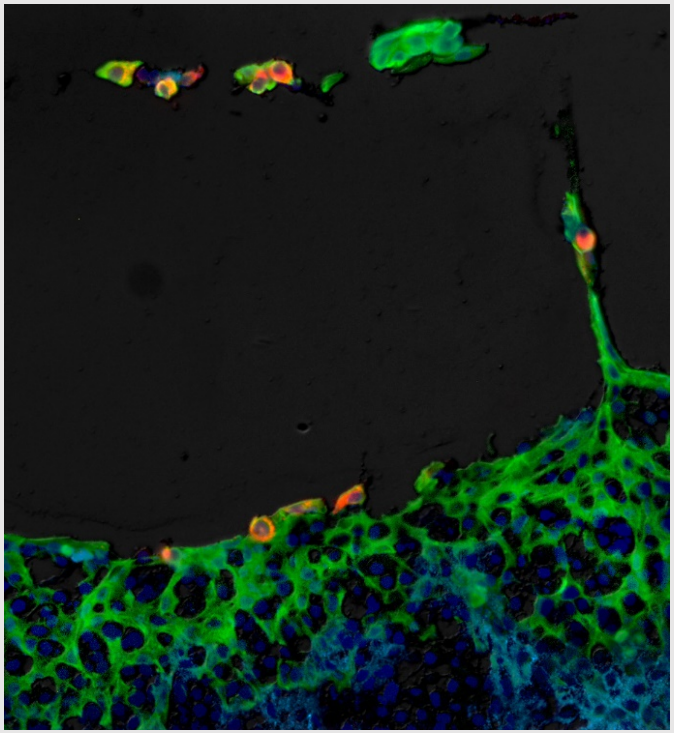
The role of asymptomatic carriers in FMD ecology; unifying knowledge from controlled laboratory experiments and field studies

Jonathan Arzt, Luis Rodriguez, Barbara Brito, Carolina Stenfeldt

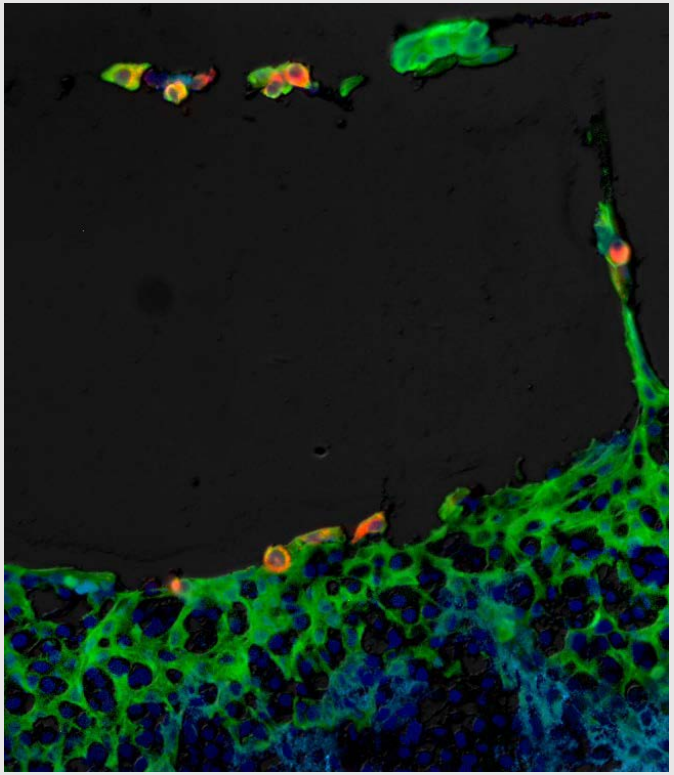


Jonathan Arzt
Veterinary Medical Officer
Foreign Animal Disease Research Unit
ARS, USDA
Plum Island Animal Disease Center

eofmd

USDA

field studies



laboratory experiments

The FMDV Carrier State; early landmark papers

- ***Van Bekkum et al., 1959***
  - Recovery of infectious FMDV in OPF (saliva) up to 8 months after recovery from clinical FMD
  - Similar subclinical persistence found in vaccinated cattle
  - Reports that FMDV carriers do not transmit infection further
- ***Sutmoller and Gaggero, 1965***
  - Standardization of probang sampling technique
    - Although, similar approach as used by van Bekkum
- ***Burrows 1966***
  - Isolation of infectious FMDV from nasopharyngeal tissues of persistently infected cattle
- ***Sutmoller et al 1968***
  - Definition of carriers at 28dpi

First description of carrier state;

Van Bekkum et al., 1959

Observations on the carrier state of cattle exposed to foot-and-mouth disease virus

by

J. G. VAN BEKKUM, H. S. FRENKEL, H. H. J. FREDERIKS and S. FRENKEL.

(Centraal Diergeneeskundig Instituut, Afd. Amsterdam)

Tijdschrift Voor Diergeneeskunde, Aflevering 20, Deel 84, 15 Okt. 1959.

At the XVIth Veterinary Congress held in Madrid in 1959, Schang gave as his opinion, that virus carriers are of no importance in the Argentine. According to Fogedby a comparable situation exists in Scandinavia. The available evidence at most seems to warrant the conclusion, that only a small proportion of recovered individuals may excrete the agent (Olitsky c.s., 1928).

In 1955 Dijkstra, basing his opinion on an epizootiologic study of the disease in the Dutch province of Friesland, reached the conclusion, that in cattle either virus excretion is not continuous, or the amount of virus liberated is so small,

Experimental studies

Advantages

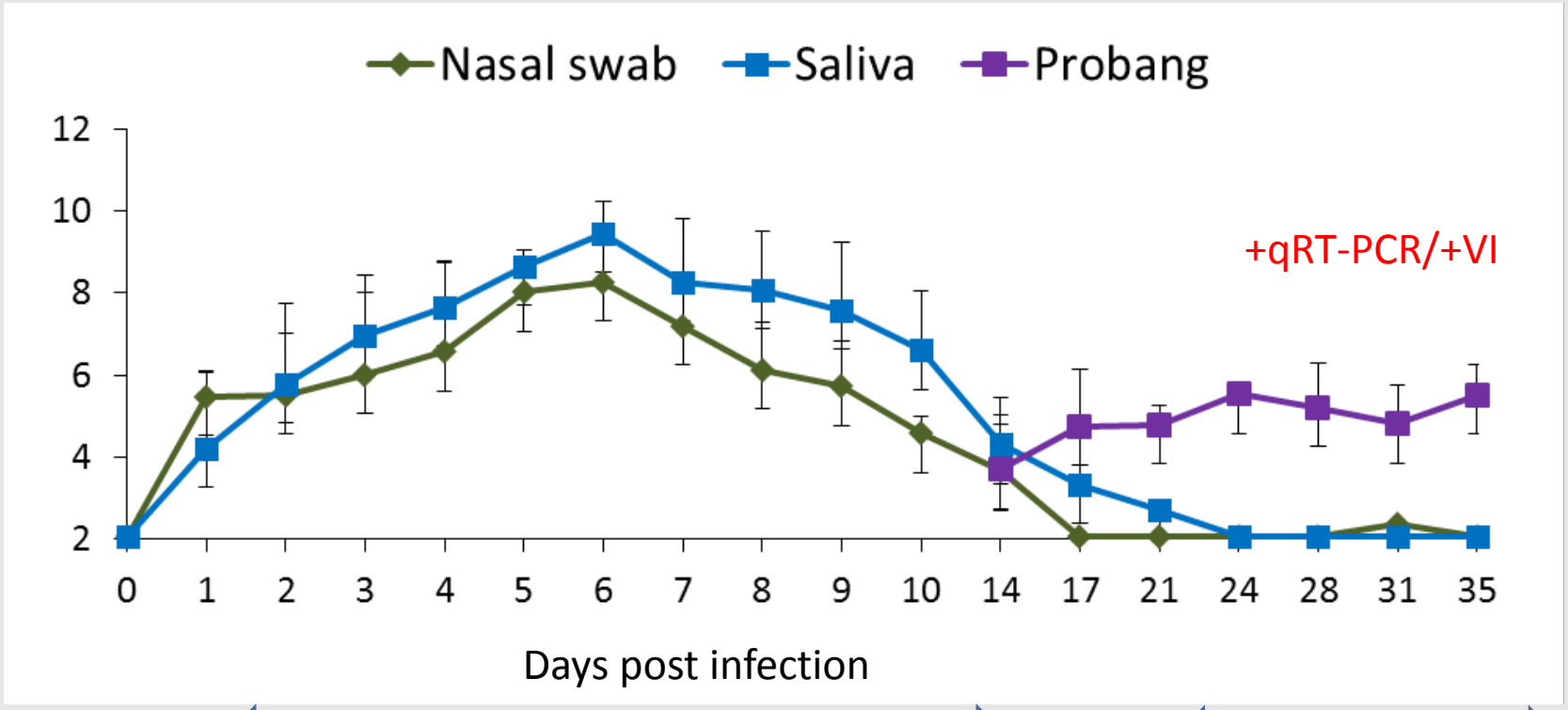
- Prospective study design
- Controlled environment
  - Known virus and exposure conditions
  - Strategic & intensive sampling
  - Possible to reduce/control confounding variables
    - No delays in sample processing
    - Reduced operator variability
- Provides detailed knowledge of FMDV pathogenesis

Limitations

Detecting and defining carriers

FMDV "'shedding'" in oral/nasal swabs and probangs

Non-Vaccinated cattle



Clinical FMD

FMDV persistence

Stenfeldt et al 2016

Sutmoller et al 1968

Plum Island Animal Disease Laboratory, Animal Disease and Parasite Research Division, Agricultural Research Service, U.S. Department of Agriculture, Greenport, Long Island, N.Y., U.S.A.

