

OIE/FAO Foot-and-Mouth Disease Reference Laboratory Network

Annual Report 2021

Editors:

Dr Donald King, Dr Antonello Di Nardo and Dr Mark Henstock, The Pirbright Institute, UK

Maps within this document

All maps within this document were drawn using the United Nations Map (UNMap) v2020, supplied to the authors by the FAO. The following disclaimers apply to the maps in this document.

The designations employed and the presentation of material on this map do not imply the expression of any opinion whatsoever on the part of the Secretariat of the United Nations concerning the legal status of any country, territory, city or area or of its authorities, or concerning the delimitation of its frontiers or boundaries.

Jammu and Kashmir: Dotted line represents approximately the Line of Control in Jammu and Kashmir agreed upon by India and Pakistan. The final status of Jammu and Kashmir has not yet been agreed upon by the parties.

Sudan and South Sudan: Final boundary between the Republic of Sudan and the Republic of South Sudan has not yet been determined.

Abyei: Final status of the Abyei area is not yet determined.

Falkland Islands (Malvinas): A dispute exists between the Governments of Argentina and the United Kingdom of Great Britain and Northern Ireland concerning sovereignty over the Falkland Islands (Malvinas).

Use of data (including all images) from this document

Copies of all the individual reports cited herein can be obtained from WRLMD (<u>www.wrlfmd.org</u>) and please seek permission before presentation, publication or other public use of these data.



Contents

1	OIE/F	AO FMD Reference Laboratory Network	3		
	1.1	Principle Goals	3		
	1.2	Reporting Period	4		
	1.3	Collated input from	4		
2	Globa	I distribution and impact of foot-and-mouth disease	6		
	2.1	Introduction	6		
	2.2	Overview of the activities of the OIE/FAO FMD Laboratory Network during 2021	10		
	2.3	Regional distribution of different FMD viral lineages	13		
	2.4	Vaccine matching and recommendations	17		
3	Overv	iew of Network surveillance activities in each of the regional endemic pools	18		
	3.1	Pool 1 Regional synopsis	18		
	3.2	Pool 2 Regional synopsis	20		
	3.3	Pool 3 Regional synopsis	21		
	3.4	Pool 4 Regional synopsis	23		
	3.5	Pool 5 Regional synopsis	25		
	3.6	Pool 6 Regional synopsis	27		
	3.7	Pool 7 Regional synopsis	28		
4	Impro	ving the quality of laboratory tests from FMD reference laboratories	30		
	4.1	Proficiency testing schemes (PTS) organised by the Network Partners	30		
	4.2	Supply of reagents	32		
	4.3	Training courses organised by Network partners	36		
	4.4	Collaborative projects	37		
	4.5	In vivo potency studies undertaken during 2021	41		
A re	ppendix 1 egions tes	Details of clinical samples from field cases from countries in FMDV endemic ted during 2021	44		
A	ppendix 2	2 Vaccine matching studies undertaken by Network partners during 2021	46		
Appendix 3 Nucleotide sequence analysis 52					
Appendix 4 Selected phylogenetic trees for 2021 54					
A	ppendix 5	The 16th Annual Meeting of the OIE/FAO FMD Reference Laboratory Network	65		



1 OIE/FAO FMD Reference Laboratory Network

1.1 Principle Goals

The Network of OIE/FAO FMD Reference Laboratories has been established with two principal goals:

1) To understand global virus distribution patterns and use these data to inform vaccine recommendations

and

2) To harmonise and improve the quality of laboratory testing carried out by international and national reference laboratories.

These activities require sharing and joint evaluation of surveillance information from laboratory diagnosis, serotyping, genetic characterisation and vaccine matching tests and harmonisation of standards for diagnostic procedures.

This report is divided into two parts providing an update on progress towards each of these goals.



1.2 Reporting Period

1st January 2021 - 31st December 2021

1.3 Collated input from



Figure 1-1: Participating laboratories







Dakar, Senegal

Ankara, Turkey



2 Global distribution and impact of foot-and-mouth disease

Foot-and-mouth disease (FMD) is a highly contagious viral disease that infects a wide variety of domestic and wild cloven-hooved hosts. Its presence impacts upon rural livelihoods and restricts trade opportunities for countries where the disease is endemic and poses a constant threat to those countries that are free of the disease. In endemic countries, the economic costs associated with FMD are estimated to be US\$6.5–21 billion annually, with outbreaks in FMD-free countries and zones potentially causing economic losses of >\$1.5 billion. FMD virus lineages are not randomly dispersed throughout the world but are associated with particular ecological niches. The distribution of these FMD virus lineages is affected by cyclical upsurges in the prevalence of particular strains that may be associated with the evolution of FMD virus sto escape protective immunity in susceptible livestock populations and/or opportunities presented by movements of animals and their products. These features can give rise to pandemic events where FMDV lineages spread widely to affect new regions.

Global surveillance for FMD is necessary to identify the current hazards and to predict heightened risk so that appropriate diagnostic tools and vaccines are available for detection and control. This requires sustained effort directed towards the monitoring of FMD outbreaks and ideally also of FMDV circulation and persistence, along with collection and characterisation of FMD viruses and integration of findings with associated epidemiological intelligence. Such an extensive effort requires a coordinated approach encompassing national and international disease laboratories of the **OIE/FAO FMD Laboratory Network** (www.foot-and-mouth.org) along with partnering laboratories, commercial vaccine and diagnostic providers. The worldwide distribution of the different serotypes and variants of FMD virus (as compiled in 2021) and the associated activities of the Network laboratories are presented in this report.

2.1 Introduction

Global surveillance undertaken by the OIE/FAO FMD Laboratory Network aims to monitor the distribution of FMD viruses to predict risk for endemic and FMD-free countries. FMDV is unevenly distributed throughout the world reflecting factors such as livestock density and species mix, patterns of husbandry, animal movement and trade, wildlife reservoirs and incentives and capacities for disease control. The virus exists as seven serotypes and multiple subtypes where cross-immunity is absent or incomplete. The situation is dynamic and complex and affected by viral evolution, waxing and waning of host immunity and changing ecosystems and trading patterns. Despite the opportunities for spread of FMDV into new regions, viruses tend to recur in the same parts of the world, presumably reflecting some degree of either ecological isolation or adaptation. On this basis, the global pool of FMD viruses can be subdivided into seven 'regional pools' in which genetically and antigenically distinctive virus strains tend to occur within a defined region.



The seven 'Regional Pools' referred to throughout this report are shown below (Figure 2-1) and represent:

- Pool 1 Southeast Asia with spill over into Eastern Asia
- Pool 2 Southern Asia
- Pool 3 Western Asia with spill over into North Africa
- Pool 4 Eastern Africa with spill over into North Africa
- Pool 5 Western Africa
- Pool 6 Southern Africa
- Pool 7 South America



Figure 2-1: Distribution of the seven endemic pools of FMD showing conjectured status of FMD in countries during 2021. Periodically, viruses spread between pools and to free regions, and countries at the interfaces between pools (such as in North Africa and Central Asia) often experience FMD outbreaks from different regional sources. Note on Pools 4-6: In Africa there are currently three FMD virus pools loosely defined as covering East Africa (pool 4), West Africa (pool 5) and Southern Africa (pool 6). A map describing the official OIE status for these countries can be found at: https://www.oie.int/en/disease/foot-and-mouth-disease/#ui-id-2

The clustering of FMD viruses into 7 virus pools, with 3 pools covering West Eurasia, South Asia and Southeast Asia, 3 pools covering East, West and Southern Africa and 1 pool covering the Americas, is now enabling a targeted approach to be applied to the 'Progressive Global Control of FMD' initiative overseen by the OIE and FAO and for which the Network laboratories will play a pivotal role.

Overview of the Global situation in 2021

Headline events (Figure2-2) in 2021 include:

The identification of serotype O (O/EA-2 topotype) in Namibia, where cases in July
represent the continued spread of this lineage that originates from East Africa.
Together with cases due to the O/EA-2 topotype that have also been detected recently
in Zambia, these outbreaks pose new threats to Southern Africa where serotype O is
not normally present.



- Increasing dominance of the O/ME-SA/Ind-2001e lineage in Pool 1 where it appears to be supplanting previously circulating serotype O lineages (including O/SEA/Mya-08 and O/ME-SA/PanAsia). This lineage continues to be detected in Pakistan (where it was first detected in 2019) and there has also been a second detection of the O/ME-SA/Ind-2001e lineage in Mauritius in 2021 that appears to be distinct to viruses that caused outbreaks in 2016.
- Detection of SAT 2 (topotype I) in KwaZulu-Natal in May 2021 that represents an FMD outbreak that has occurred away from the usual areas of high concern in southern Africa.
- FMD viruses detected in animals imported from East Africa to Bahrain were characterised as O/EA-3 and A/AFRICA/G-1 highlighting the ease by which FMD viruses can be imported into new areas via the trade in live animals.



Figure 2-2: Headline FMD events for 2020 (highlighted in yellow – important epidemiological events from 2019 are also shown in grey)

Specific information regarding contemporary FMD outbreaks can be found on the World Animal Health Information Database (WAHID) located on the OIE website (<u>https://wahis.oie.int/#/home</u>), as well as the EMPRES Global Animal Disease Information System (<u>http://empres-i.fao.org/</u>) provided by FAO. Further supplementary data and updates are provided in the WRLFMD/EuFMD Quaterly Report for FMD (<u>https://www.wrlfmd.org/ref-lab-reports</u>).

During 2021, FMD outbreaks have continued to affect countries in the established endemic regions of the world. Particular attention has been focussed upon new FMD outbreaks and events that have occurred at the margins of these endemic regions (reported on the OIE



WAHIS Interface: <u>https://wahis.oie.int/#/home</u>, summarised in Figure 2-3, Table 2-1 and described elsewhere in this report). Further details of many of the characterisation of viruses retrieved from these outbreaks are provided later in this report.



