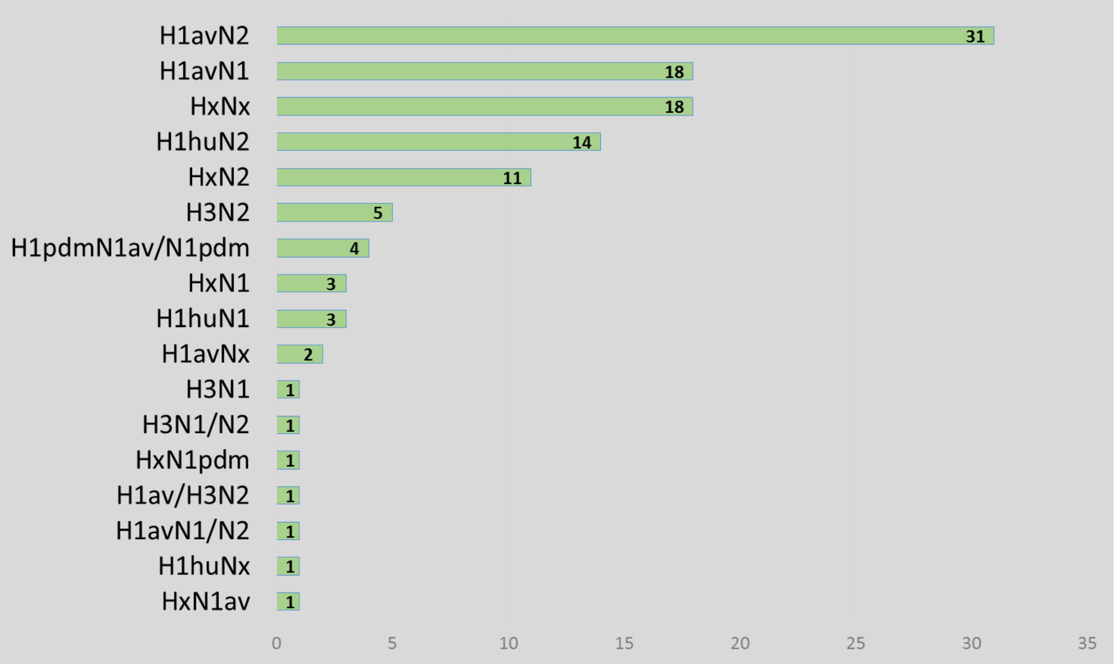
168 outbreaks > 107 positive
17 randomly selected farms
> 14 positive

Subtyping (RT-qPCR) of 116 positives cases

Isolation of 44 swIAV strains

NGS genotyping (Watson et al., 2015)