The Epizootiological Importance of Foot-and-Mouth Disease Carriers

I. Experimentally Produced Foot-and-Mouth Disease Carriers in Susceptible and Immune Cattle

By

Paul Sutmoller, John W. McVicar, and **George E. Cottral**

With 1 Figure

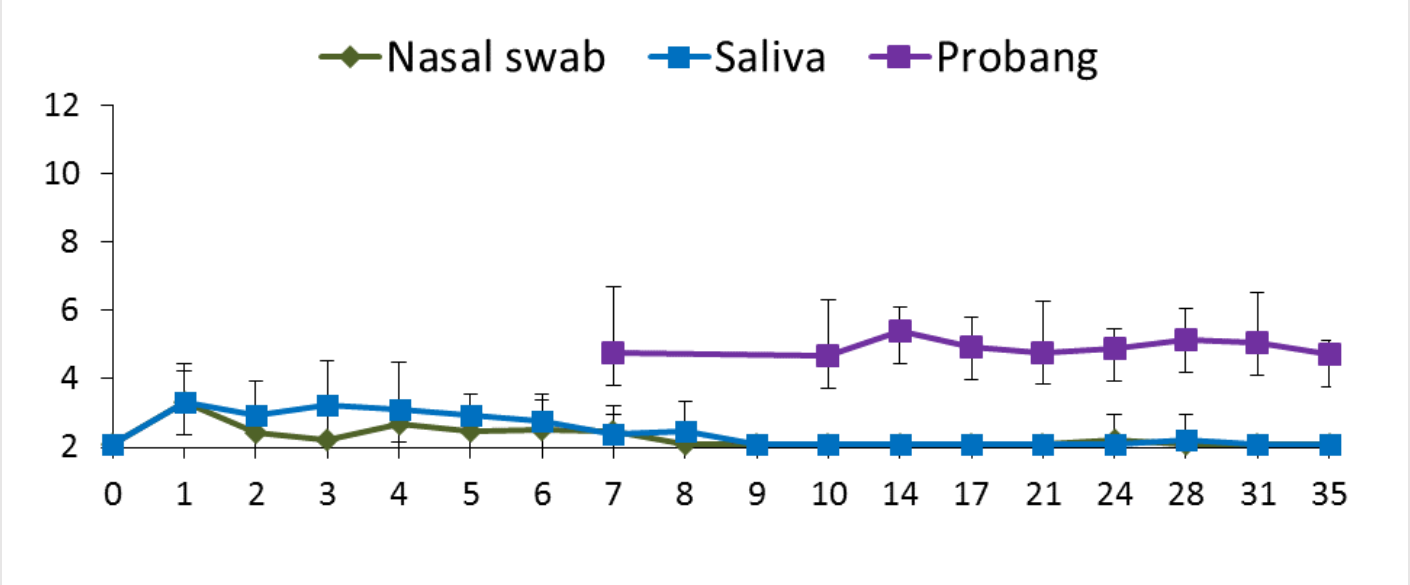
(Received September 9, 1967)

Cattle examination. The data reported are limited to cattle observed for 4 weeks following inoculation. The term virus "carrier" is applied to cattle in which virus was detectable to the end of that period. For a period of 14 DPI, the cattle were examined daily for clinical signs of FMD. Most OP fluid specimens were collected and tested (16) at weekly intervals. However, from each of groups III and IV, 8 cattle were tested only at 28 DPI.

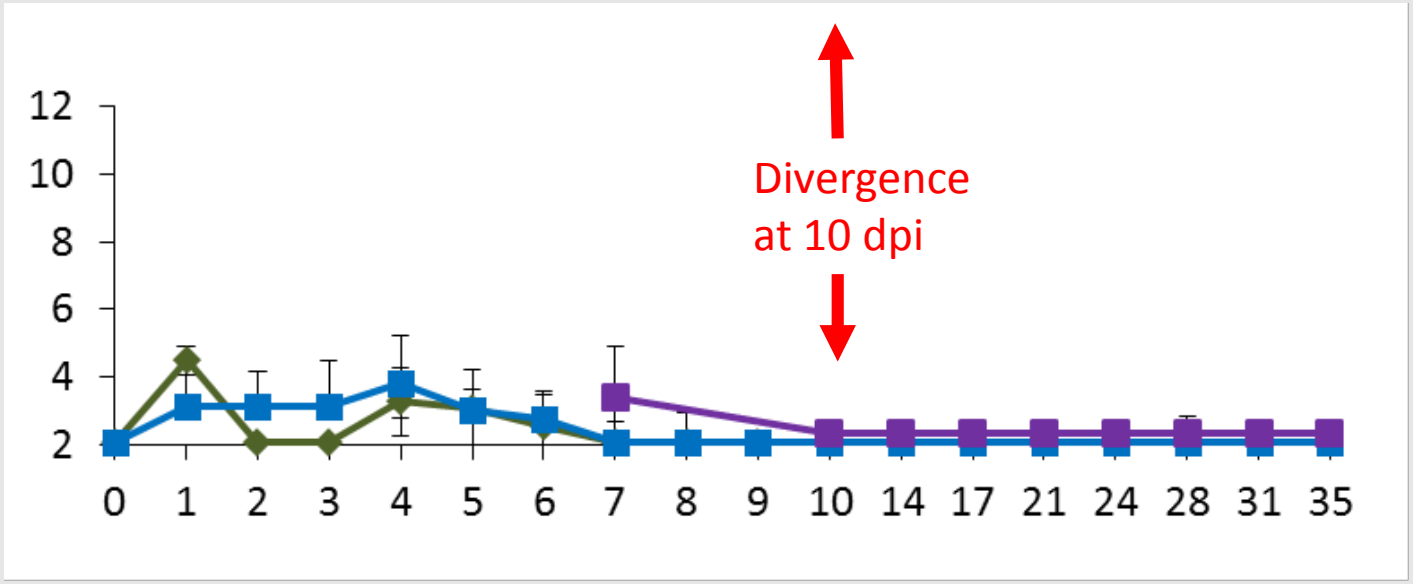
FMDV persistence definition; from laboratory data