Figure 2-3: Map indicating the location of significant epidemiological events and disease outbreaks reported to OIE in immediate notifications or follow-up reports in 2021 (data, available from: https://wahis.oie.int/#/home, downloaded on 9 February 2022)

Table 2-1: New FMD	outbreaks	reported to	o OIE	during	2021	(data	retrieved	from	WAHIS	on
www.oie.int on 9th Feb	ruary 2022)									

				Number of	animals		
Country	New outbreaks	Susceptible	Cases	Killed and disposed of	Slaughtered	Deaths	Vaccinated
China (People's Rep. of)	2	74	45	70	0	4	0
Israel	19	6978	812	0	0	114	0
Jordan	8	4925	421	0	0	27	351760
Libya	3	84	49	0	0	16	0
Malawi	1	11200	85	0	0	0	5466
Mauritius	1	57	16	0	0	0	0
Mongolia	9	1094	1097	0	0	0	12603
Namibia	5	408	178	0	0	0	0
Pakistan	994	192195	11903	0	0	299	30208253
Palestine	4	2212	560	0	0	19	0
							9 Page



				Number o	of animals		
Country	New outbreaks	Susceptible	Cases	Killed and disposed of	Slaughtered	Deaths	Vaccinated
Russia	1	39	2	39	0	0	2557
Somalia	2	1100	12	0	0	0	0
South Africa	62	145004	665	0	34510	0	0
United Arab Emirates	3	1067	65	65	0	0	0
Zambia	18	26915	1196	0	0	0	0
Zimbabwe	2	1188	63	0	0	0	4471

2.2 Overview of the activities of the OIE/FAO FMD Laboratory Network during 2021

The OIE/FAO FMD Reference Laboratory Network provides important support to the global control of FMD and provides opportunities and expertise for developing and sustaining laboratory capacity and capability, exchange of materials and technologies, harmonising approaches to diagnosis and supporting complementary research. Laboratories within the Network regularly receive samples for FMD diagnosis from many parts of the world. The *in vitro* antigenic properties of selected isolates are assessed for vaccine matching and nucleotide sequencing allows precise characterisation of new isolates and tracing of their origin by comparison with viruses held in virus collections. This analysis assists the monitoring of the 'real time' emergence and spread of FMD virus globally.

1672 clinical samples from suspect cases of FMD were tested by laboratories in the Network (and associated laboratories) during 2021. These samples were collected from 30 countries from all seven FMD endemic pools (Figure 2-4). **However, sampling within these pools is not equivalent:** and efforts are currently underway with the Network to improve sample collection in regions where sampling is particularly under-represented.





Figure 2-4: Distribution of samples collected from suspect cases of FMD and reported by the OIE/FAO FMD Laboratory network during 2021. Routine surveillance that is undertaken in countries that are FMD-free without vaccination is not shown (NB: PDFMD reports retrospective for India which is not shown on this figure)



Figure 2-5: Clinical samples (n=1735) tested for FMD investigation (virology) by the OIE/FAO FMD Laboratory Network from FMD endemic countries during 2021 and their distribution across the seven FMD endemic pools (see Figure 2-1).





Figure 2-6: Summary of results for characterised isolates (n=1076) from FMD endemic countries were reported by the Network during 2021. FMDV GD denotes samples that were only positive using molecular (RT-PCR methods), while a further 678 samples were tested but found to be negative for FMDV using all diagnostic methods.



Figure 2-7: Summary of 817 samples (viruses and field isolates) that were sequenced (VP1/capsid/complete genome) during 2021 (see Appendix 3).

The results for the individual samples are reported later in this report. Characterization results obtained on samples received by WRLFMD and PANAFTOSA can also be found respectively at: <u>http://www.wrlfmd.org/</u> and at: <u>http://new.paho.org/panaftosa</u>.



2.3 Regional distribution of different FMD viral lineages

In regions where FMD is endemic, continuous evolution of the virus generates geographically discrete lineages that are genetically distinct from FMD viruses found elsewhere. The conjectured global status for FMD (see Figure 2-1) masks the underlying complexity of FMDV virus distribution in the different pools (at serotype, topotype and lineage levels). This report showcases a new format to display how different FMD lineages ciruculate in different regions of the world. Using a new tool (called PRAGMATIST) that has been developed in partnership between WRLFMD and EuFMD, analyses accommodate the latest epidemiological data collected by the Network and presented in this report regarding FMDV lineages detected in samples to assess the relative importance of the viral strains circulating within each *source regions* (see Table below).

Table 2-2: Conjectured distribution of important FMDV lineages in different endemic regions. For each of the regions, data represent the relative importance of the different lineages [prevalence score estimated as a proportion (%) of total FMD cases that occur in domesticated host animals]. NB: Arrows highlight changes from the figures published in this table in last year's report

FMDV Lineage	West Eurasia	East Asia	North Africa	South Asia	East Africa	West & Central Africa	Southern Africa	South America
O/ME-SA/PanAsia-2	35							
O/ME-SA/PanAsia		10						
O/SEA/Mya-98		↓ 21.5₃₃						
O/ME-SA/Ind2001	7	1 40 20	₽ 10	186 80				
O/EA/WA	3		55		1 55.5	465 70	16 °	
O/EURO-SA								190 80
O/CATHAY		10.5						
A/ASIA/Sea-97		18 26						
A/ASIA/Iran-05	132 27	0						
A/ASIA/G-VII	10 15			10 16				
A/AFRICA			133 25		22	17 15		
A/EURO-SA								10 20
Asia-1	12.5	0 0.5		4				
SAT 1			0		8	3 5	16 27	
SAT 2	0.5		10		14	15 10	1 52 57	
SAT 3					0.5 1		16	
С								



Based on these data, a *prevalence score* is defined by estimating the proportion of each of the local viral strains that would be represented if 100 animals infected with FMDV were randomly selected from each source area.

In order to help visualise the changing patterns in FMDV distribution and recognise risks for the emergence of new lineages, the Network has reviewed available intelligence for epidemiologically important FMDV lineages (Table 2-2), focussing on those that have already demonstrated a potential for long-distance trans-pool spread: O/ME-SA/Ind-2001, O/ME-SA/PanAsia, O/ME-SA/PanAsia-2, O/SEA/Mya-98, O/EA-3, A/ASIA/G-VII, A/ASIA/Iran-05, A/ASIA/Sea-97 and SAT 2/VII.

The current known and conjectured distribution of these different FMD viral lineages are represented in the maps below: The extent of current distribution for each of the viral lineages is represented within the black lines, while the location of individual outbreaks (dots) and affected countries (shaded colours, according to dates) are shown. NB: Arrows are drawn to highlight the regions that are now threatened by these lineages and text boxes highlight some of the headline events and changes that have occurred during 2020-21.



FMDV O





Figure 2-8: Conjectured distribution of important serotype O FMDV lineages.

Main events in 2021:

- O/ME-SA/Ind-2001e has become established as the dominant serotype O lineage in Southeast Asia]
- A new incursion of O/ME-SA/Ind-2001e in Mauritius which is genetically distinct to the virus that caused outbreaks in 2106
- O/ME-SA/PanAsia-2^{ANT-10} outbreaks in the eastern Mediterranean (Jordan, Israel, Palestine)
- FMD cases due to O/EA-3 in Bahrain (animals recently imported from East Africa)
- Elsewhere, spread of O/EA-2 in southern Africa (not shown here)



FMDV A



FMDV Asia 1



Figure 2-10: Conjectured distribution of serotype Asia 1.

Main events in 2021:

• No further spread of this serotype in Southeast Asia (beyond cases reported in 2017)

FMDV SAT 2



Figure 2-11: Conjectured distribution of serotype the SAT 2 (topotype VII) FMDV lineage.



2.4 Vaccine matching and recommendations

These take two forms: regional recommendations and details of locally produced vaccines for each of the FMD endemic pools are summarised later in this report, whilst the WRLFMD recommendations for FMD free countries are given in Figure 2-12 below. Details of vaccine matching work undertaken by the Network are summarised in Appendix 2.



Vaccine Antigen Prioritisation: Europe

Figure 2-12: Recommendations from WRLFMD on FMD virus strains to be included in FMDV vaccine antigen bank for Europe

Outputs from WRLFMD are generated with a tool (called PRAGMATIST) that has been developed in partnership between WRLFMD and EuFMD. These analyses accommodate the latest epidemiological data collected by the Network regarding FMDV lineages that are present in different source regions (see Table 2-2 above), as well as available in vitro, in vivo and field data to score the ability of vaccines to protect against these FMDV lineages. Further information about FMD vaccine producers is available on the Network website: https://www.foot-and-mouth.org/fmd-vaccine-producers

The figure highlights the importance of these source regions for Europe (using data collected at the EU-RL Workshop); please contact WRLFMD/EuFMD for assistance to tailor these outputs to other geographical regions. NB: Vaccine-coverage data presented is based on available data and may under-represent the true performance of individual vaccines.



3 Overview of Network surveillance activities in each of the regional endemic pools

3.1 Pool 1 Regional synopsis

3.1.1 Conjectured circulating FMD viral lineages in Pool 1 during 2021

- Serotype O:
 - o SEA/Mya-98
 - o ME-SA/PanAsia
 - o ME-SA/Ind2001
 - CATHAY
- Serotype A:
 - o ASIA/Sea-97
- Serotype Asia-1 (no outbreaks detected since 2017, Myanmar)

 Table 3-1: Overview of clinical samples collected and tested from Pool 1 in 2021 (countries highlighted in blue; graph represents clinical submissions since 2005)



		Number of	f Samples
Laboratory	Countries of Origin	Clinical Field	Surveillance
		Cases	Activities
AQPA	Cambodia, Republic of Korea, Vietnam	43	898279
ARC-OVI	Thailand	0	1
FGBI ARRIAH	Russia	0	116287
LVRI	China	17	2551
RRLSEA	Lao PDR, Thailand	95	5332
WRLFMD	Mongolia, Vietnam	32	0

* 54684 from Russia. Some of these samples may be from Pool 3



Pool 1 headlines:

• O/ME-SA/Ind-2001e lineage is the predominant serotype O lineage in the region. Data below from WRLFMD summarises sequencing data from WRLFMD for samples collected from mainland southeast Asian countries (Cambodia, Laos, Myanmar, Thailand and Vietnam):



- Serotype A outbreaks due to A/ASIA/Sea-97 have also been reported in Thailand
- In China, two FMD outbreaks due to O/ME-SA/Ind-2001e have also been reported as well as an outbreak due to O/CATHAY (see Appendix 4.1).
- O/ME-SA/Ind-2001e lineage viruses from two separate clades have also caused FMD outbreaks in Mongolia (see Appendix 4.2).
- No FMD outbreaks have been reported in the Republic of Korea since 2019.
- No new outbreaks due to serotype Asia1 were detected in 2021. This serotype has been absent since 1998, with the exception of outbreaks in Vietnam (2006) and Myanmar (2017).

3.1.2 Vaccine recommendations for Pool 1

- Internationally produced vaccines:
 - o O: Campos, Manisa, Primosky, TUR/5/2009 & 3039
 - A: Arg2001, A24 Cruzeiro, Iran/05, A22/Iraq/64, Malaysia/97, TUR/20/06 & Zabaikalsky.
 - o Asia 1: Shamir
- Locally produced vaccines (at RRL SEA):
 - o 0: 189/87 (Udornthani/87)
 - o A: Lopburi/12, Sakolnakorn/97
 - o Asia1: Petchaburi/85
- Locally produced vaccines (at FGBI ARRIAH):
 - o O: Ind-2001, Mya-98, PanAsia, PanAsia-2
 - o A; G-VII, Iran-05, Sea-97



- Asia1: Shamir, Sindh-08
- Locally used vaccine strains (by Chinese manufactures):
 - o O/Mya-98 (O/Mya98/BY/2010 and Re-O/Mya98), O/HK99
 - Re-A/Sea-97 (Re-A/WH/09)
 - o Asia1/GV (Asia1/JSL/06).