> 26 strains for the moment - ongoing



Genotype	Quantity
A	8
B	2
C	2
D	5
M	2
N	3
Not described ¿"X"?	4*

IDT Biologika

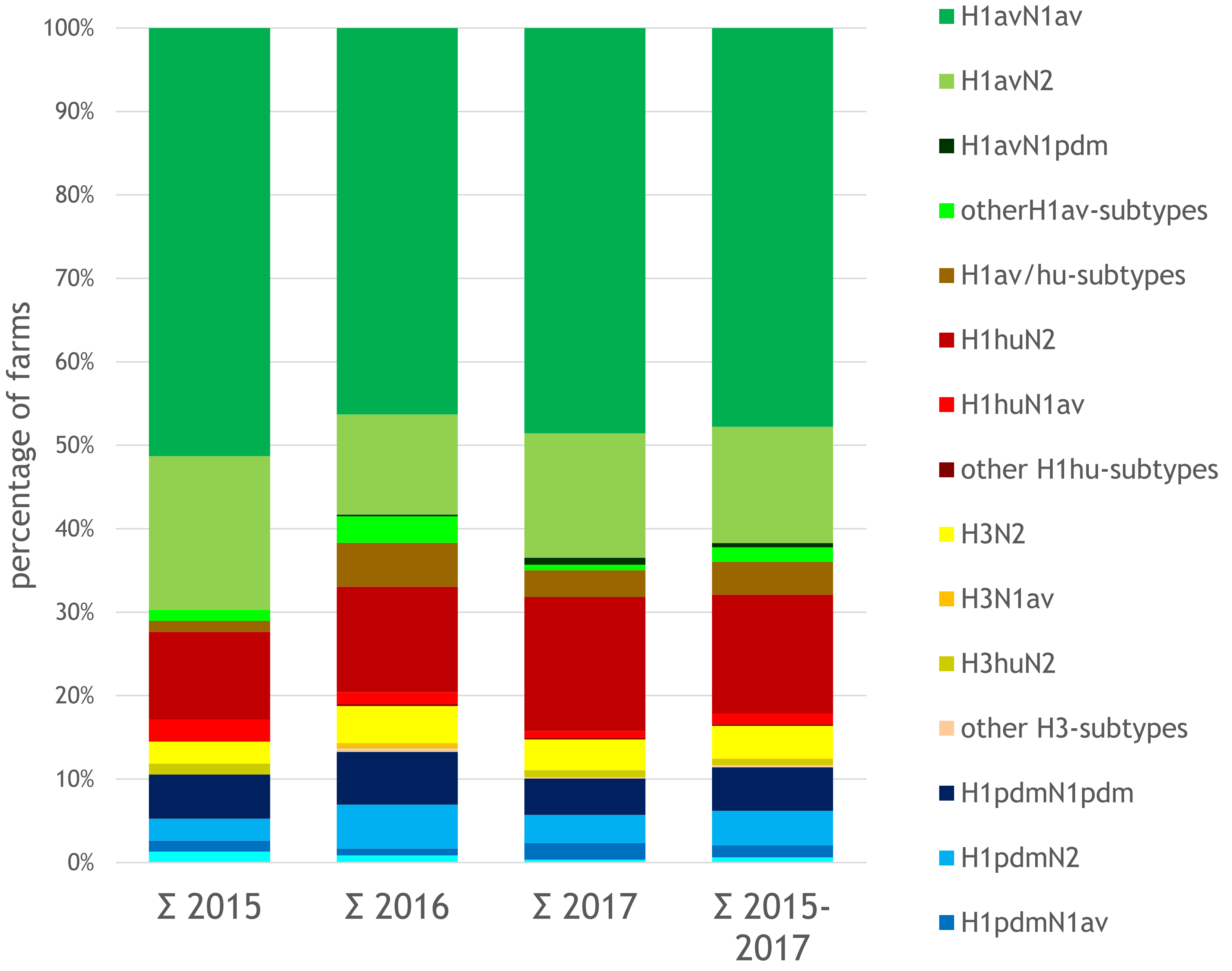
European surveillance program (1)