Vaccinated cattle

Carriers

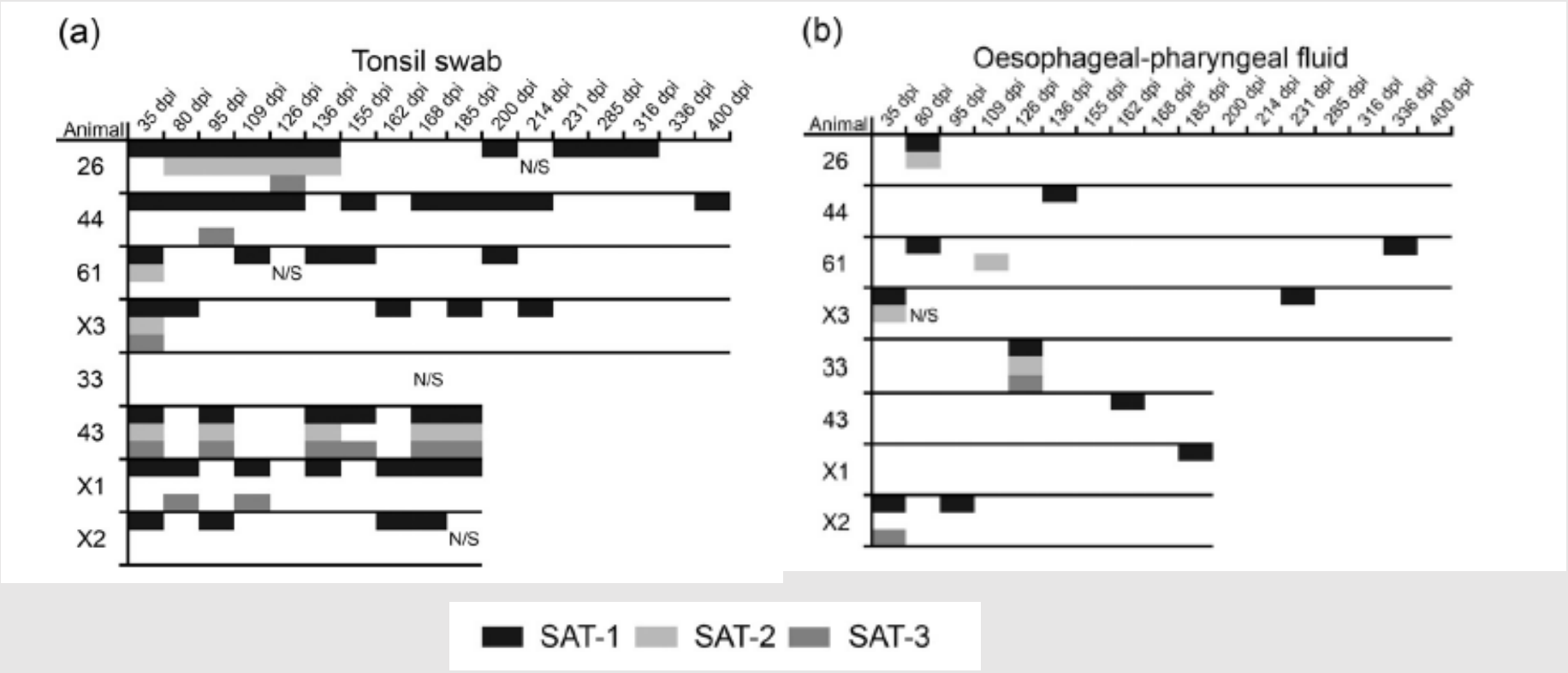


Non-Carriers

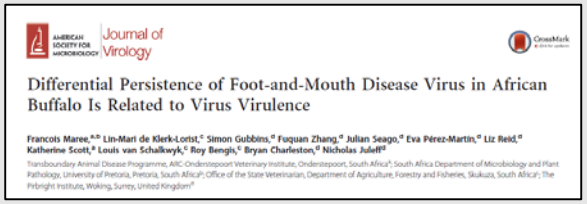


Stenfeldt et al 2016

FMDV shedding in tonsil swabs and probangs



Maree et al '16



Experimental studies

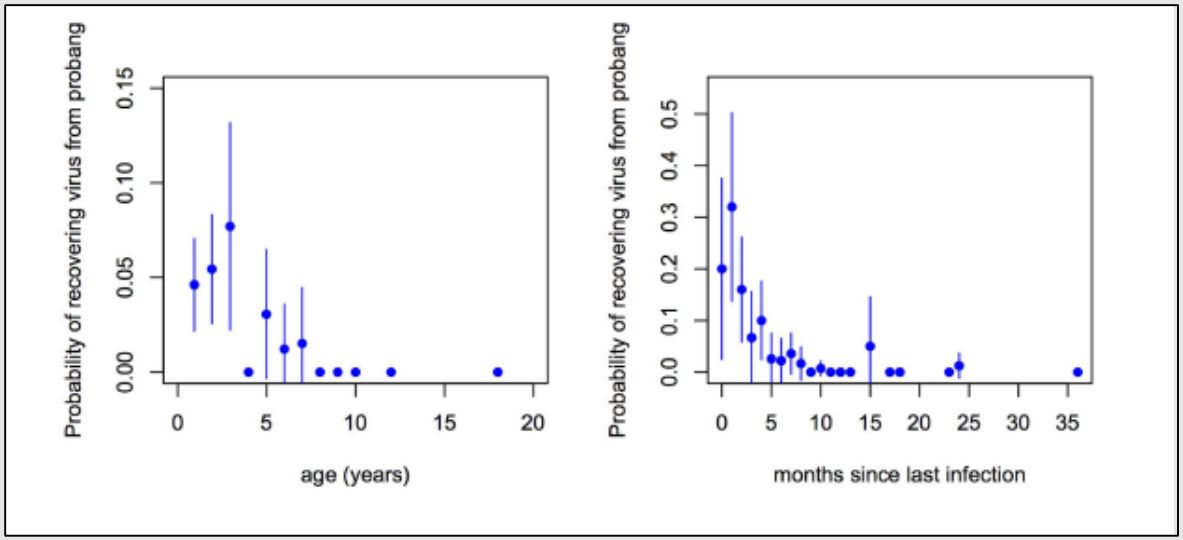
Limitations

- Experimental capacity (practical burden)
  - Restrictions in animal numbers and duration of investigations
    - Logistics and Animal Welfare concerns
- Questionable applicability to field situation
    - Virus used
    - Environmental conditions
    - Findings limited to experimental virus strain/host species
    - Feed
    - husbandry

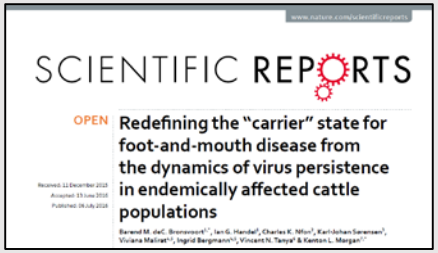
FMDV persistence definition; from modeling field data

Predictive statistical models for identifying persistently infected cattle:

$$Prob(probang+'ve) = invlogit(-2.91 - 0.32 \times monlast + 0.22 \times age - 0.14 \times age^2 + 2.07 \times VNTany + 0.04 \times age \times monlast)$$



Bronsvoort et al


SCIENTIFIC REPORTS

OPEN

Redefining the "carrier" state for foot-and-mouth disease from the dynamics of virus persistence in endemically affected cattle populations

Field studies

Advantages

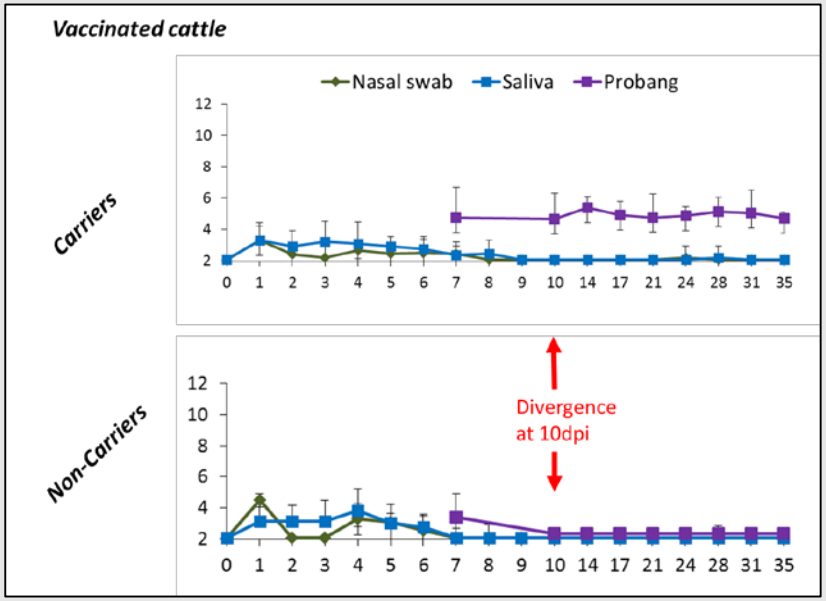
- Scale, scope, and duration unlimited
- Undeniable link to reality
- Enables association of specific findings and known events
  - Risk analysis based on actual
- Spatiotemporal relationships of viruses and outbreaks
  - Retrospective estimation of transmission pathways
- Multi-factorial analysis → FMD Epidemiology **&** Ecology

Limitations

- Unknown (uncontrolled) background context
- Limited information available
- Outcome and conclusions specific to geographic region investigated
- Intrinsic technical challenges(cold chain, transport)
- Limited capability (no daily sampling, operator variability)
- Breadth of relevance difficult to ascertain

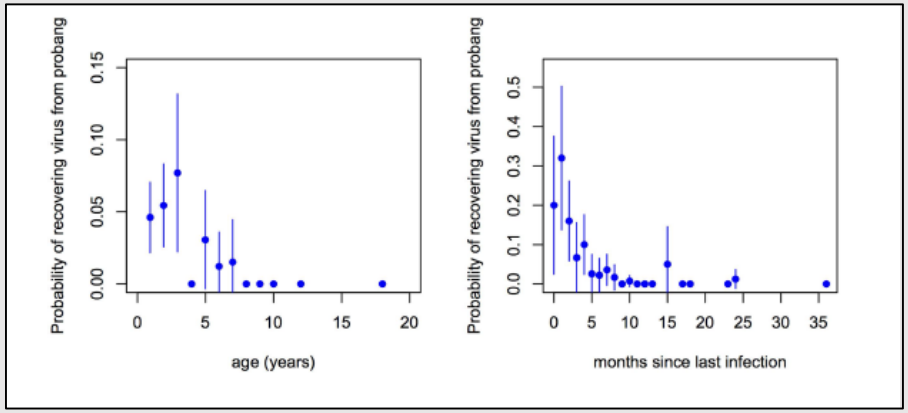
2 new criteria to characterize the carrier state

Unified



Stenfeldt et al 2016

$$Prob(probang{+}'ve) = invlogit(-2.91 - 0.32 \times monlast + 0.22 \times age - 0.14 \times age^2 + 2.07 \times VNTany + 0.04 \times age \times monlast)$$