3.2 Pool 2 Regional synopsis

3.2.1 Conjectured circulating FMD viral lineages in Pool 2 during 2021

- Serotype O:
 - o ME-SA/Ind-2001
 - o ME-SA/SA-2018
- Serotype A:
 - ASIA/IND (genotype VII also known as genotype 18)
- Serotype Asia-1

Table 3-2: Overview of clinical samples collected and tested from Pool 2 in 2021 (countries highlighted in blue; graph represents clinical submissions since 2005)



		Number of	f Samples
Laboratory	Countries of Origin	Clinical Field	Surveillance
		Cases	Activities
ANSES	Mauritius	18	47
APQA	Bangladesh	20	0
ARC-OVI	India	0	1
WRLFMD	Nepal	70	0



Pool 2 headlines:

- No data provided from ICAR-DFMD, India for 2021
- Clinical samples from Nepal demonstrate that A/ASIA/G-VII and O/ME-SA/Ind-2001e are actively circulating in the pool.
- There have been new FMD cases in Mauritius where sequence data supports a new introduction of the virus from Pool 2 (see: Appendix 4.3)
- Vaccine protection studies highlight the antigenic differences between A/ASIA/G-VII and A/ASIA/Iran-05 (see: Singanallur et al., 2022).

Singanallur et al., (2022) A Vaccine Based on the A/ASIA/G-VII Lineage of Foot-and-Mouth Disease Virus Offers Low Levels of Protection against Circulating Viruses from the A/ASIA/Iran-05 lineage. Viruses. 14(1):97.

3.2.2 Vaccine recommendations for Pool 2

- Internationally produced vaccines:
 - O/ME-SA/PanAsia-2 (or suitable alternative). *In vitro* vaccine matching data for O/ME-SA/Ind2001 provides evidence for an antigenic match with O/TUR/09 vaccine (MSD) and O-3039 (Boehringer Ingelheim).
- Locally produced vaccines (by Indian suppliers):
 - o O/IND/R2/1975
 - o A/IND/40/2000
 - o Asia1/IND/63/1972

3.3 Pool 3 Regional synopsis

3.3.1 Conjectured circulating FMD viral lineages in Pool 3 during 2021

- Serotype O:
 - ME-SA/PanAsia-2 [comprising at least two viral sublineages (ANT-10 and QOM-15) present in different countries].
 - ME-SA/Ind-2001 (via introductions from South Asia: Pool 2)
 - EA-3 (in Israel & Palestinian Autonomous Territories)
- Serotype A:
 - ASIA/Iran-05 [comprising 4 predominant viral sublineages (SIS-10, SIS-12, SIS-13 and FAR-11)]
 - o ASIA/G-VII
- Serotype Asia-1:
 - o Sindh-08



Table 3-3: Overview of clinical samples collected and tested from Pool 3 in 2021 (countries highlighted in blue; graph represents clinical submissions since 2005)



Pool 3 headlines:

- New FMD outbreaks in Eastern Mediterranean countries (Jordan, Israel and Palestine) have been caused by the O/ME-SA/PanAsia-2^{ANT-10} lineage – viruses which share closest genetic relationship to viruses collected in Pakistan in 2019 (see Appendix 4.4). Previously, the dominant serotype O virus in the region was O/ME-SA/PanAsia-2^{QOM-15}.
- In Turkey, only serotype O FMD outbreaks have been detected during 2021 (from PanAsia-2 QOM-15 and ANT-10 sublineages; see Appendix 4.5). Serotypes A and Asia-1 have not been detected since 2018 and 2015 respectively.
- Serotype A outbreaks (due to A/ASIA/Iran-05^{FAR-11}) have been detected in Iran which generated poor antigenic matching data when isolates were tested at the ŞAP Institute, Turkey. See: https://rr-europe.oie.int/en/our-missions/animaldiseases/foot-and-mouth-disease/west-eurasia-fmd-roadmaps /
- New FMD outbreaks due to a FMD virus from the O/ME-SA/Ind-2001e lineage most closely related to viruses from Mongolia.have been reported in Russia (Orenburg region) in December 2021 (see Appendix 4.6)
- In Bahrain, FMD cases due to viruses from serotype O (O/EA-3) and A (A/AFRICA/G-I) with an East African origin were detected (see Appendices 4.7 and 4.8)

3.3.2 Vaccine recommendations for Pool 3

Internationally produced vaccines



- MSD and Boehringer-Ingelheim (Merial)*:
 - O/ME-SA/PanAsia-2 (or suitable alternative)
 - o O/Manisa
 - A Iran-05 (or A TUR 06)
 - o A22/Iraq
 - o Asia-1 Shamir
 - o A/G-VII
- Locally produced vaccines (at FGBI ARRIAH):
 - o O: Ind-2001, Mya-98, PanAsia, PanAsia-2
 - o A; G-VII, Iran-05, Sea-97
 - Asia1: Shamir, Sindh-08
- Locally produced vaccines:
 - o O/TUR/07 (PanAsia 2)
 - A05 (A/IRN/17)
 - o A/ASIA/Iran 05^{FAR-11}
 - o A/Asia/G-VII
 - o Asia 1/Sindh-08
- Locally produced vaccines (other suppliers in the region):
 - o Vetal
 - o MEVAC

3.4 Pool 4 Regional synopsis

3.4.1 Conjectured circulating FMD viral lineages in Pool 4 during 2021

- Serotype O:
 - o EA-2 (Namibia, Zambia, Kenya, Tanzania, DR Congo, Uganda)
 - o EA-3 (Egypt, Ethiopia, Eritrea, Sudan)
 - EA-4 (Ethiopia, Kenya, Uganda)
 - o ME-SA/Sharqia-72 (detected in samples collected in Egypt in 2009)
 - o ME-SA/Ind2001 (in Libya, Tunisia, Algeria and Morocco)
- Serotype A
 - o AFRICA/I (Kenya, Tanzania, D.R. Congo)
 - AFRICA/IV (Algeria, Sudan, Eritrea, Egypt)
 - AFRICA/VII (Ethiopia, Egypt)
 - ASIA/Iran-05^{BAR-08} (Egypt)
- Serotype SAT 1
 - o I (Kenya, Tanzania)
 - o IX (Ethiopia)
- Serotype SAT 2:
 - o IV (Kenya, Tanzania)
 - VII (Sudan, Egypt, Mauritania)
 - o XIII (Ethiopia, Sudan)
- Serotype SAT 3
 - Only detected in African buffalo in the south of the Queen Elizabeth National Park, Uganda in 1970, 1997 and 2013).



Table 3-4: Overview of clinical samples collected and tested from Pool 4 in 2021 (countries highlighted in blue; graph represents clinical submissions since 2005). *Note: These figures include samples collected in countries in North Africa where FMD outbreaks have occurred since 2013.*



Pool 4 headlines:

WRLFMD

• Recent analyses of FMDV sequences support the idea that the East African pool represents two separate ecosystems that maintain discrete viral lineages (see: Gizaw et al., 2020).

Dem. Rep. of Congo,

Kenya, Uganda

97

0

• A new initiative launched during late 2019/early 2020 aims to motivate vaccine producers to supply good quality FMD vaccines into the East African market (see: https://agresults.org/projects/fmd-vaccine)

Gizaw et al., (2020) Molecular characterization of foot-and-mouth disease viruses circulating in Ethiopia between 2008 and 2019. Transbound Emerg Dis. 67(6): 2983-2992

3.4.2 Vaccine recommendations for Pool 4

- Internationally produced vaccines:
 - o O: Manisa, 3039
 - o O: PanAsia-2 (or equivalent)
 - o A: Eritrea
 - o SAT 1: Sat105, SAT109
 - o SAT 2: SAT251, Eritrea
 - o SAT 3: SAT306, SAT309
- Locally produced vaccines from KEVIVAPI (Kenya):
 - o O: K 77/78 EA1
 - o A: K5/80 G1



- SAT1: T155/71 NWZ
- o SAT2: K52/84 IV
- Locally produced vaccines from Ethiopia:
 - o Serotype O (EA-3)
 - Serotype A (Africa/G-III)
 - Serotype SAT 2 (XIII)
- Locally produced vaccines from BVI (Botswana including the following strains O/Manisa 1/78, O/3039, SAT105, SAT109,SAT2035, SAT251, SAT306 & SAT309

3.5 Pool 5 Regional synopsis

3.5.1 Conjectured circulating FMD viral lineages in Pool 5 during 2021

- Serotype O:
 - WA and EA-3 (Nigeria)
- Serotype A:
 - o AFRICA/G-IV & G-VI
- Serotype SAT 1
 - Topotype X (Nigeria and Cameroon)
- Serotype SAT 2:
 - Topotype VII (Mauritania)

Table 3-5: Overview of clinical samples collected and tested from Pool 5 in 2021 (countries highlightedin blue; graph represents clinical submissions since 2005)





Pool 5 headlines:

- Collection of good-quality samples from this region remains an important challenge and Network laboratories have implemented novel approaches using nucleic acid recovery from lateral-flow devices as well as RNA transfection methods to characterize FMD viruses causing outbreaks and to fill gaps in surveillance
- The Network is providing support to FMD reference laboratories in the region via collaborative projects coordinated by NVRI, ANSES, NCFAD, WRLFMD.
- Samples analyzed by ANSES have detected SAT/VII in Burkina Faso and A/AFRICA/G-IV and SAT 2/VII in Niger.
- Samples recently analyzed by NVRI and NCFAD have confirmed the presence of O/EA-3, A/AFRICA/G-IV and SAT 2/VII lineages in Nigeria.