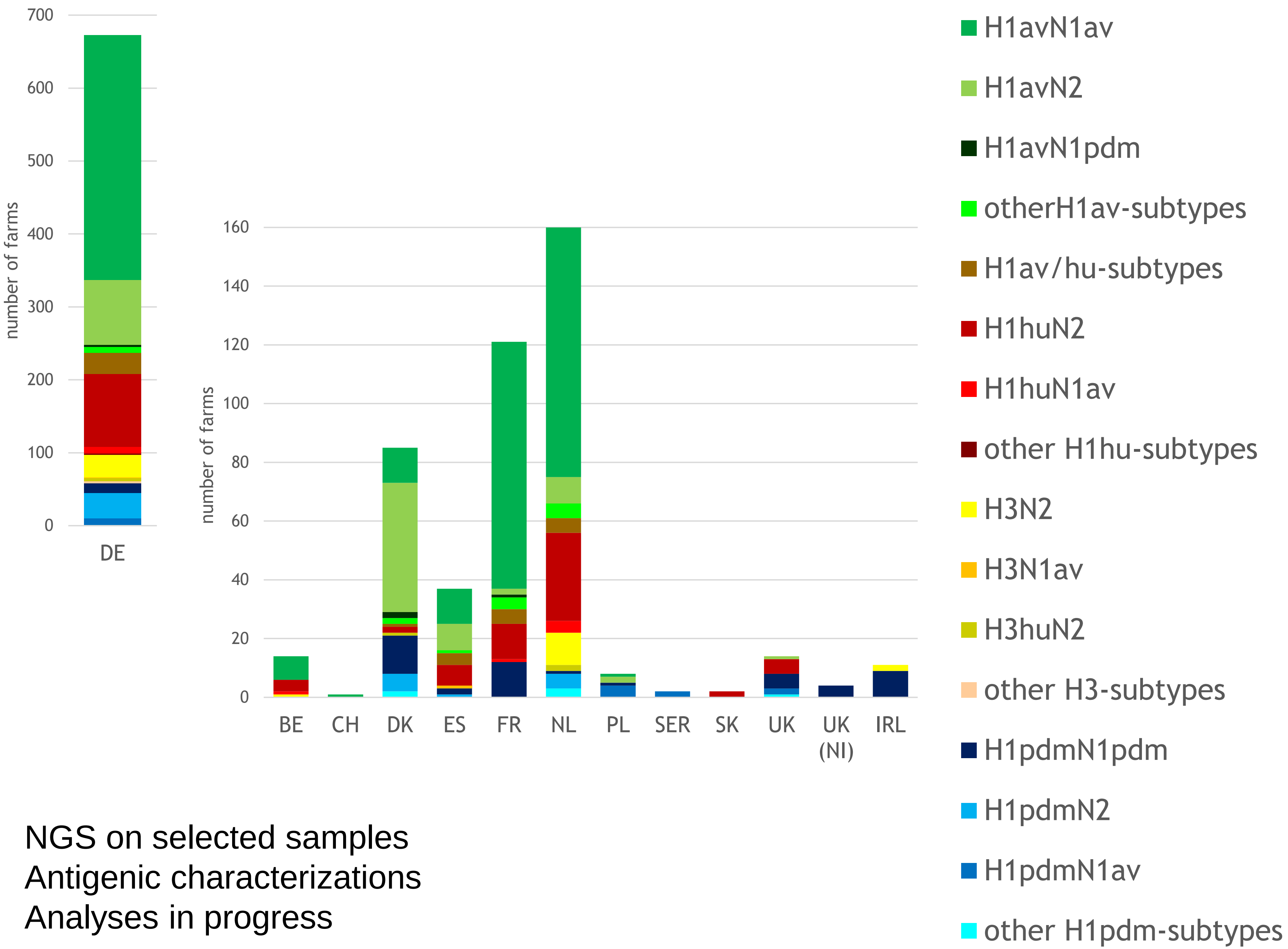
2015-2017

in coll. with FLI, Germany

Pigs sampled in 18 countries

Countries		Total number	positive	
			n	%
Σ	samples	18 216	5 522	30.31 %
	farms	**2 434**	**1 368**	**56.20 %**
Germany	samples	3 973	1 636	41.17 %
	farms	1 529	822	53.76 %
Netherlands	samples	4 468	1 357	30.37 %
	farms	270	184	68.14 %
France	samples	3 878	1 218	31.40 %
	farms	279	151	54.12 %
Denmark	samples	2 463	605	24.56 %
	farms	146	101	69.17 %
United-Kingdom	samples	811	113	13.93 %
	farms	49	17	34.69 %
Spain	samples	791	260	32.86 %
	farms	61	42	68.85 %
Poland	samples	524	67	12.78 %
	farms	13	6	46.15 %
Belgium	samples	410	103	25.12 %
	farms	35	15	42.85 %
Ireland	samples	394	99	25.12 %
	farms	24	15	62.50 %
Slovakia	samples	199	23	11.55 %
	farms	4	3	75.00 %
UK (North Ireland)	samples	146	27	18.49 %
	farms	10	6	60.00 %
SER	samples	39	4	10.25 %
	farms	3	2	66.66 %
Portugal	samples	33	6	18.18 %
	farms	3	2	66.66 %
Luxembourg	samples	25	2	8.00 %
	farms	1	1	100.00 %
Austria	samples	20	0	0.00 %
	farms	3	0	0.00 %
Sweden	samples	20	0	0.00 %
	farms	1	0	0.00 %
REU	samples	19	0	0.00 %
	farms	1	0	0.00 %
Switzerland	samples	3	2	66.66 %
	farms	1	1	100.00 %


100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
percentage of farms
Σ 2015
Σ 2016
Σ 2017
Σ 2015-2017
H1avN1av
H1avN2
H1avN1pdm
otherH1av-subtypes
H1av/hu-subtypes
H1huN2
H1huN1av
other H1hu-subtypes
H3N2
H3N1av
H3huN2
other H3-subtypes
H1pdmN1pdm
H1pdmN2
H1pdmN1av
other H1pdm-subtypes