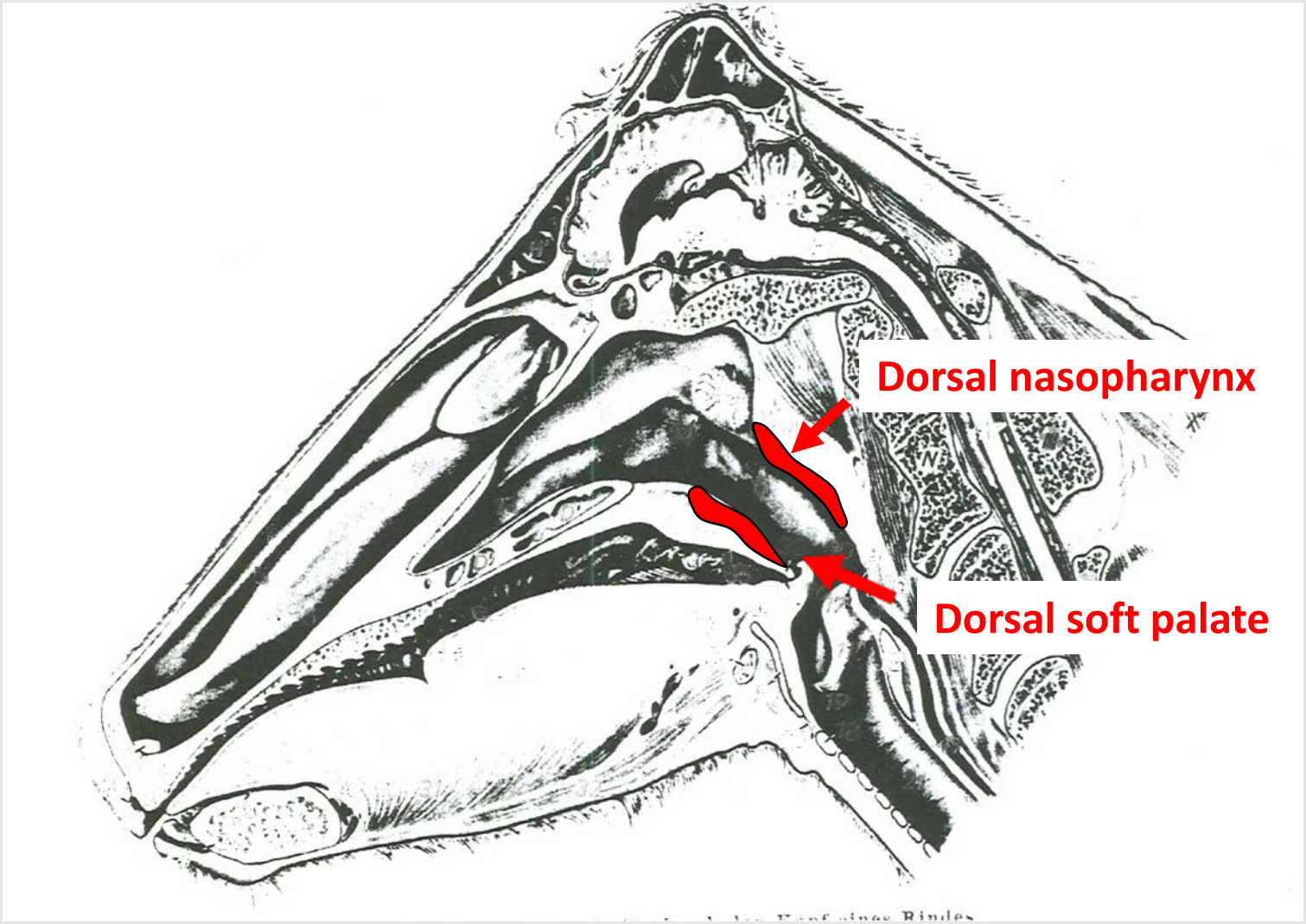


Bronsvoort et al

Comprehensive

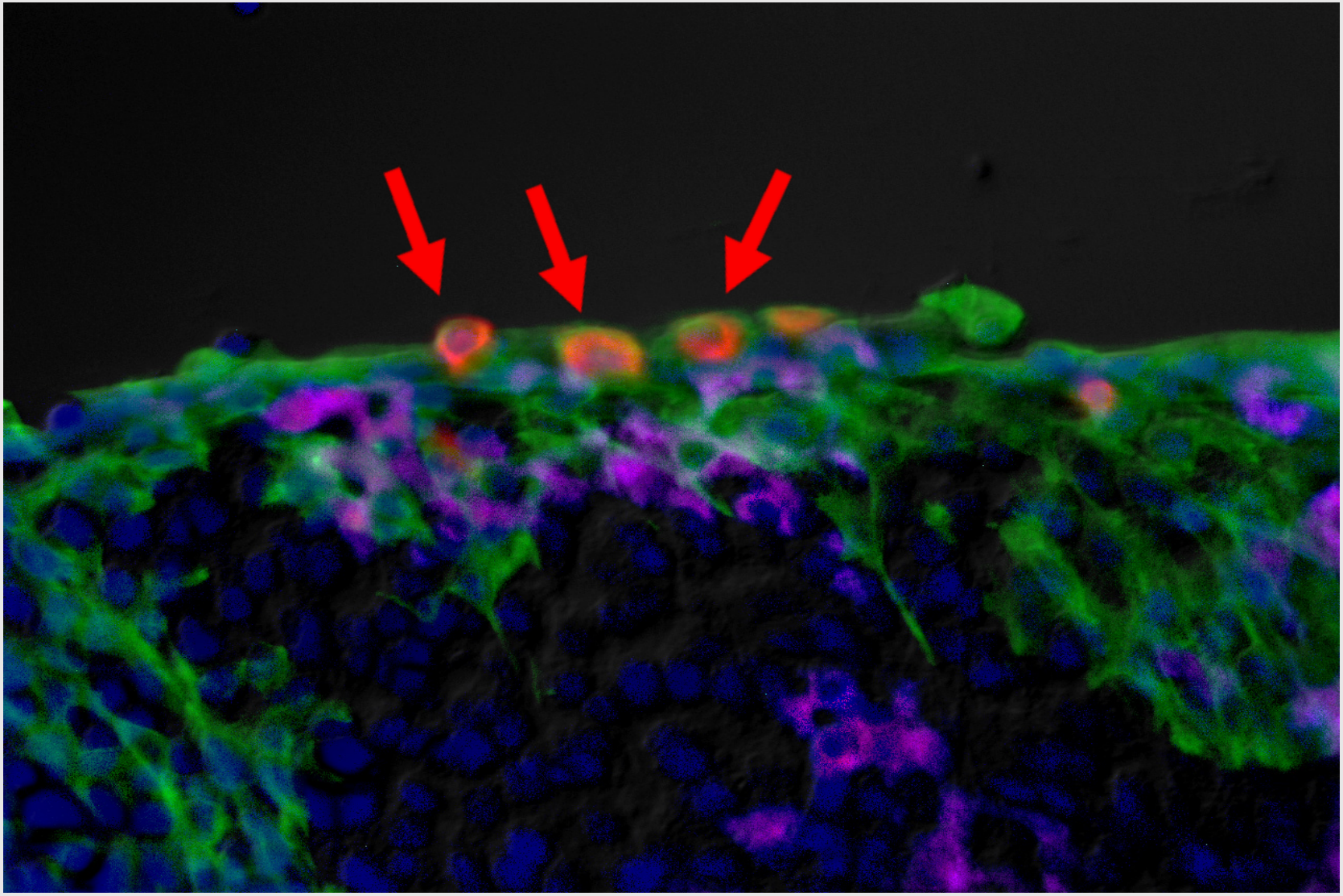
Sites of infection in carriers

Anatomic localization of persistent FMDV: Nasopharynx



Not the oropharynx.

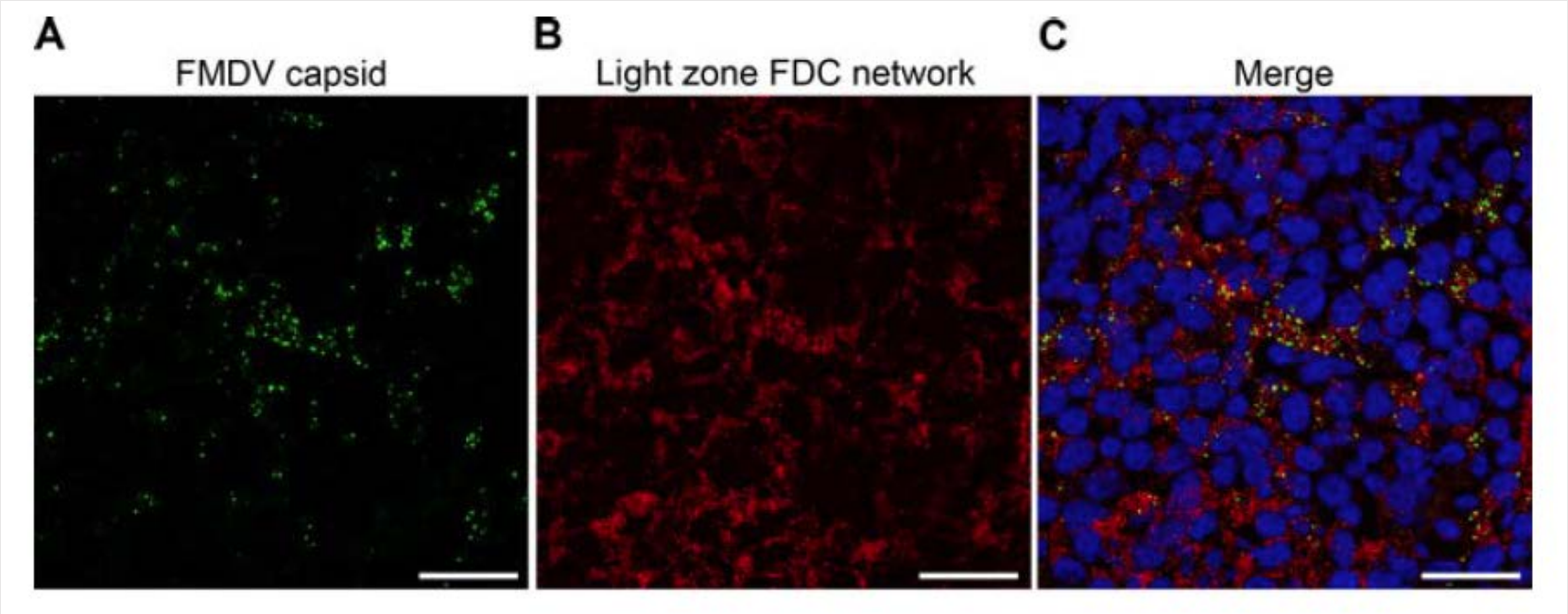
Anatomic localization of persistent FMDV in the bovine nasopharyngeal mucosa