3.5.2 Vaccine recommendations for Pool 5

- Internationally produced vaccines:
 - o **O/Manisa**
 - o O/Maghreb
 - o O/PanAsia-2 (or equivalent)
 - o O: 3039
 - o A: Eritrea
 - SAT 2: Eritrea & Zimbabwe
 - Locally produced vaccines
 - O: NIG 04/14
 - o O: WA and EA-3 topotypes
 - o A: NIG 07/13
 - o A: West Africa (G-IV lineage)
 - o SAT 1: Topotype X
 - o SAT 2: NIG 03/12
 - SAT 2: Topotype VII
 - o O, A, SAT 1 & SAT 2 (Boru-Vacc, Nigeria)

3.6 Pool 6 Regional synopsis

3.6.1 Conjectured circulating FMD viral lineages in pool 6 during 2021

- Serotype O
 - o O/EA-2 topotype
- Serotype SAT 1:
 - o Topotypes I, II and III
- Serotype SAT 2:
 - Topotypes I, II and III
- Serotype SAT 3:



• Topotypes I, II and III

 Table 3-6:
 Overview of clinical samples collected and tested from Pool 6 in 2021 (countries highlighted in blue; graph represents clinical submissions since 2005)



Pool 6 headlines:

- Serotype O viruses (O/EA-2 topotype) have spread from Pool 4 to cause outbreaks in central/southern Zambia and Namibia (see Appendix 4.9). These cases pose new risks to countries in southern Africa where serotype O has not been detected since 2000.
- BVI has recently detected serotype SAT 2 (also in Namibia), SAT 1 (Malawi).
- South Africa reported new FMD outbreaks due to SAT 2 (KwaZulu-Natal Province see Appendix 4.10) and SAT 3 (in the protection zone adjacent to the Limpopo TFP see Appendix 4.11). For the SAT 3 outbreaks, anecdotal evidence suggests that the viruses are being maintained within the cattle population (i.e., not involving buffalo)
- Many of the FMD outbreaks in South Africa are characterised by mild clinical symptoms or subclinical infections

3.6.2 Vaccine recommendations for Pool 6

- Internationally produced vaccines:
 - o O: O Manisa
 - SAT 1: SAT105, SAT 109
 - o SAT 2: SAT251
 - o SAT 3: SAT306, SAT 309
- Locally produced vaccines



- o O: O Manisa
- o SAT 1: SAT105, SAT109, a South African isolate and a Botswana isolate
- o SAT 2: SAT251, SAT2035 and a South African isolate
- SAT 3: SAT306, SAT309 and a South African isolate

3.7 Pool 7 Regional synopsis

Table 3-7: Overview of clinical samples collected and tested from Pool 7 in 2021 (countries highlighted in blue; graph represents clinical submissions since 2005)



Pool 7 headlines:

• Except for Venezuela which has no official FMD status with the OIE, there have been no suspected cases of FMD anywhere in South America during 2021.

3.7.1 Vaccine recommendations for Pool 7

- Internationally produced vaccines:
 - All vaccines used in the region are produced in South America (Argentina, Brazil, Colombia, Paraguay & Venezuela have vaccine manufacturers)
- Locally produced vaccines
 - o O: O1 Campos
 - o A: A₂₄ Cruzeiro, A/Arg/2001*
 - o C: C₃ Indaial*

*Only Argentina uses A/Arg/2001 and C_3 Indaial.



4 Improving the quality of laboratory tests from FMD reference laboratories

4.1 Proficiency testing schemes (PTS) organised by the Network Partners

Brazil

- Proficiency testing scheme organised during 2021:
 - 25 laboratories from 17 countries were invited to participate (Argentina, Bolivia Brazil, Canada, Chile, Colombia Ecuador, United States, Guyana Mexico, Panama, Paraguay, Peru, Surinam Trinidad and Tobago, Uruguay and Venezuela)
 - \circ 22 laboratories confirmed participation.
 - The tests aim to detect FMDV or VSV
 - Molecular biology techniques (RT qPCR, RT PCR and sequencing)
 - Antigen Detection Techniques (ELISA LPBE)
 - Serological techniques (ELISA 3 ABC EITB and viral neutralization)
 - The interlaboratory is in process and is currently in the phase of execution of the techniques by the national laboratories

China

- National PT for major animal disease organized by CADC and FMDRL during 2021
 - Funded by MARA, China.
 - 50 sets of FMD (serotypes O and A) blind samples prepared and provided by FMDRL for FMDV typing real-time RT-PCR test.

France

- FMD/SVD Proficiency testing scheme 2021
 - Objective: to evaluate the existing ability of each laboratory to diagnose FMD/SVD outbreaks using virological and serological methods.
 - o 42 participants from 40 countries
 - 26 EU member states (EURL funding)
 - 5 candidate countries (EURL funding)
 - 6 countries from SEE (EuFMD funding)
 - 3 third countries (self funding)

Italy

- National Proficiency Testing Scheme organised for regional laboratories in Italy.
 - Objective: To practise the regional laboratories in the use of serological and molecular tests for maintaining preparedness in case of national emergency
 - Completion of Proficiency testing scheme 2020
 - Molecular test Panel: 3 samples (inactivated virus FMDV type O). Test performed: Real Time RT-PCR (3D)
 - Serological test Panel: 20 sera (naive and positive against FMDV type Asia1). Test performed: SP-ELISA type Asia1



- Proficiency testing scheme 2021
 - Molecular test Panel: 3 samples (inactivated virus FMDV). Test: Real Time RT-PCR (3D)
 - Serological test Panel: on the same panel used for the RT 2020. Test performed: 3ABC trapping ELISA
 - Dead Line: 30th of January 2022

Republic of Korea

- Proficiency testing scheme 2021:
 - 46 Regional Diagnostic Laboratories
 - Virological test Panel: 6 samples (inactivated virus FMDV). Test: RT-PCR and real time RT-PCR
 - Serological test Panel: 6 sera (Sero-positive and negative against type O) Test performed: Two types of NSP ELISA and One type of SP-O ELISA
- Objective: to evaluate the existing ability of each laboratory to diagnose FMD outbreaks using virological and serological methods

Russian Federation

• LPB ELISA for 6 labs from Kyrgyzstan, Kazakhstan, Belarus and Armenia

United Kingdom

Phase XXXIII (2021)					
Total invited laboratories ¹	75				
Total number of shipments ¹	44				
	EUFMD funded participants				
Participants from Global Network	Argentina, Botswana, Brazil, C	anada³, China, Ethiopia, Kenya,			
Labs ²	Nigeria, Russia, So	uth Africa, Thailand.			
	Cat-1	0 %			
% of labs meeting target	Cat-2	20 %			
performance ⁴	Cat-3	20 %			
	Cat-4	60 %			
Participants from EuFMD Member states (non-EU)	Georgi	Georgia, Israel.			
	Cat-1	0 %			
% of labs meeting target	Cat-2	0 %			
performance ⁴	Cat-3	50 %			
	Cat-4	50 %			
Participants from neighbourhood countries	Algeria, Armenia, Azerbaijar	n, Jordan, Lebanon, Morocco			
	Cat-1	0 %			
% of labs meeting target	Cat-2	0 %			
performance ⁴	Cat-3	16.67 %			
	Cat-4	83.3 %			
	Self-funded participants				
Participants	Australia, Namibia, Nepal, New Ze	aland, Republic of Korea, Senegal,			
	Singapor	e, Taiwan			
	Cat-1	0 %			
% of labs meeting target	Cat-2	0 %			
performance ⁴	Cat-3	0 %			
	Cat-4	100 %			



¹ Additional laboratories (non-NRL) participate in the PTS at their own expense;

² Not including IZSLER and Sciensano who participate as European NRLs;

³ is self-funded;

⁴ Scored according criteria agreed by the NRLs within Europe, each laboratory receives a personalized anonymous feedback letter to highlight areas in which they could improve, and performance of each laboratory is broadly categorized into one of four groups: (**Category 1**) to emphasize critical issues where immediate action is required that impact upon the laboratory to correctly identify FMD virus (virology tests) or FMDV infected animals (serological tests), (**Category 2**) laboratories with serious issues with the performance of individual tests that need to be addressed, (**Category 3**) to record additional observations which may need to be considered by the laboratory to improve the local performance of individual tests and (**Category 4**) laboratories whose tests which are fit for purpose and where no further action is required.

4.2 Supply of reagents

Belgium

Type of reagent	Quantity	Recipient of the reagent (Laboratories/Countries)
FMD virus	6 virus strains	Joint EU-RL partner ANSES (France)
FMD antibody positive serum	720 ml serum in total	Joint EU-RL partner ANSES (France)
FMD virus	8 virus strains	AGES (NRL Austria)
FMD antibody positive serum	Reference serum corresponding to each of the 8 virus strains and NSP positive serum	AGES (NRL Austria)

Canada

Type of reagent	Quantity	Recipient of the reagent (Laboratories/Countries)
Pre-coated FMDV 3ABC cELISA plates		Canadian Animal Health Surveillance Network (CAHSN) laboratories
FMD 3ABC cELISA panels		CAHSN laboratories
FMD rRT-PCR panels		CAHSN laboratories
Hybridomas secreting monoclonal antibodies to FMDV serotypes A, O, SAT 1, 2, 3 and one that recognizes all 7 serotypes. These were used in the production of prototype lateral flow strips for FMD antigen detection.	2mL each	Mologics LTD, United Kingdom.
Purified monoclonal antibodies to FMDV serotypes A, O, SAT 1, 2, 3 and one that recognizes all 7 serotypes. These were used in the production of prototype lateral flow strips for FMD antigen detection.	Various	Mologics LTD, United Kingdom.



Type of reagent	Quantity	Recipient of the reagent (Laboratories/Countries)
Hybridomas secreting monoclonal antibodies to FMDV serotypes Asia 1 and O. These were used in the production and commercialization of serotype-specific solid phase competitive ELISA (SPCE) kits.	Various	BioStone Animal Health LLC, United States of America.
Monoclonal antibodies to FMDV serotypes Asia 1 and O. These were used in optimization of SPCE.	Various	BioStone Animal Health LLC, United States of America.
Reagents for FMD 3ABC blocking ELISA (recombinant antigen, conjugated monoclonal antibody and positive control sera). For the development of in-house diagnostic capacity	Various	The National Veterinary Research Institute (NVRI), Vom, Nigeria.
Reagents for FMD serotypes A, O, SAT1 and 2 SPCE (BEI inactivated antigens, rabbit and guinea pig anti- FMDV polyclonal sera, positive control sera). For the development of in-house diagnostic capacity	Various	The National Veterinary Research Institute (NVRI), Vom, Nigeria.
Reagents for FMDV 3D and beta-actin real-time RT-PCR (primers, probes, positive controls mastermixes). For the development of in-house diagnostic capacity	Various	The National Veterinary Research Institute (NVRI), Vom, Nigeria.

China

Type of reagent	Quantity	Recipient of the reagent (Laboratories/Countries)
LPB-ELISA (type O/A/Asia1)	20624	Veterinary laboratories and large-scale
NSP-3ABC-ELISA	1618	breeding companies, China
SPC-ELISA	583	
Conventional Multi-RT-PCR	42	
Real-time RT-PCR	705	Provincial veterinary laboratory in China
Typing real-time RT-PCR	261	

France

Type of reagent	Quantity	Recipient of the reagent (Laboratories/Countries)
NSP ELISA	8 kits	Mauritius



Type of reagent	Quantity	Recipient of the reagent (Laboratories/Countries)
Type O ELISA	6 kits	Mauritius
Master Mix duplex rtRT-PCR	300 reactions	Jordan
Primers for FMDV typing	200 reactions	Nigeria

Italy

Type of reagent		Quantity
FMDV antigen detection ELISA type O, A, C, Asia1, SAT1-2		145
NSP Ab ELISA KIT (3ABC)		35
SP-Antibody ELISA Kit	FMDV O	87
	FMDV A	81
	FMDV Asia1	55
	FMDV SAT 1	8
	FMDV SAT 2	16

• In 2021 saw the minimum number of the kits distributed since 2014. A decrease of about 20% compared with 2020 of the countries that requested kits.

• In 2021 the requests for FMDV antigen detection ELISA kits has is increased by about 30% compared to 2020.