NGS on selected samples
Antigenic characterizations
Analyses in progress

IDT Biologika - European surveillance program (2)

in coll. with IVD GmbH, Germany

Germany - IDT Biologika (IVD GmbH)

					Number of strains per lineage					
					H1N1		H3N2	H1N2		
Period	Number of herds investigated	Number of Influenza A positive cases	Frequency of positive cases (%)	Number of subtyped viruses*	$H1_{av}N1$	H1N1pdm	H3N2	$H1_{hu}N2$	$H1_{av}N2$	$H1_{pdm}N2$
2018	1463	804	59.96	667	358	26	55	114	77	37

Other European countries: data collection in progress

Greece

Zoonoses and Public Health

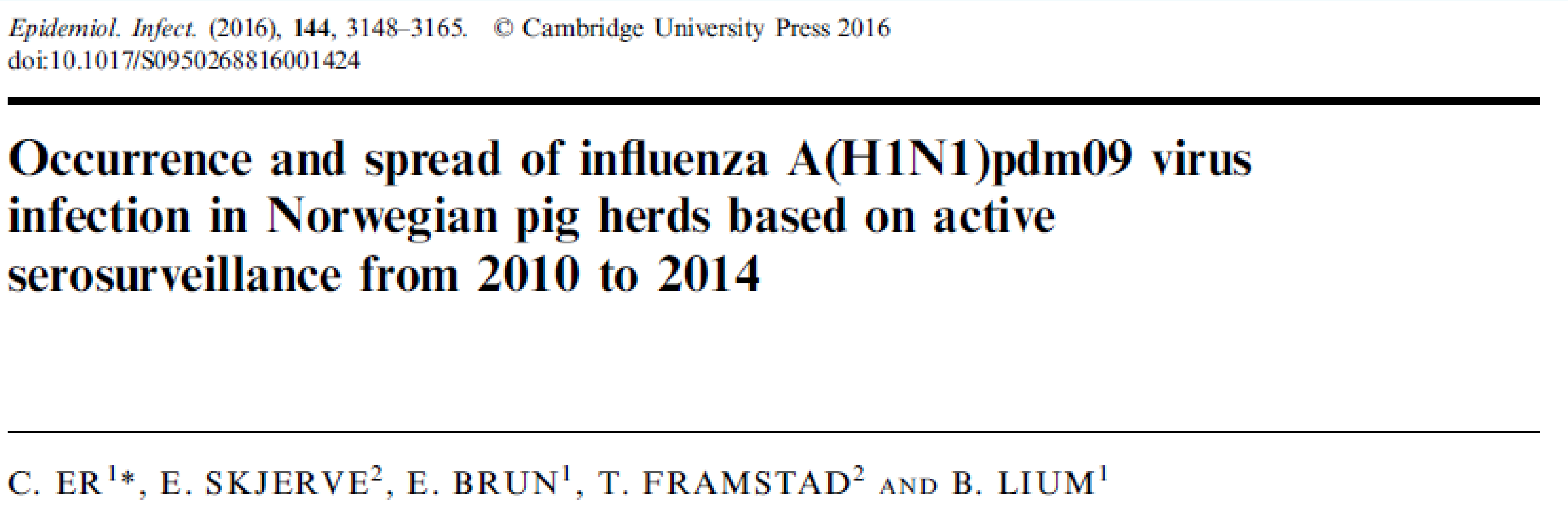
ORIGINAL ARTICLE

Serological Evidence of Pandemic H1N1 Influenza Virus Infections in Greek Swine

C. S. Kyriakis[1], V. G. Papatsiros[2], L. V. Athanasiou[2], G. Valiakos[1], I. H. Brown[3], G. Simon[4,5], K. Van Reeth[6], S. Tsiodras[7,8], V. Spyrou[9] and C. Billinis[1]

- Confirmed the presence of $H1_{av}$, $H1_{hu}$ and H3 swIAVs
- + 4% of pig sera had antibodies against the H1pdm only

Norway



Epidemiol. Infect. (2016), **144**, 3148–3165. © Cambridge University Press 2016
doi:10.1017/S0950268816001424