Cytokeratin, FMDV VP1, FMDV 3D, MHC II

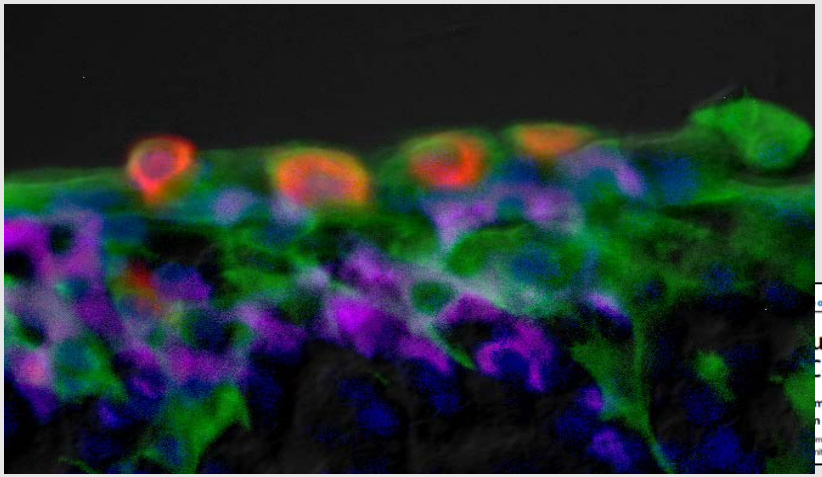
Stenfeldt et al 2016

Anatomic localization of persistent FMDV



Submandibular LN, 38 dpc

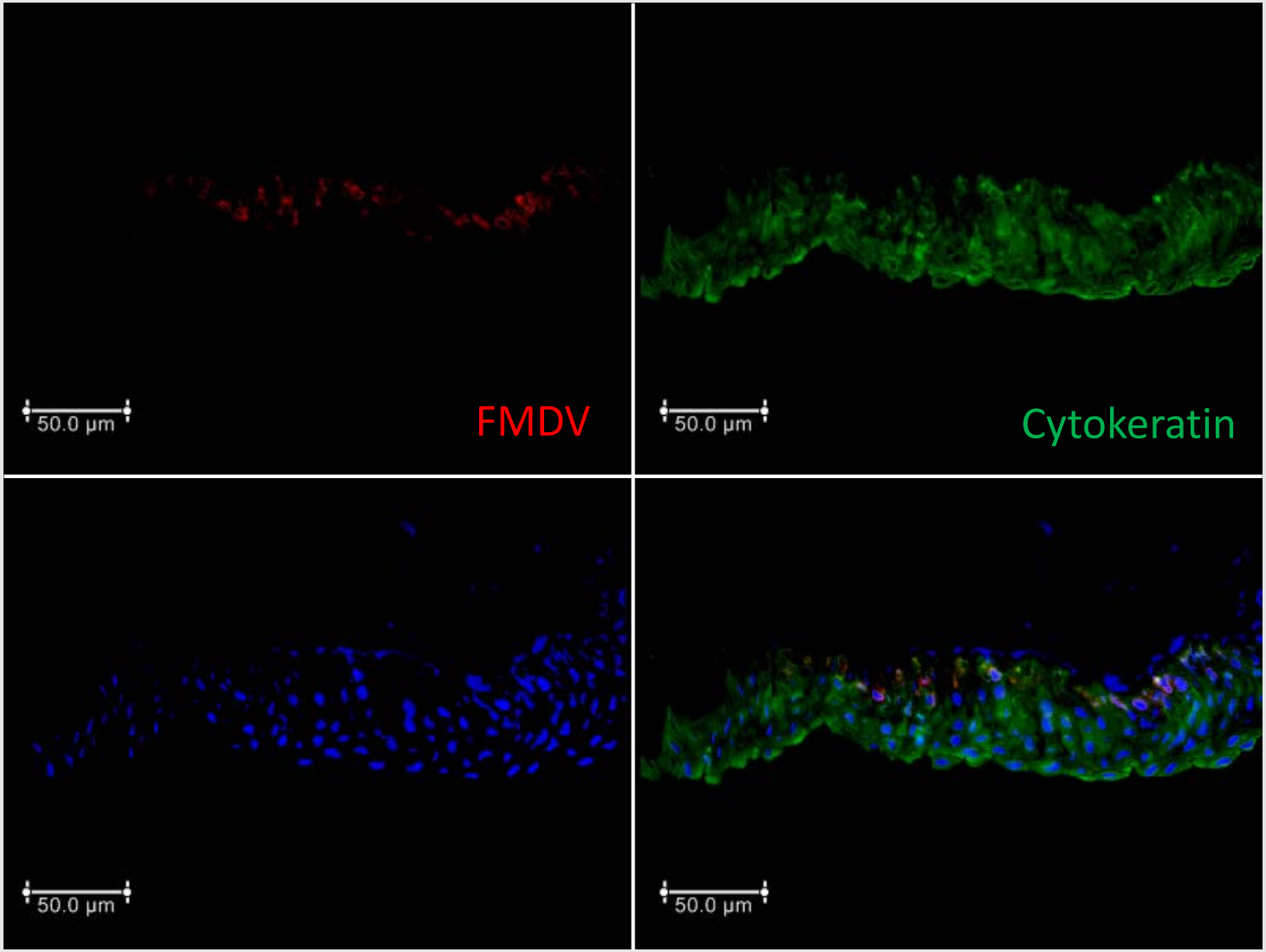
Not mutually exclusive findings.



Juleff et al., Plos ONE (2008)

Anatomic confirmation from the field;

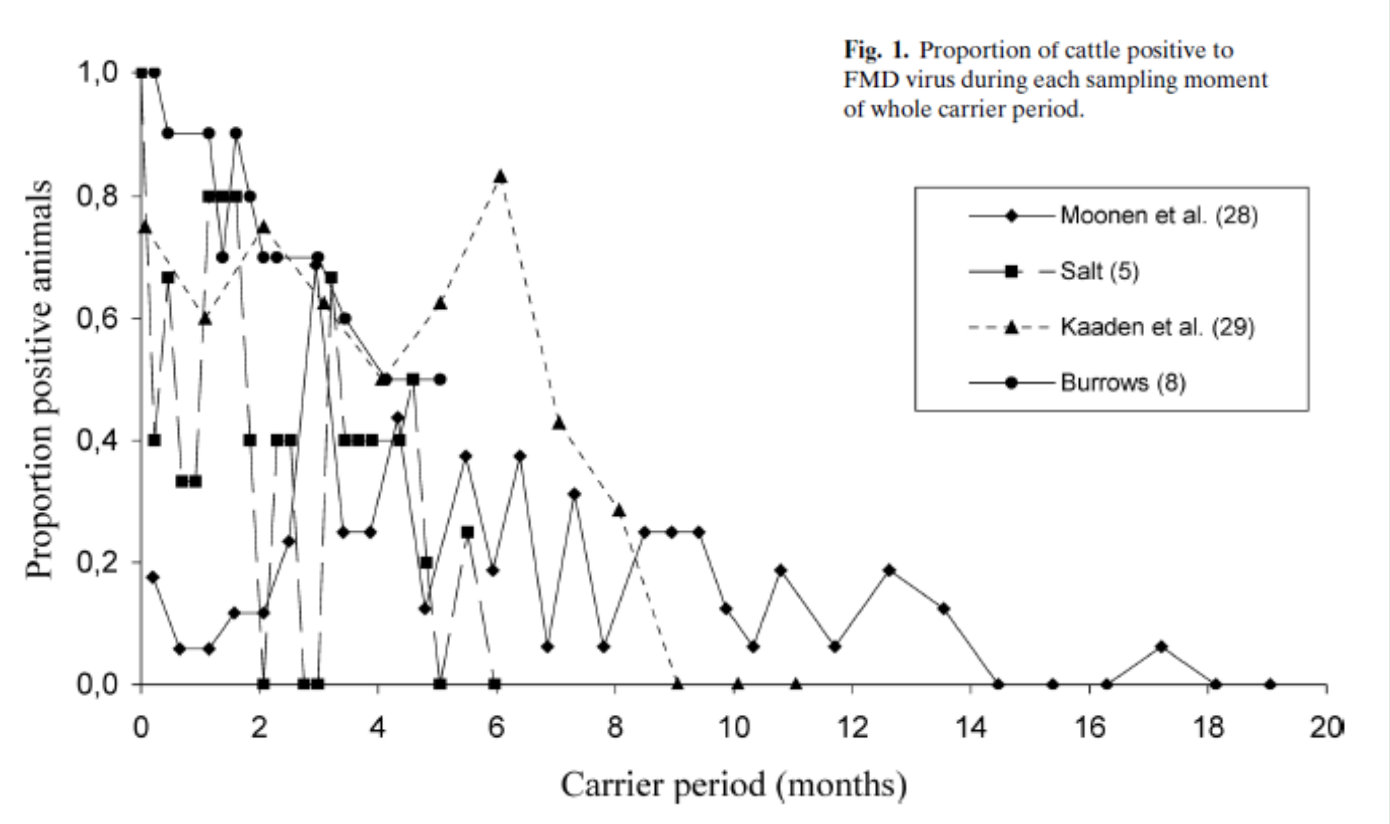
Nasopharynx of Asian buffalo



Ranjan et al, in progress

Duration of carrier state

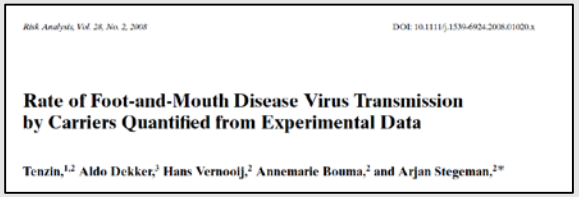
Duration of FMDV persistence (extinction)



Meta-analysis of published experimental studies

Decrease in proportion positive animals: 0.11 per carrier month

Tenzin et al., 2008

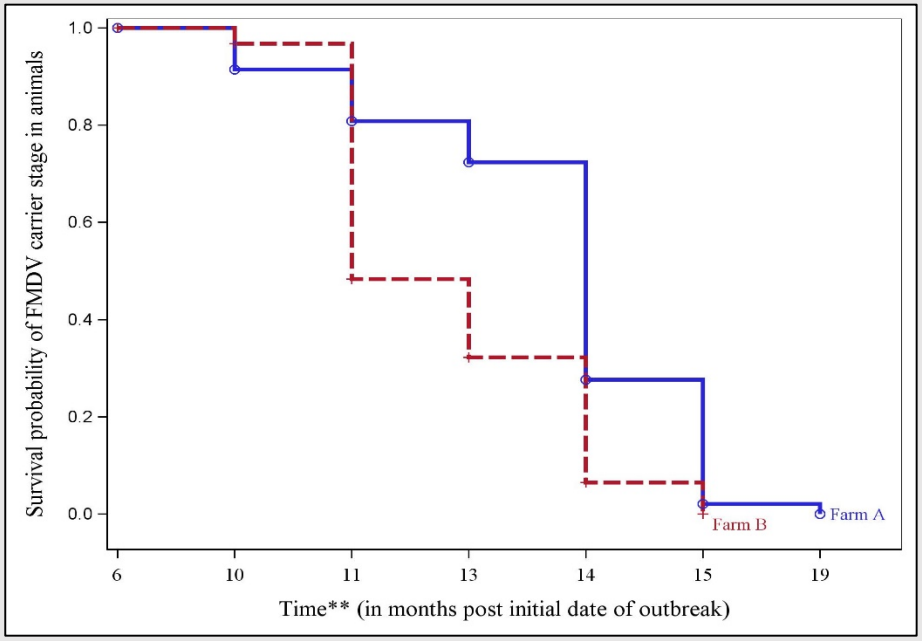

Risk Analysis, Vol. 28, No. 2, 2008 DOI: 10.1111/j.1539-6924.2008.01020.x

Rate of Foot-and-Mouth Disease Virus Transmission by Carriers Quantified from Experimental Data