Republic of Korea

Type of Reagent	Quantity	Recipient of the reagent (Lab / Countries)
FMDV Rapid kits for Pan, serotype	150 tests	
rRT-PCR for serotype O, A or	192 tests	
rRT-PCR for genotype O/PanAsia,	192 tests	Bangladesh
rRT-PCR for genotype A/Sea-97 or	192 tests	

Russian Federation

Type of reagent	Quantity	Recipient of the reagent (Laboratories / Countries)
LPB ELISA	2054	Russia, Belarus & Kyrgyzstan
Ag ELISA	6	Russia & Kyrgyzstan
FMD RT-PCR-RT	10	Russia



Type of reagent	Quantity	antity Recipient of the reagent	
		(Laboratories / Countries)	
NSP-ELISA	65	Russia	

Thailand

Type of reagents	Supplied nationally and own lab	Remarks
Rabbit type O	18	-
Rabbit type A	16	
Rabbit type Asia1	15	
Guinea pig type O	20	
Guinea pig type A	24	
Guinea pig type Asia1	21	
Antigen Type O	15	
Antigen Type A	10	
Antigen Type Asia 1	12	
Control serum c++, c+, c-	11	

United Kingdom

Type of reagent	Quantity	Recipient of the reagent
		(Laboratories / Countries)
Antigen (Serotypes: O, A, Asia-1, SAT 1, SAT 2 & SAT 3)	96 vials	Republic of Korea, USA & UK
Antisera (Serotypes: O, A, Asia-1, SAT 1, SAT 2 & SAT 3)	41 vials	Italy & UK
Controls (Serotypes: O, A, C, Asia-1, SAT 1, SAT 2 & SAT 3)	39 vials	France, Republic of Korea & Taiwan
NSP controls	5 vials	Romania
NSP panel	80 vials	Taiwan
Antigen and antisera (Serotypes: O, A, C, SAT 1, SAT 2 & SAT 3)	336 vials	Botswana & Poland
Controls and NSP controls (Serotypes: O, A, & Asia-1)	47 vials	Republic of Korea



Type of reagent	Quantity	Recipient of the reagent (Laboratories /
		Countries)
Antigen, antisera and controls (Serotypes: O, A & Asia-1)	958 vials	Iraq & Vietnam
Virus Isolates	159 aliquots	Argentina, Brazil, France, Germany, Israel, Republic of Korea, Russia, Turkey & United Kingdom

4.3 Training courses organised by Network partners

Canada

• Foreign Animal Disease Recognition course for Canadian Veterinarians

China

- 2 national training courses on FMD diagnostic techniques and control & prevention (Tianshui, Gansu).
- 14 reports or seminars at workshops organized by provincial labs, China.
- 2 Biosafety trainings held in LVRI, China.
- Real time on field training (sampling during active surveillance).
- •

Republic of Korea

Type of technical training provided	Country of origin of the expert(s) provided with training	No. participants from the corresponding country
	Malaysia	9
	Vietnam	9
Seminars	Indonesia	8
	Kazakhstan	5
	Philippines	3
	Sri Lanka	1

Russian Federation

- Three hands-on training courses
 - o 2: Russia (4 participants)
 - o 1: Pakistan (3 participants)


4.4 Collaborative projects

Australia

Collaborators	Purpose of collaboration	Outcomes
FLI, Germany	Safe transport of epithelium samples infected with FMD virus	 Investigate whether FMDV in epithelium tissue will be inactivated using commercially available buffers Determine if virus can be recovered from the genetic material to allow further investigation under suitable biocontainment levels
FLI, Germany	Understanding virus survival at different temps and humidity to aid accurate modelling	

Belgium

Collaborators	Purpose of collaboration	Outcomes
BVI, Botswana	bilateral collaboration	Participation of Sciensano in PT organised by BVI in 2021
LNV, Burundi	bilateral collaboration	on hold due to Covid restrictions – resumed in 2022
NVRI, Nigeria	bilateral collaboration	on hold due to Covid restrictions – to be resumed
KULeuven, Belgium (promoter); Nairobi University, Kenya; Addis Ababa University, Ethiopia	JOINT international networking project	on hold due to Covid restrictions – resumed in 2022

Brazil

Collaborators	Purpose of collaboration	Outcomes
NCFAD, Canada	Whole genome sequencing of the 95 isolates of FMDV that are under Panaftosa custody, relative to the period from 1950 to 2018	understaning the evolutionary history of these strains of FMDV serotypes O and A and identifying molecular epidemiological patterns.



Canada

Collaborators	Purpose of collaboration	Outcomes
Botswana Institute for Technology Research and Innovation (BITRI), Botswana; Botswana Vaccine Institute (BVI), Botswana; The Pirbright Institute, UK	Development of lateral flow strip test for FMDV antigen detection and serotyping	Lateral flow strip test for FMDV serotypes A, O, SAT 1, 2, 3 and panserotype
National Veterinary Research Institute (NVRI), Nigeria	Capacity building for National and Regional Foot-and-Mouth- Disease Control Strategy in Nigeria	Enhanced capacity for FMDV diagnosis, characterization of circulating FMDV serotypes
Boehringer Ingelheim	Determination of the 50% protective dose (PD ₅₀) of FMD vaccines in pigs	Data on potency of FMD vaccines in pigs
Animal and Plant Quarantine Agency, South Korea	Comparative studies for Foot and Mouth Disease virus diagnostics and vaccines in Cattle	Validated FMD diagnostic kits, efficacy of FMD vaccines in cattle
University of Calgary, Alberta, Canada	Application of the split TreA enzyme assay for detection of antibodies to FMDV Non- Structural Proteins (3ABC)	Rapid FMD 3ABC antibody detection assay
Shaddari Inc., Montreal, QC, Canada	Computational tool for foot- and-mouth disease vaccine matching	A vaccine matching tool that can rely on either VNT data or FMDV P1 sequences
The Pirbright Institute, UK	Point-of-care test for FMDV detection	Validated strip test for FMDV antigen detection

China

Collaborators	Purpose of collaboration	Outcomes
Japan; Korea	Prevention and control of TADs in China- Japan-Korea	exchange of information
Mongolia; Russia	prevention and control of TADs, such as FMD, PPR, HPAI	exchange of information
Korea Atomic Energy Research Institute	Research and development of FMD virus like particle (VLP)	Immune optimization and mechanism of targeting dendritic cells with FMD VLPs



Italy

Collaborators	Purpose of collaboration	Outcomes	
The Pirbright Institute, UK	Research agreement aimed to continuous validation and improvement of diagnostic kits (ELISA), new developments	 Continued development of antigen detection assays for FMDV and other vesicular diseases Development of assays for FMD vaccine quality Validation of a novel universal serology assay for diagnosis of FMD Research activities in the framework of AgResult project 	
EUFMD and Trancaucasus countries	Field trial to estimate effectiveness of vaccination program against FMD in Transcaucasian countries – Azerbaijan	 framework of AgResult project VNT testing on sera from vaccinated animals, collected sequentially after first and second vaccination. Obtained results are under evaluation Overall, data appear to be similar with those obtained in the neighboring countries (Georgia and Armenia) 	
University of Turin, Italy; The Pirbright Institute, UK; SAP Institute, Turkey; FMDV-NRL, Embakasi, Kenya	Development of multiplex LFD for FMDV serotyping in field conditions	 LFA 1 (pan-FMDV and simultaneous serotyping of O, A, Asia 1) performed similarly to the IZSLER/Pirbright MAb ELISA kit. LFA 2 prototype (pan-FMDV and simultaneous serotyping of SAT 1 & 2) validation on 24 clinical samples sensitivity is lower than Ag ELISA 	

Republic of Korea

Collaborators	Purpose of collaboration	Outcomes
NCVD, Vietnam	To carry out comparative studies of Avian influenza virus and Foot-and- mouth disease virus between Korea and Vietnam	Data and materials (2016-2024)



Collaborators	Purpose of collaboration	Outcomes
NAHPRI, Cambodia; NAHL, Lao PDR	To study on genetic characterization of foot-and-mouth disease viruses and avian influenza virus in FMD and AI endemic countries (Cambodia and LAO PDR)	Data and materials (2018-2022)
CDIL, Bangladesh	To study on genetic characterization of Foot-and-mouth disease viruses in Bangladesh	Data and materials (2020~2024)
NCFAD, Canada	Collaborative validation studies of solid phase competitive enzyme-linked immunosorbent assay and rapid detection kits for antibodies to NSPs for FMDV	Data and materials (2019-2021)
The Pirbright Institute, UK	To carry out a collaborative research project on Molecular epidemiology and NGS platform studies on Foot-and- mouth disease virus (FMDV) between APQA, Korea and WRLFMD, United Kingdom	Data and materials (2020-2021)

Russian Federation

Collaborators	Purpose of collaboration	Outcomes
Mongolia	Assessment of immunity level in animals vaccinated against FMD and detection of possible virus circulation in zones where vaccination is practiced (at the stage of signing)	Eradication of highly dangerous diseases including FMD in Mongolian Livestock. Continues to 2022
China, People's Republic of; Mongolia	Agreement on cross-border trade and TADs risk reduction between China, Mongolia and Russia	Interactions in case of emergencies associated with dangerous animal diseases including FMD
Armenia; Azerbaijan; Georgia; Iran; Turkey	Cooperation on the prevention and control of foot and mouth disease and other transboundary animal diseases between the countries of the Caucasus, Russia and Iran (GF- TADs)	Exchange of information on outbreaks of diseases, vaccination of animals
	Joint CIS measures for FMD prevention and control	FMD prevention and control Continues to 2025



Thailand

Collaborators	Purpose of collaboration	Outcomes
SATREPS Project (JICA)	Differential diagnosis systems of foot-and-mouth disease (FMD) and similar vesicular diseases	Immunochromatography test kits research work
WRLFMD, UK.	Reference control serum for in- house reagent preparation plan	 FMD vaccine production (Animal facility) WRLFMD (Supporting and recommendation)

Turkey

Collaborators	Purpose of collaboration	Outcomes
ANSES;	ICRAD ERA-NET PROJECT	In progress
SLU; FLI; SCIENSANO	From proteogenomic host response signatures of persistent foot-and-mouth disease virus (FMDV) infection to diagnostic markers and therapeutic control	

United Kingdom

Collaborators	Collaborative project	Outcomes
AU-PANVAC	OIE Twinning Project	Vaccine QA/QC for Africa
Sciensano, Belgium; Lelystad, Netherlands; IZSLER, Italy	Calibration of VNT methods	To compare VNT methods used in different laboratories
IZSLER, Italy	Development of FMD ELISA and LFD tests	On-going new ELISA tests for FMD diagnosis
KSRVI, Kazakhstan	Serological assays for FMD	Post-vaccination testing and surveillance

4.5 *In vivo* potency studies undertaken during 2021

Argentina

Purpose of study	Study design	Name of Vaccine strain	Name of Challenge strain	Result
Vaccine control for certification	PD ₅₀	 Trivalent Scheme O₁ Campos, A24 Cruzeiro, A Argentina 2001 Full dose; IM 	O/SKR/1/19	8.00
	PD ₅₀	 Trivalent Scheme O1 Campos, A24 Cruzeiro, A Argentina 2001 Reduced dose; ID 	O/SKR/1/19	7.49



	Trivalent Scheme	O/SKD/04 vdm	
PD50	 O1 Campos, A24 Cruzeiro, A Argentina 2001 		6.24
	· · · · · · · · · · · · · · · · · · ·	Jincheon 2014	

reduced dose; ID

Brazil

		30 d after v	accination	365 d after vaccination			
Country of origin	Serotype	Average of protective titres FMDV specific antibody	Average of unprotective titres FMDV specific antibody	Average of protective titres FMDV specific antibody	Average of unprotective titres FMDV specific antibody		
Brazil	A24 Cruzeiro	3.29 (334/347)	0.96 (12/347)	2.94 (322/347)	1.04 (25/347)		
Brazil	O1 Campos	3.01 (96/110) *	1.3 (14/110) *	2.66 (164/347)	1.23 (183/347)		
* in progre	SS						

Canada

Efficacy of a bivalent (O₁ Campos and A24 Cruzeiro/A Argentina 2001) Foot-and-mouth disease (FMD) vaccine against challenge with FMDV A/ASIA/SEA-97 in cattle.