Occurrence and spread of influenza A(H1N1)pdm09 virus infection in Norwegian pig herds based on active serosurveillance from 2010 to 2014

C. ER[1]*, E. SKJERVE[2], E. BRUN[1], T. FRAMSTAD[2] AND B. LIUM[1]

Prevalence of H1N1pdm seropositive herds has not fallen under 40%
> adaptation to the pig population

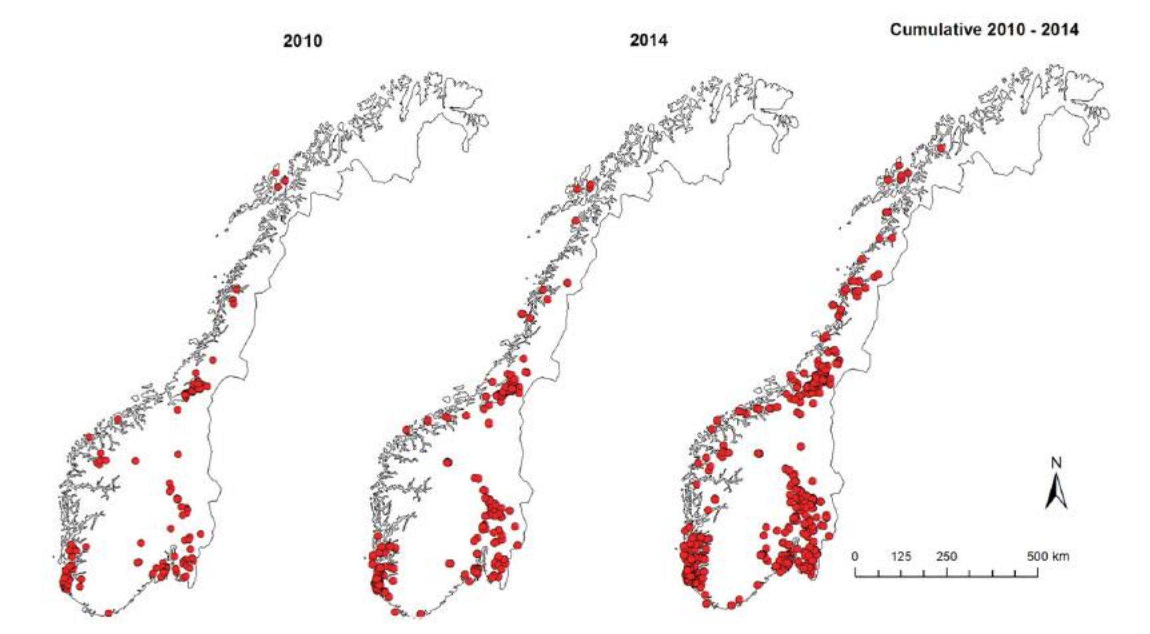


Fig. 3. Spatial distribution of Norwegian pig herds testing positive for antibodies against influenza A(H1N1)pdm09 virus in 2010 [41%, 95% confidence interval (CI) 37–45 seroprevalence]; 2014 (48%, 95% CI 45–51 herd seroprevalence); cumulative 2010–2014 (53%, 95% CI 50–56 herd seroprevalence).

Conclusion

- **Four enzootic viruses co-circulating across Europe**: $H1_{av}N1$, H3N2, $H1_{hu}N2$, H1N1pdm
- **Increasing frequency at which viral diversification occurs** in this viral reservoir
- Antigenic shifts (reassortments) + Antigenic drifts (immune escape, host adaptation)
- **Increasing frequencies of (multi) reassortant viruses** containing ≥1 gene from H1N1pdm
- Many "sporadic" (?) viruses
- **Emergence of a swine-divergent H1N1pdm genogroup** (*versus* seasonal-like H1N1pdm)
- **Frequent introductions of HA or NA genes from seasonal human IAVs** (H1N1pdm, H3N2)
- **Genotypes of public-health interest** (ex: reassortant IGC with Mpdm)
- **Significant regional variations**

  > Novel enzootic viruses in some countries (ex: UK, Germany, DK)

  > Disappearance of "old" enzootic viruses in some regions

No significant modifications in the global European picture since ESNIP3 but **a still increasing complexity of European swIAVs**

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