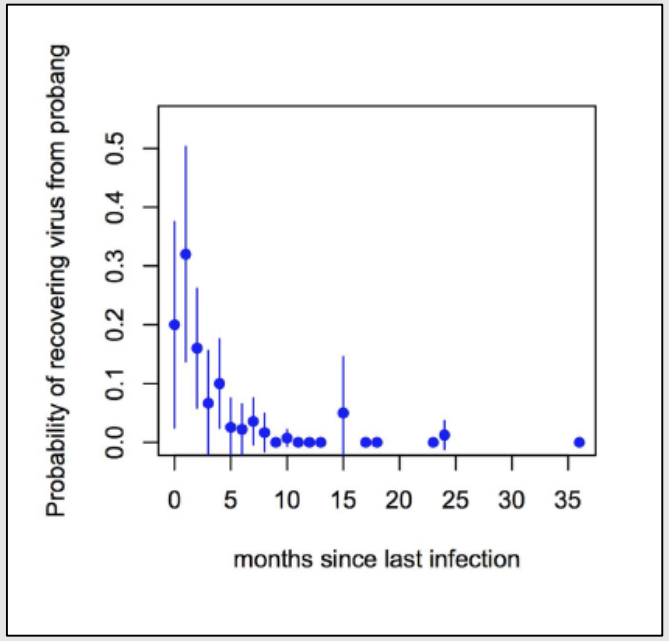
Tenzin,[1,2] Aldo Dekker,[3] Hans Vernooij,[2] Annemarie Bouma,[2] and Arjan Stegeman,[2*]

Duration of FMDV persistence: field confirmation

India (field-based)



Cameroon (field-based)



Hayer et al., Duration of FMDV carrier state based on detection of FMDV RNA in probang samples from two farms in Uttarakhand, India

Bronswoort et al., Unlikely to detect probang-positive animals (by VI) >12 months post-outbreak

Viral genomic changes during the carrier state

Experimental studies

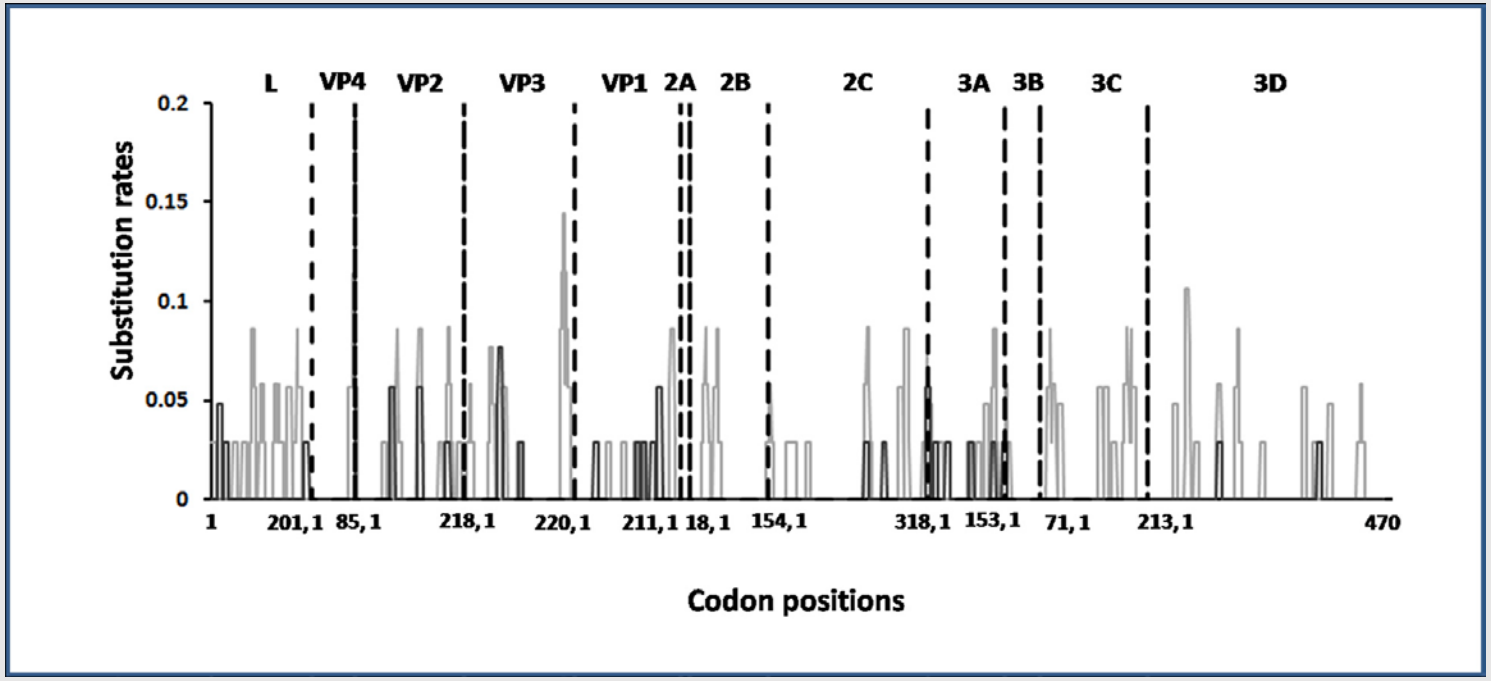
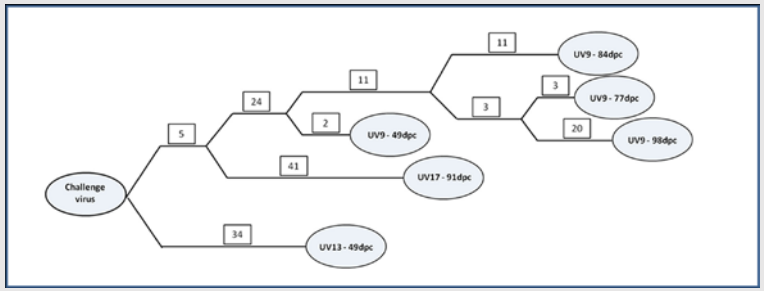
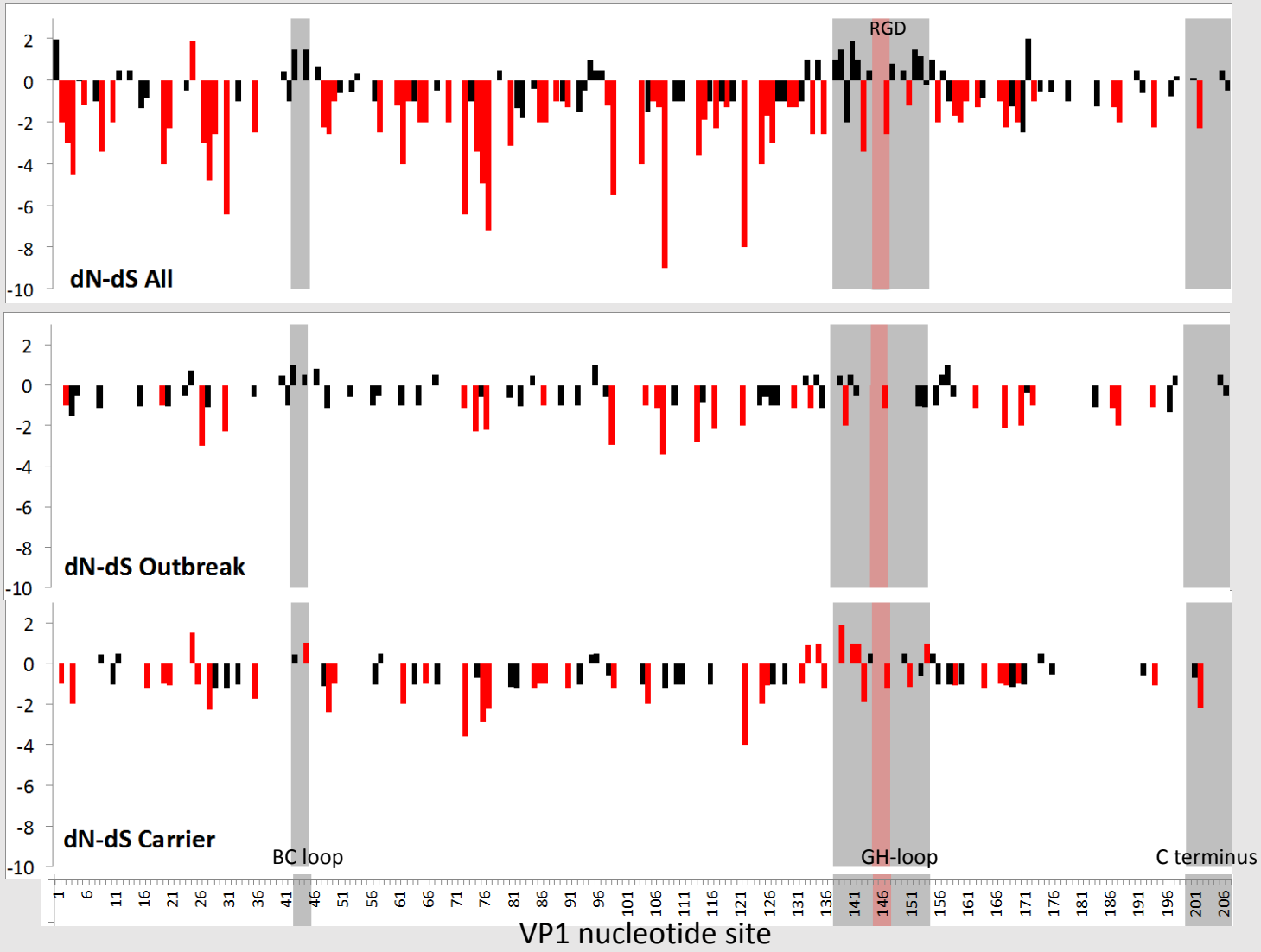


Fig 2. Synonymous (grey) and non-synonymous (black) changes in the ORF of the genomes of the FMD viruses obtained from carrier animals.