Republic of Korea

Purpose of study	Study design	Vaccine strain used in Korea	Challenge strain	Result
	Two doses (Vaccinated	O1 Manisa + O3039		Protection (3/3)
	with 2 weeks interval and challenged at 7	O/RUS/Primorsky/20 14 (O/SEA/Mya-98)	O/VIT/30/2017 (O/Cathay)	Protection (4/4)
	vaccination)	O1 Campos		Protection (4/4)
	One dose (Challenged at 28 dpv)	A22 Iraq		Protection (4/4)
Research study on		A/RUS/Zabaikalsky/2 013 (A/ASIA/Sea-97)	A/BHU/03/2017 (A/G-\/II)	Protection (2/4)
pigs		A24 Cruzeiro + A/ARG/2001	(700 vii)	Protection (4/4)
		A22 Iraq		Protection (1/4) *
	One dose	A/RUS/Zabaikalsky/2 013 (A/ASIA/Sea-97)	A/TUR/13/2017 (A/G-\/II)	Protection (1/3) *
	(Ghanenged at 20 upv)	A24 Cruzeiro + A/ARG/2001	(///0-/11)	Protection (4/4)

* Mostly minor clinical signs were observed.

China

Purpose of study	Study design (i.e., PD50, PGP etc)	Name of Vaccine strain	Name of Challenge strain	Result



Immune potency pre-test	Percentage of	Re-O (vaccine strain)		10/10 protective
Immune potency pre-test	protection (10 pigs vaccinated, challenged 28dpv)	Re-O/17002 (alternative vaccine strain)	O/Mya-98 (O/2021008)	9/(10-1) protective
ID 50	10-3,10-4,10-5,10-6 (4 pigs per dilution)		O/CATHAY (O/19031)	ID ₅₀ =4.5

Kenya

Purpose of study	Study design	Name of Vaccine strain	Name of Challenge strain	Result
Vaccine lot Potency test	PD50	O K77/78	O K77/78	20.2
Vaccine lot Potency test	PD50	A K5/80	A K5/80	29.5
Vaccine lot Potency test	PD50	A K5/80	A K5/80	6.6
Vaccine lot Potency test	PD50	SAT2 K52/84	SAT2 K52/84	20.2
Vaccine lot Potency test	PD50	SAT2 K52/84	SAT2 K52/84	29.5



Appendix 1 Details of clinical samples from field cases from countries in FMDV endemic regions tested during 2021

Laboratory	Samples from	Total	0	А	ပ	Asia 1	Sat 1	Sat 2	Sat 3	Untyped	NVD	Comments
ANSES	Burkina Faso	12	-	-	-	-	-	6	-	5	1	
	Mauritius	18	2	-	-	-	-	-	-	-	16	
	Niger	66	-	18	-	-	-	1	-	8	39	
APQA	Bangladesh	20	18	2								
	Cambodia	35	25	4	-	-	-	-	-	6	-	
	Vietnam	8	6	-	-	-	-	-	-	-	2	
ARC-OVI	South Africa	42	-	-	-	-	-	2	15	-	25	
	Eswatini	8	-	-	-	-	-	-	-	-	8	
BVI	Namibia	18	7	-	-	-		4	-	-	7	
	Malawi	5	-	-	-	-	1	-	-	-	4	
	Botswana	4	-	-	-	-	-	-	-	-	4	
	Zambia	16	4	-	-	-	-	2	-	-	10	
	Eswatini	8	-	-	-	-	-	-	-	-	8	
CSIRO	-	-	-	-	-	-	-	-	-	-	-	
FADDL	-	-	-	-	-	-	-	-	-	-	-	
FGI-ARRIAH	Russia	133	2	-	-	-	-	-	-	-	131	
FMD laboratory	Kenya	98	37	7	-	-	1	17			36	
ICAR- Directorate of FMD	-	-	-	-	-	-	-	-	-	-	-	
LNERV	Senegal	-	-	-	-	-	-	-	-	-	-	
LVRI	China	17	4	-	-	-	-	-	-	-	13	
NAHDIC	Ethiopia	112	22	-	-	-	7	24	-	8	61	
NCFAD	Nigeria	177	103	22	-	-	-	24	-	20	8	
NVRI	Nigeria	24	9	-	-	-	-	7	-	8	-	
PANAFTOSA	-	-	-	-	-	-	-	-	-	-	-	
RRLSEA	Thailand	75	17	34	-	-	-	-	-	17	7	
	PDR Lao	20	-	-	-	-	-	-	-	-	20	
Şap Institute	Iran	25	14	7	-	3	-	-	-	-	1	
	Turkey	120	64	-	-	-	-	-	-	6	50	
SENASA	Argentina	22	-	-	-	-	-	-	-	-	22	
WRLFMD	Bahrain	40	3	1	-	-	-	-	-	1	35	
	DR of Congo	63	15	-	-	-	-	-	-	23	25	
	Iran	50	22	18	-	1	-	-	-	9	-	
	Israel	6	6	-	-	-	-	-	-	-	-	
	Jordan	13	7	-	-	-	-	-	-	4	2	
	Kenva	20	13	1	-	-	2	-	-	3	1	
	Mongolia	_0 16	4	-	-	-	-	-	-	5	12	
	Nepal	70	38	1	-	-	-	-	-	16	15	



Laboratory	Samples from	Total	0	А	C	Asia 1	Sat 1	Sat 2	Sat 3	Untyped	NVD	Comments
	Nigeria	137	53	28	-	-	-	20	-	26	11	One sample dually infected with O and SAT 2
	Uganda	14	4	-	-	-	-	-	-	4	6	
	Vietnam	16	16	-	-	-	-	-	-			
	Zambia	97	33	-	-	-	-	7	1	17	39	



Appendix 2 Vaccine matching studies undertaken by Network partners during 2021

Vaccine efficacy is influenced by both vaccine potency and vaccine match and it is possible that a poor match may to some extent be compensated by high potency vaccines and by administering more than one dose at suitable intervals. The use of oil adjuvant is also expected to improve efficacy. Thus, a vaccine with a weak antigenic match to a field isolate, as determined by serology, may nevertheless afford some protection if it is of sufficiently high potency. Therefore, in the absence of a good match, or where the match is unknown, vaccines of high potency should preferably be used. The r_1 values shown below, represent the one way serological match between vaccine strain and field isolate, calculated from the comparative reactivity of an antiserum, raised against the vaccine in question, to the vaccine virus and the field isolate.

Key:



For VNT:

 $r_1 \ge 0.3 -$ suggest that there is a close relationship between field isolate and vaccine strain. A potent vaccine containing the vaccine strain is likely to confer protection

 $r_1 \le 0.3$ - suggest that the field isolate is so different from the vaccine strain that the vaccine is unlikely to protect.

For LB-ELISA:

 $r_1 \ge 0.4 -$ suggest that there is a close relationship between field isolate and vaccine strain. A potent vaccine containing the vaccine strain is likely to confer protection

 $r_1 \le 0.4$ - suggest that the field isolate is so different from the vaccine strain that the vaccine is unlikely to protect.



Argentina

		Vaccine strain	
	O₁ Campos Highpot Sera Pig	A 24 Cruzeiro	O₁ Campos/A 24 Cruzeiro/A Argentina 2001
O VTN 6/18 cathay	0.37		
O SKR 1/19 ind2001	0.63		
O VTN 10/18 mesa oanasia	0.57		
O.VTN/1/19 sea mya98	0.65		
A/SKR/2/10 AsiaSEA97		POOR MATCH	0.98
A/SKR/3/17 AsiaSEA97		POOR MATCH	0.77
A/SKR/4/18 AsiaSEA97		POOR MATCH	0.89

Botswana

	R1 value per Vac	cine virus strain
	O/Manisa	O/3039
NAM 14/2021	0.52	0.38

China

Field is alata	lineage	onimal	Vaccine Strain			
Field ISolate	imeage	ammai	O/BY/2010	Re-O		
2021-008	Mya-98	pig	Ν	М		
2021-A011	Ind-2001	cattle	М	Μ		
2021-A052	CATHAY	pig	М	Μ		
2021-021	CATHAY	pig	Ν	nd		
2020-A136	Ind-2001	cattle	М	Μ		

India

Serotype	Strain	Genotype/Topotype	Lineage
0	INDR2/1975	ME-SA	Branch B
А	IND40/2000	Genotype 18	Non-deletion
Asia1	IND63/1972	Genotype 1	Lineage B



Republic of Korea									
Field vir	us (type O)	Matching with Vaccine strain (Matching rate)							
Topotype/ Genotype	Vietnam 2016-18		01/Manisa	O/3039	O/RUS/ Primorsky/2014	O1/Campos	Om-O/PanAsia2 (Korean Vaccine candidate)	O/BE/SKR/2017 (Korean Vaccine candidate)	
	Vietnam 2016-18	4	100%	75%	25%	0%	25%	100%	
O/ME-SA/PanAsia	Cambodia 2018	2	100%	100%	0%	50%	100%	100%	
	Laos 2018	1	100%	100%	100%	100%	0%	100%	
O/ME-SA/Ind-2001d	Laos 2017	1	100%	100%	100%	100%	100%	100%	
O/SEA/Mya98	Vietnam 2015-17	2	100%	100%	50%	50%	0%	100%	
O/Cathay	Vietnam 2017	1	100%	0%	0%	100%	0%	0%	