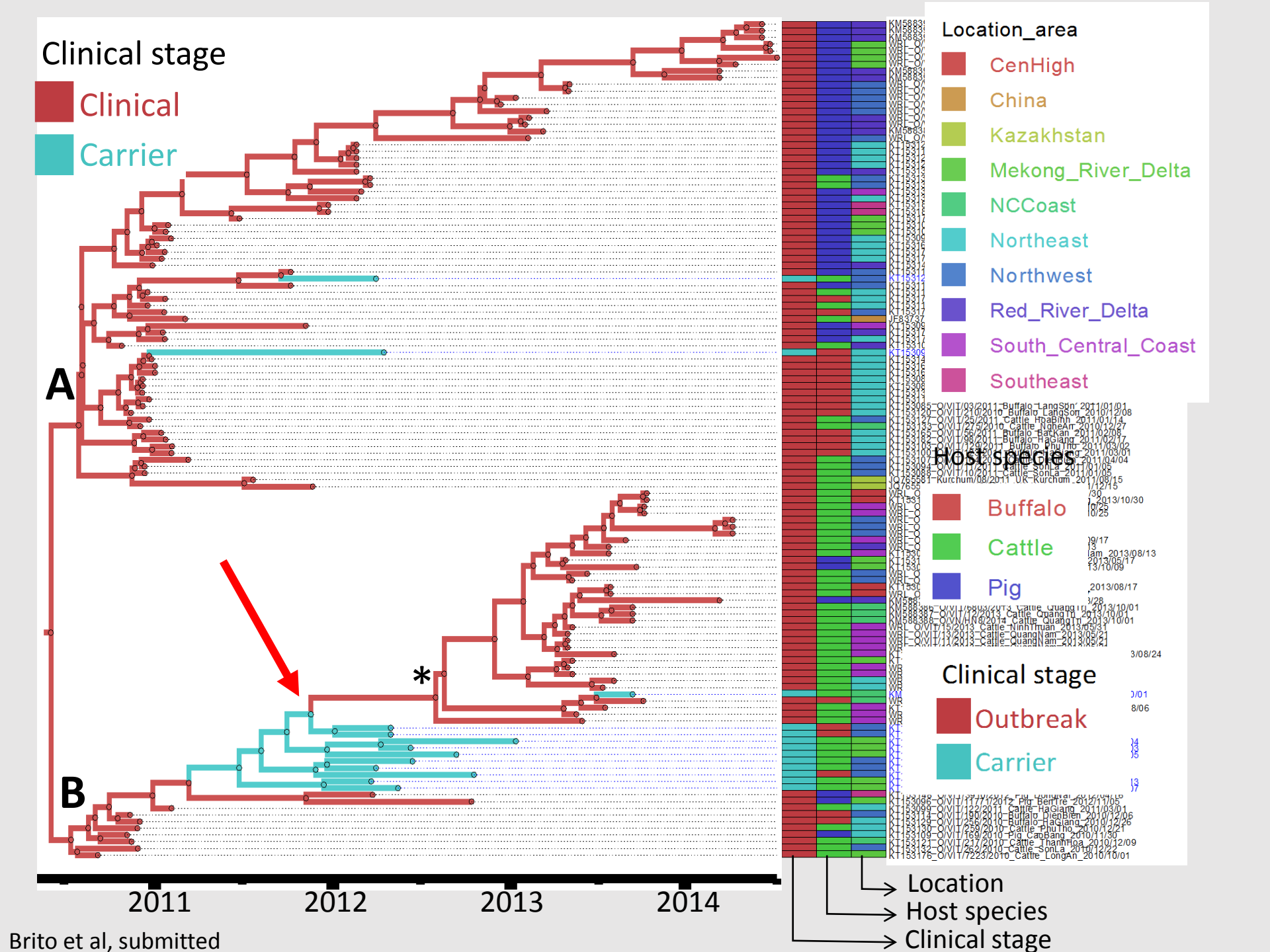


Parthiban et al., 2015

Genomic variation; carriers from the field Vietnam



Brito et al, submitted



Brito et al, submitted

Transmission during the carrier state

Transmission from FMDV carriers

Table II. List of Experiments Used for the Estimation of Transmission Rate Parameter β

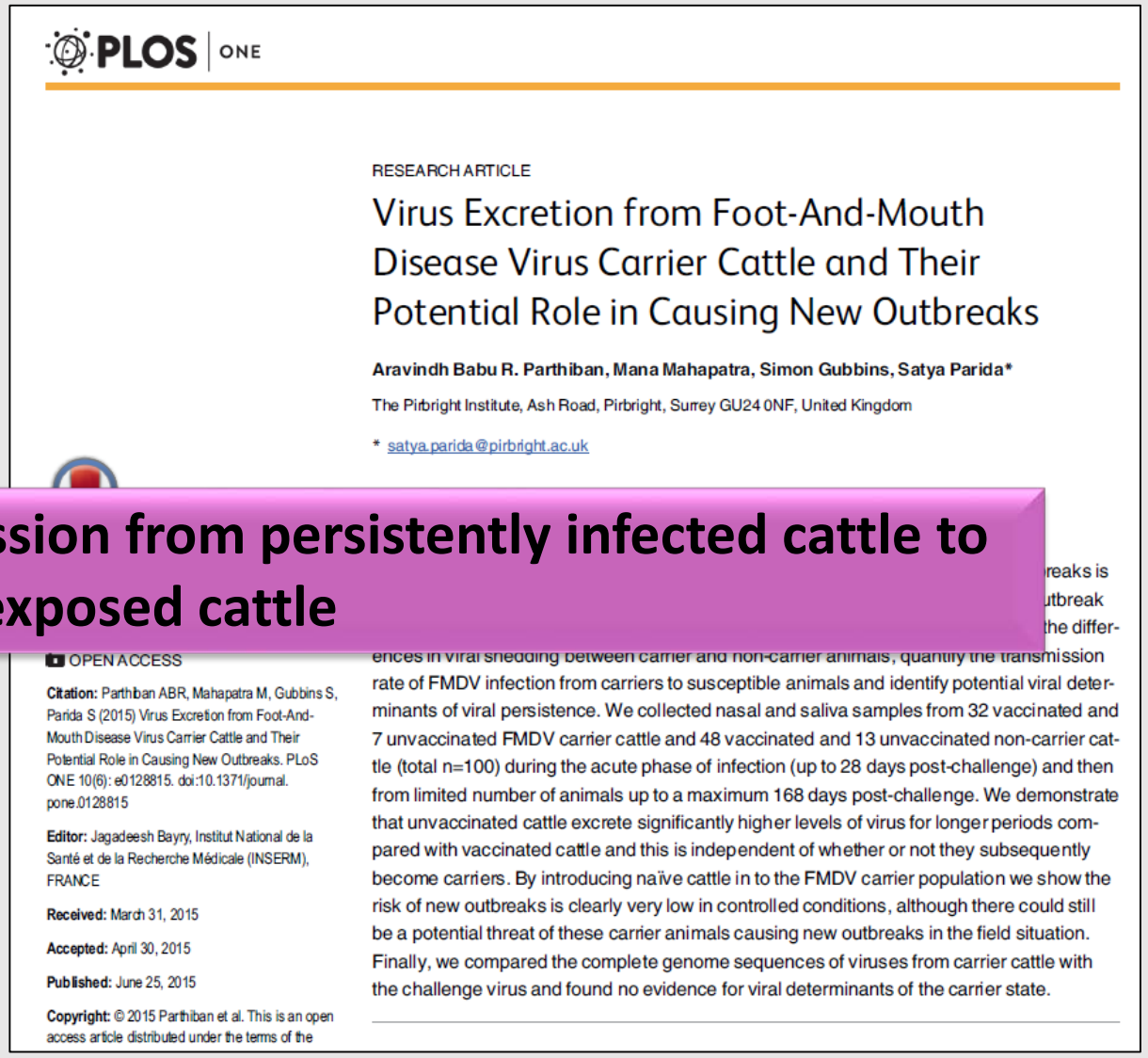
Ref. No.[a]	Number of Cases	Number of Carriers	Number of Susceptibles	N[b]	Contact Days	Species
(7)	0	6	4	10	35	Cattle to cattle
(30)	0	14	8	22	43	Cattle to cattle
(28)	0	17	1	18	549	Cattle to cattle
(31)	0	1	1	2	61	Cattle to cattle
(31)	0	2	2	4	274	Cattle to cattle
(31)	0	2	2	4	274	Cattle to cattle
(31)	0	2	1	3	274	Cattle to cattle
(11)	0	1	2	3	42	Cattle to cattle
(11)	0	1	2	3	42	Cattle to cattle
(11)	0	1	2	3	42	Cattle to cattle
(11)	0	1	2	3	28	Cattle to cattle
(11)	0	1	2	3	28	Cattle to cattle
(11)	0	1	2	3	28	Cattle to cattle
(31)	0	2	1	3	84	Sheep to cattle
(9)	0	5	6	11	548	Buffalo[c] to cattle
(32)	0	1	2	3	175	Buffalo to cattle
(32)	0	1	2	3	152	Buffalo to cattle
(33)	0	6	6	12	456	Buffalo to cattle
(34)	1	3	4	7	168	Buffalo to cattle
(35)	0	6	3	9	731	Buffalo to cattle
(36)	2[d]	3	2	5	312	Buffalo to cattle
(37)	0	1	7	8	122	Buffalo to buffalo
(36)	1	3	1	4	198	Buffalo to buffalo
(38)	0	4	6	10	30	Cattle to pigs
(38)	0	4	6	10	30	Cattle to pigs
(38)	0	4	6	10	30	Cattle to pigs
(38)	0	4	6	10	30	Cattle to pigs
(38)	0	4	6	10	30	Cattle to pigs
(38)	0	4	6	10	30	Cattle to pigs
(39)	2	3	6	9	75	Cattle to pigs
(39)	0	2	6	8	86	Cattle to pigs
(39)	0	3	4	7	44	Cattle to pigs
(39)	0	1	4	5	34	Cattle to pigs
(39)	0	1	4	5	35	Cattle to pigs
(39)	0	2	4	6	29	Cattle to pigs
(31)	0	2	4	6	91	Cattle to pigs