Field	virus (type A)		Matchii strain	Matching with Vaccine strain (Matching rate)				
Topotype/ Genotype	Country/Year	Number of samples	01/Manisa	O/3039	O/RUS/ Primorsky/2014			
	Vietnam 2015	7	33.3%	0%	33.3%			
A/Asia/Sea-97	Cambodia 2019	3	66.7%	0%	66.7%			
	Laos 2018	1	0%	0%	100%			
	Turkey 2017	1	0%	100%	0%			
	Israel 2017	1	0%	0%	0%			
A/ASId/G-VII	Bhutan 2017	1	0%	0%	0%			
	Iran 2018	1	0%	0%	0%			

South Africa

Isolate	Vaccine strain (SAR 3/04/2)
SAR 15/13/2	0.75
KNP 12/08/2	0.77
ZIM 2/12/2	0.65



SAR 1/01/2	0.69
SAR 1/01/2	0.71
SAR 1/03/2	0.63

Thailand

		r-value [O/189/87 (Thai vaccine strain)]										
	Country		LF	LPB ELISA			VNT		VP1 Genotyping			
Year		Number	0-0.19	0.2-0.39	≥ 0.4	Number	<0.3	≥ <i>0</i> .3	Sequence			
2021	Thailand	9	1	1	7	1	-	1	Me-SA/Ind2001e			

	[A/S	r-value [A/Sakolnakorn/97 (Thai vaccine strain)]									
			LF	LPB ELISA			VNT		VP1 Constyning		
Year	Country	Number	0-0.19	0.2-0.39	≥ 0.4	Number	<0.3	≥ <i>0</i> .3	Sequence		
2021	Thailand	7	0	1	6	7	1	6	ASIA/Sea-97		

	[A	r-value [A/Lopburi/2012 (Thai vaccine strain)]									
			LF	B ELI	SA	VNT			VP1 Genetyping		
Year	Country	Number	0-0.19	0.2-0.39	≥ 0.4	Number	<0.3	≥0.3	Sequence		
2021	Thailand	7	0	6	1	7	6	1	ASIA/Sea-97		

Turkey

	Serotype A			a	۲ - ۱	4	~	90
Isolate	Genotype	Lineage	A/GVII	A22/IRG	A/IRN/1 (A05SIS 13)	A/TUR/1 (A05)	A/TUR/1 (A05)	A/TUR/0 (A05)
Irn 20/21	A-05	FAR-11	0	0	0.13			
Irn 16/21	A-05	FAR-11	0	0	0.13			
Irn 2/21	A-05	FAR-11	0	0	0.13	0.11	0.10	0.15
Irn 25/21	A-05	FAR-11			0.23	0.07	0.3	0.27



	Serotype	07	18	sa	
Isolate	Topotype	Lineage	ο/Τυκ/	O/TUR/	O1 Mani
Irn 19/21	ME-SA	PanAsia-2 ^{ANT-10}	>1	0.04	>1

United Kingdom

Note:

Ν	No Match (r ₁ < 0.3)
М	Match (r ₁ ≥ 0.30)

Isolate	Serotype O		O 3 Boeh Ingel	O 3039 Boehringer Ingelheim		O Campos Boehringer Ingelheim		O Manisa Boehringer Ingelheim		O/TUR/5/09 MSD		O1 Campos Biogénesis Bagó		O Panasia 2 Boehringer Ingelheim	
	Topotype	Lineage	<i>r</i> 1	Titre	<i>r</i> 1	Titre	<i>r</i> 1	Titre	<i>r</i> 1	Titre	<i>r</i> 1	Titre	<i>r</i> 1	Titre	
KEN/10/2020	EA-2	-	0.45	1.79	NT	-	0.4	2.04	0.5	1.98	0.35	2.35	0.34	2.01	
KEN/6/2021	EA-2	-	0.74	2.01	NT		0.74	2.31	0.85	2.21	0.51	2.51	0.43	2.11	
ZAM/2/2020	EA-2	-	0.4	1.7	NT		0.26	1.92	0.46	1.97	0.47	2.43	0.28	1.95	
ZAM/7/2021	EA-2	-	0.59	1.87	NT		0.47	2.17	1	2.35	1	2.81	0.68	2.33	
BAR/18/2021	EA-3	-	0.54	1.78	NT	-	0.49	2.15	0.65	2.31	0.54	2.52	NT	-	
BAR/20/2021	EA-3	-	0.62	1.84	NT		0.5	2.16	0.56	2.25	0.63	2.59	NT		
ISR/15/2017	EA-3	-	NT		NT		NT		NT		0.21	2.13	NT		
ISR/1/2021	ME-SA	Ind-2001	0.35	1.63			0.36	1.92	0.85	2.21	0.52	2.57	NT	-	
ISR/5/2021	ME-SA	Ind-2001	0.48	1.77			0.46	2.02	1	2.34	0.76	2.73	NT		
LAO/1/2020	ME-SA	Ind-2001	0.65	1.92	0.28	1.81	0.47	2.1	0.5	2.07	0.48	2.48	NT		
LAO/5/2020	ME-SA	Ind-2001	0.39	1.7	0.32	1.87	0.62	2.22	0.76	2.25	0.55	2.54	NT		
MOG/2/2021	ME-SA	Ind-2001	0.69	1.94	0.33	2.2	0.51	2.29	1	2.37	0.65	2.61	NT		
MOG/3/2021	ME-SA	Ind-2001	0.62	1.89	0.33	2.2	0.52	2.3	0.87	2.22	0.6	2.58	NT		
TAI/12/2020	ME-SA	Ind-2001	0.47	1.74	0.26	1.79	0.32	1.94	0.54	2.12	0.4	2.32	NT		
TAI/9/2020	ME-SA	Ind-2001	0.58	1.83	0.34	1.9	0.51	2.15	0.6	2.17	0.65	2.57	NT		
VIT/1/2021	ME-SA	Ind-2001	0.38	1.53	NT		0.29	0.98	0.72	2.16	0.45	2.33	NT		
VIT/21/2020	ME-SA	Ind-2001	0.59	1.79	NT		0.3	2.06	0.54	2.12	0.38	2.42	NT		
VIT/24/2020	ME-SA	Ind-2001	0.39	1.61	NT	-	0.2	1.89	0.42	2.01	0.37	2.41	NT	-	
CAM/1/2018	ME-SA	PanAsia	0.33	1.63	0.25	1.91	0.29	1.91	0.46	2.01	0.43	2.37	NT	-	
CAM/6/2018	ME-SA	PanAsia	0.4	1.71	0.32	2.05	0.33	1.97	0.59	2.12	0.49	2.43	NT	-	
IRN/3/2021	ME-SA	PanAsia-2	0.58	1.84	NT	-	0.22	1.95	0.63	2.12	0.59		0.26	2.02	
IRN/7/2021	ME-SA	PanAsia-2	0.5	1.78	NT		0.31	1.94	0.13		0.48	2.46	0.22	1.95	
ISR/12/2019	ME-SA	PanAsia-2	NT		NT		NT		NT	-	0.29	2.28	NT	-	
ISR/27/2019	ME-SA	PanAsia-2	NT	-	NT	-	NT	-	NT	-	0.42	2.25	NT	-	
VIT/47/2018	SEA	Mya-98	0.34	1.48	NT	-	0.14	1.67	0.3	1.78	0.49	2.37	NT	-	



	Seroty	vpe A	A I 20	ran 05	A Sa	audi 95	A/TUR/	20/06	A Ma 9	laysia 7	A22	Iraq	A Eri 9	itrea 8	A G 20	IVII 15
Isolate			Boeh	ringer heim	Boe Inge	hringer elheim	MSD A Hea	nimal lth	Boehi Ingel	ringer heim	Boehi Ingel	ringer heim	Boehi Ingel	ringer heim	Boehi Ingel	ringer heim
	Topotype	Lineage	<i>r</i> ₁	Titre	<i>r</i> ₁	Titre	<i>r</i> ₁	Titre	<i>r</i> ₁	Titre	<i>r</i> ₁	Titre	<i>r</i> ₁	Titre	<i>r</i> ₁	Titre
BAR/21/2021	Africa	G-I	0	-	0.35	1.98	0	-	NT	-	0.31	1.94	0.12	1.69	0.39	1.58
KEN/10/2021	Africa	G-I	0.08	1.48	0.25	1.82	0.08		NT	-	0.27	2.11	0.11	1.71	0	-
NIG/1/2017	Africa	G-IV	0	-	0.16	1.72	0.15	1	0.12	1.44	0.18	1.75	0.17	1.86	NT	-
NIG/3/2017	Africa	G-IV	0.04	1.11	0.37	2.08	0.21	1.15	0.17	1.58	0.23	1.86	0.15	1.82	NT	
NIG/5/2015	Africa	G-IV	0.05	1.17	0.35	2.05	0.2	1.13	0.19	1.63	0.3	1.98	0.21	1.95	NT	-
IRN/18/2021	Asia	Iran-05	0.39	2.2	NT	-	0.23	1.25	NT	-	0.14	1.81	NT	-	0	-
IRN/22/2021	Asia	Iran-05	0.28	2.05	NT	-	0.36	1.44	NT	-	0.32	2.17	NT	-	0	-
TAI/10/2019	Asia	Sea-97	NT	-	NT	-	0	-	0.14	1.42	0.5	2.19	NT	-	0.2	1.13
TAI/11/2019	Asia	Sea-97	NT	-	NT	-	0.13	1.01	0.39	1.87	0.25	1.89	NT	-	0.62	1.62

			Asia 1	Shamir
Isolate	Serotyp	e Asia-1	Boeh Ingel	ringer heim
	Topotype	Lineage	<i>r</i> 1	Titre
IRN/1/2020	ASIA	Sindh-08	0.24	1.95

Isolate	Serotype	e SAT 1	SAT1 F Boehi Ingel	Rho 78 ringer heim
KEN/3/2020	I (NW7)	-	1	2.46
KEN/9/2020	I (NWZ)	-	0.43	1.88

Isolate	Serotype SAT 2		SAT 2 Er Boeh Ingel	r itrea 98 ringer heim	SAT 2 Zim 83 Boehringer Ingelheim		
	Topotype	Lineage	<i>r</i> ₁	Titre	<i>r</i> ₁	Titre	
ZAM/5/2021		-	0.14	0.95	0.16	1.71	
ZAM/2/2021	IV	-	0	-	0.06	1.25	
NIG/1/2020	VII	Lib-12	0.76	1.70	0.23	1.96	
NIG/6/2020	VII	Lib-12	0.95	1.80	0.27	1.98	

Isolate	Seroty	pe A	SAT3 Z Boehr Ingel	lim 83 Tinger heim
	Topotype	Lineage	<i>r</i> 1	Titre
ZAM/9/2018	II (WZ)	-	0	-