[a]Number of reference (in list). In one paper, more than one experiment or experimental unit could be included.
[b]N = Total number of animals (carriers + susceptible).
[c]African buffalo (*Syncerus caffer*).
[d]It was assumed that only one susceptible became infected due to contact with the carrier, and the second was infected by the first contact-infected animal.

Tenzin et al., 2008: Meta-analysis of FMDV transmission from persistently infected cattle and buffalo

0.0148 infections per carrier per month

Transmission from FMDV carriers



PLOS | ONE

RESEARCH ARTICLE

Virus Excretion from Foot-And-Mouth Disease Virus Carrier Cattle and Their Potential Role in Causing New Outbreaks

Aravindh Babu R. Parthiban, Mana Mahapatra, Simon Gubbins, Satya Parida*

The Pirbright Institute, Ash Road, Pirbright, Surrey GU24 0NF, United Kingdom

* satya.parida@pirbright.ac.uk

No transmission from persistently infected cattle to (4) contact exposed cattle

reaks is
utbreak
he differ-
ences in viral shedding between carrier and non-carrier animals, quantify the transmission rate of FMDV infection from carriers to susceptible animals and identify potential viral determinants of viral persistence. We collected nasal and saliva samples from 32 vaccinated and 7 unvaccinated FMDV carrier cattle and 48 vaccinated and 13 unvaccinated non-carrier cattle (total n=100) during the acute phase of infection (up to 28 days post-challenge) and then from limited number of animals up to a maximum 168 days post-challenge. We demonstrate that unvaccinated cattle excrete significantly higher levels of virus for longer periods compared with vaccinated cattle and this is independent of whether or not they subsequently become carriers. By introducing naïve cattle in to the FMDV carrier population we show the risk of new outbreaks is clearly very low in controlled conditions, although there could still be a potential threat of these carrier animals causing new outbreaks in the field situation. Finally, we compared the complete genome sequences of viruses from carrier cattle with the challenge virus and found no evidence for viral determinants of the carrier state.

OPEN ACCESS

Citation: Parthiban ABR, Mahapatra M, Gubbins S, Parida S (2015) Virus Excretion from Foot-And-Mouth Disease Virus Carrier Cattle and Their Potential Role in Causing New Outbreaks. PLoS ONE 10(6): e0128815. doi:10.1371/journal.pone.0128815

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Transmission from FMDV carriers

AMERICAN SOCIETY FOR MICROBIOLOGY | Journal of Virology

Differential Persistence of Foot-and-Mouth Disease Virus in African Buffalo Is Related to Virus Virulence

Francois Maree,[a,b] Lin-Mari de Klerk-Lorist,[c] Simon Gubbins,[d] Fuquan Zhang,[d] Julian Seago,[d] Eva Pérez-Martín,[d] Liz Reid,[d] Katherine Scott,[a] Louis van Schalkwyk,[c] Roy Bengis,[c] Bryan Charleston,[d] Nicholas Juleff[d]

Transboundary Animal Disease Programme, ARC-Onderstepoort Veterinary Institute, Onderstepoort, South Africa[a]; South Africa Department of Microbiology and Plant Pathology, University of Pretoria, Pretoria, South Africa[b]; Office of the State Veterinarian, Department of Agriculture, Forestry and Fisheries, Skukuza, South Africa[c]; The Pirbright Institute, Woking, Surrey, United Kingdom[d]

NO transmission from persistently infected African buffalo to contact exposed cattle

Transmission from FMDV carriers?

Field trial: Vietnam

- **NO transmission from persistently infected cattle to contact exposed cattle**

Transboundary and Emerging Diseases

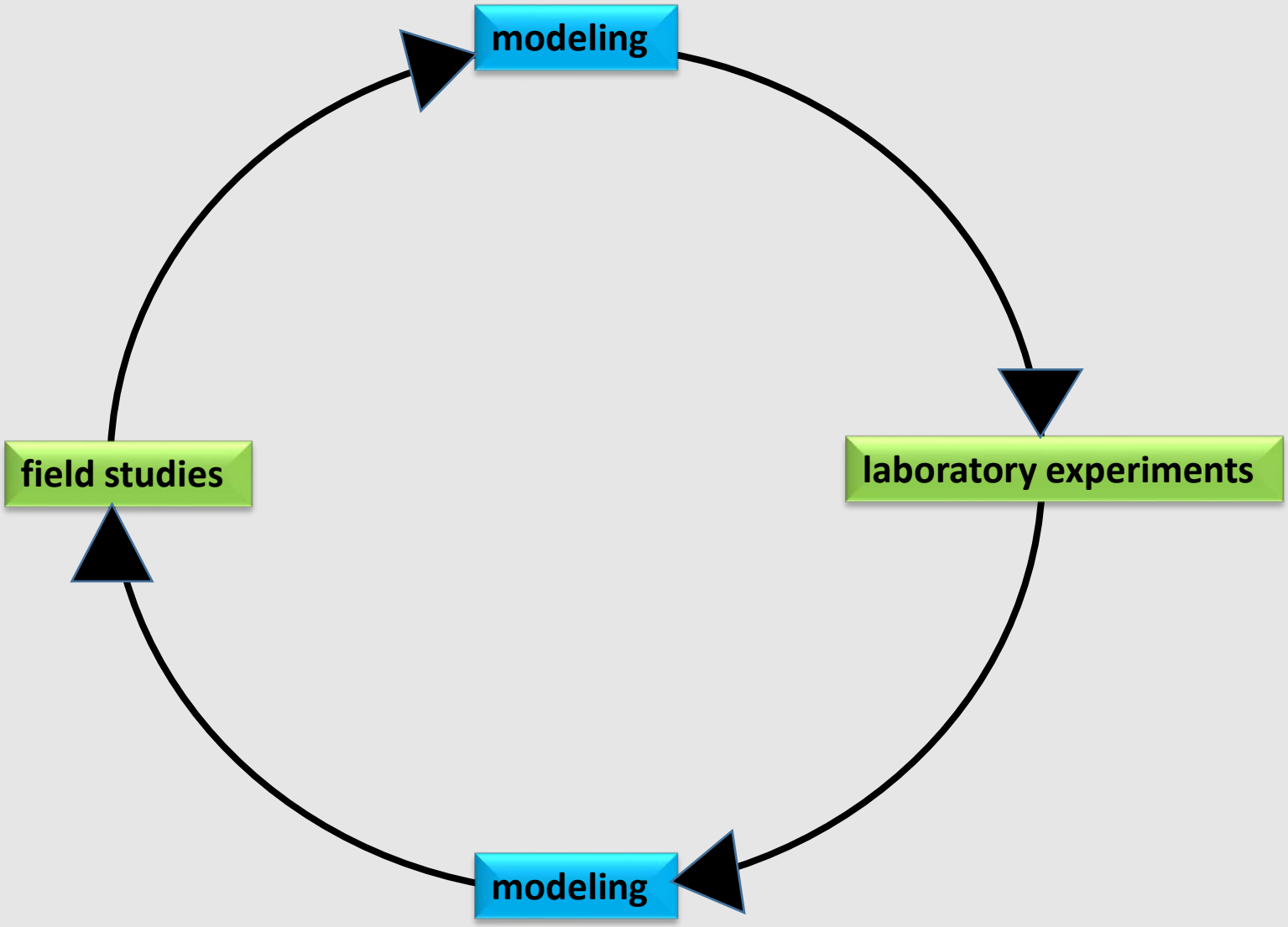
Transboundary and Emerging Diseases

ORIGINAL ARTICLE

An Integrative Analysis of Foot-and-Mouth Disease Virus Carriers in Vietnam Achieved Through Targeted Surveillance and Molecular Epidemiology

H. C. de Carvalho Ferreira[1,2], S. J. Pauszek[1], A. Ludi[1,2,*], C. L. Huston[3], J. M. Pacheco[1], V. T. Le[4], P. T. Nguyen[4], H. H. Bui[4], T. D. Nguyen[5], T. Nguyen[5], T. T. Nguyen[6], L. T. Ngo[4], D. H. Do[6], L. Rodriguez[1] and J. Arzt[1]

Keeping it real



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- Carolina Stenfeldt
- Barbara Brito

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- Kimberly VanderWaal
- Amy Delgado

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- Ngo Thanh Long
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- Jitendra K. Biswal
- Bramhadev Pattnaik

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- DTRA-DOD
- CEAH-APHIS-USDA (IAA)
- Science and Technology Directorate, DHS (IAA; primary animal work)

Keeping it real