Appendix 3 Nucleotide sequence analysis

FMDV nucleotide sequence data for phylogenetic analysis

Testing Laboratory	Sample Country	Region Sequenced	Total	0	А	C	ASIA-1	SAT 1	SAT 2	SAT 3	FMDV GD	Notes
	Burkina Faso	VP1	4						4			
ANSES	Niger	VP1	19		18				1			
	Mauritius	VP1	2	2								
	Bangladesh	VP1	20	18	2							
APQA	Cambodia	VP1	35	16	4						15	
	Vietnam	VP1	6	6								
ARC-OVI	South Africa	VP1	4						1	3		
	Namibia	VP1	18	7					4		7	
BVI	Malawi	VP1	5					2			3	
	Zambia	VP1	14	4							10	
FGI-ARRIAH	Russia	VP1	2	2								
	China	VP1	34	34								
LVRI	China	Complete Genome	5	5								
NCFAD	Nigeria	Complete Genome	117	103	22				24		28	
	Argentina	Complete Genome	19	5	14							historical samples
	Bolivia	Complete Genome	1	1								historical samples
	Brazil	Complete Genome	38	7	31							historical samples
	Chile	Complete Genome	1	1								historical samples
	Colombia	Complete Genome	19	11	8							historical samples
PANAFTOSA	Ecuador	Complete Genome	3	1	2							historical samples
	Guyana	Complete Genome	2	2								historical samples
	Paraguay	Complete Genome	2	1	1							historical samples
	Peru	Complete Genome	2	1	1							historical samples
	Uruguay	Complete Genome	2	1	1							historical samples
	Venezuela	Complete Genome	6	3	3							historical samples
RRL SEA	Thailand	VP1	10	5	5							•



Testing Laboratory	Sample Country	Region Sequenced	Total	0	А	C	ASIA-1	SAT 1	SAT 2	SAT 3	FMDV GD	Notes
Sciensano	-	VP1	16	5	3			1	2	3	2	Samples from PTS schemes
	Turkey	VP1	21	21								
	Iran	VP1	15	10	4		1					
	Bahrain	VP1	4	3	1							
	DR of Congo	VP1	15	15								
	Iran	VP1	41	22	18		1					
	Israel	VP1	6	6								
	Kenya	VP1	16	13	1			2				
WRLFIND	Mongolia	VP1	4	4								
	Nepal	VP1	39	38	1							
	Nigeria	VP1	102	53	29				20			
	Vietnam	VP1	16	16								
	Zambia	VP1	41	33					7	1		



Appendix 4 Selected phylogenetic trees for 2021

A4.1: Sequencing of viruses associated with outbreaks in China

Data from FGBI ARRIAH





A4.2: Sequences from Mongolia

Report on FMDV O in Mongolia in 2021 Batch: WRLFMD/2021/00012



^{*,} not a WRLFMD Reference Number



A4.3: Sequences from Mauritius

Data from WRLFMD and ANSES

Report on FMDV O in Mauritius in 2021 Batch: WRLMEG/2021/00004





A 4.4: Sequences from Jordan

Data from Jordan University of Science and Technology (JUST) and WRLFMD

Report on FMDV O in Jordan in 2021 Batch: WRLMEG/2021/00022







A4.5: Serotype O sequences from Turkey and Iran

Data from *ŞAP* Institute, Turkey

Type O (Turkey and Iran Isolates)



0,03



A4.6: Serotype O sequences from Russia

Data from FGBI, ARRIAH and WRLFMD

Report on FMDV O in Russia in 2021 Batch: WRLMEG/2022/00005



*, not a WRLFMD Reference Number



A4.7: Serotype O sequences from Bahrain

ETH/6/2015 VEM/29/2006 VEM/3/2006 VEM/3/2006 VEM/3/2009 VEM/3/2009 VEM/4/2008 VEM/4/2008 VEM/6/2008 100 83 1 100 YEM/6/2008 YEM/5/2008 YEM/8/2008 ETH/31/2008 ETH/32/2008 /2009 2009 YEM/28/2004 - YEM/28/2004 YEM/28/2003 YEM/22/2003 YEM/1/2004 KEN/1/99 KEN/3/98 KEN/7/98 SOM/4/2007 93 SOM/2/2007 SOM/1/2007 100

Report on FMDV O in Bahrain in 2021 Batch: WRLFMD/2021/00005





A4.8: Serotype A sequences from Bahrain

Report on FMDV A in Bahrain in 2021 Batch: WRLFMD/2021/00005



*, not a WRLFMD Reference Number



A4.9: O/EA-2 outbreaks in Namibia

Report on FMDV O in Namibia in 2021 Batch: WRLMEG/2021/00012



^{*,} not a WRLFMD Reference Number



A4.10: SAT 2 (topotype I) outbreaks in South Africa

Data from ARC-OVI





A4.11: SAT 2 (topotype I) outbreaks in South Africa

Data from ARC-OVI





Appendix 5 The 16th Annual Meeting of the OIE/FAO FMD Reference Laboratory Network

23rd and 24th November 2021

Core Members

	OIE Reference Laboratory for Foot and Mouth Disease, Dirección de Laboratorio Animal,
•	SENASA, Argentina
	Participants: Sabrina Galdo
	OIE collaborating Centre for validation, quality assessment and quality control of diagnostic
	assays and vaccine testing for vesicular diseases in Europe, and FAO Reference Centre for
	Vesicular Diseases
	Sciensano, Belgium
	Participants: David Lefebvre, Nick De Regge
	OIE Regional Reference Laboratory for Sub-Saharan Africa (RRLSSA)
	Botswana Vaccine Institute (BVI), Botswana
	Participants: Elliot Fana, Mokganedi Mokopasetso
	Centro Panamericano de Fiebre Aftosa (PANAFTOSA) and PAHO /WHO and OIE Reference
	Laboratory for FMD, Brazil
	Participants: Edviges Maristela Pituco
	FAU FIND Reference Laboratory, National Centre for Foreign Animal Disease National Centres
	for Animal Disease, Canadian Food Inspection Agency, Canada
	OIE and China National EMD Reference Laboratory Lanzbou Veterinary Research Institute
*)	(IVRI) CAAS People's Republic of China
	Participants: Haixue Zheng , Jianhong Guo, Jiiun He. Wen Dang, Xiangtao Liu
	OIE FMD Reference Laboratory. French Agency for Food and. Environmental and
	Occupational Health & Safety (ANSES), France
	Participants: Labib Bakkali Kassimi, Souheyla Benfrid
	FAO Reference Centre for FMD in South Asia, ICAR – Directorate of Foot-and-Mouth Disease,
۲	Indian Council for Agricultural Research, Mukteswar, Nainital (Uttarakhand), India
	Participants: J. K Biswal, Jajati Keshari Mohapatra, Rabindra Prasad Singh, Samir Kumar Sarangi, Saravanan Subramaniam
	OIE/FAO FMD Reference Laboratory, Istituto Zooprofilattico Sperimentale della Lombardia e
	dell'Emilia Romagna (IZSLER), Italy
	Participants: Efram Foglia, Giulia Pezzoni, Santina Grazioli
# \$	OIE Reference laboratory for Foot and Mouth Disease, Animal and Plant Quarantine Agency
	(QIA), Republic of Korea
	FACT FMD Reference Laboratory, Wageningen Liniversity & Research (WILIR). The Netherlands
	Participants: Aldo Dekker. Phaedra Eble
	FAO Reference Centre for FMD for Central Asia and West Eurasia and OIE Reference
	Laboratory for FMD, Federal Governmental Institute, Centre for Animal Health (FGI ARRIAH),
	Vladimir, Russian Federation
	Participants: Svetlana Fomina, Ms Karaulov
	FAO Reference Laboratory for FMD in Africa and OIE FMD Reference Laboratory,
	Transboundary Animal Diseases Programme, ARC-Onderstepoort Veterinary Institute (ARC-
	OVI), South Africa
	Participants: Livio Heath, Melanie Chitray, Pamela Opperman
	OIE Regional Reference Laboratory for Foot and Mouth Disease in the South East (RRLSEA)
	Department of Livestock Development, Thailand
	Participants: Kingkarn Boonsuya Seeyo, Sahawatchara Ungvanijban



FAO	World Reference Laboratory and OIE FMD Reference Laboratory
🛌 The	Pirbright Institute Pirbright, United Kingdom
Partic Afzal,	ipants: Abdelaziz Yassin, Ali Burman, Amin Asfor, Antonello Di Nardo, David Paton, Donald King, Julie Maryan, Madeeha Nick Knowles, Sarah Belgrave, Valérie Mioulet
FAO	Reference Centre for FMD and other vesicular diseases for the Americas and the
Caril	bbean and OIE FMD Reference Laboratory
Fore	ign Animal Disease Diagnostic Lab, Plum Island Animal Disease Center (PIADC), United
State	es of America
Partic	ipants: Robin Holland, Sulee Robbe-Austerman

Affiliates

*	Australian Centre for Disease Preparedness (ACDP), Australia Participants: Nagendra Singanallur, Petrus Jansen van Vuren, Wilna Vosloo
.	NATIONAL Animal Health Diagnostic & Investigation Center (NAHDIC), Ethiopia Participants: Daniel Gizaw
	Foot and Mouth Disease Laboratory, Kenya Participants: Abraham Sangula
	National Veterinary Research Institute, Vom, Plateau State, Nigeria Participants: Hussaini Ularamu, Wungak Yiltawe
C*	Şap Institute (and WELNET FMD), Ankara, Turkey Participants: Can Cokcaliskan, Unal Parlak
	Pan African Veterinary Vaccine Center for African Union (AU-PANVAC), Ethiopia Participants: Ethel Chitsungo, Cisse Rahamatou

OIE/FAO Representatives

eofmd	The European Commission for the Control for Foot-and-Mouth Disease
	Participants: Bryony Armson, Carsten Potzsn, Fabrizio Rosso, Kees van Maanen
E	Food and Agriculture Organization of the United Nations
	Participants: Melissa McLaws, Samia Metwally
Oie	OIE – World Organisation for Animal Health
	Participants: Bolortuya Purevsuren, Min-Kyung Park

PCP Support Officers (PSOs) & Heads of Regional Laboratory Networks/Epi/Lab leaders

Epi Leader, West Eurasia
 Participant: Satenik Knaratyan
Epi Leader, Middle East
Participant: Rehab Abdelkader
Lab Leader, SAARC, India
 Participant: Rajeev Ranjan
PSO, Azerbaijan, Kyrgystan
 Participant: Carsten Potzsch
PSO, South Sudan, Sudan, Palestine
 Participant: Kees Van Maanen
PSO, The Gambia, Sierra Leone
Participant: Austine Bitek

EPI-interactive



E[#] EPI-interactive Participants: Petra Muellner, Uli Muellner



TUESDAY 23rd NOVEMBER 2021, DAY 1

Opening of the 16th annual meeting and adoption of agenda (Don King)

Opening remarks from Dr King provided a brief review of the history and core activities of the Network which was established in 2004. Since the last meeting in 2000, WBR Lelystad (The Netherlands) has been designated as an FAO Reference Laboratory for FMD and has signed the MoU as a core member of the Network. In view of the COVID-19 situation, this is the second meeting of the Network in a "virtual format" which allows wider participation from FAO, OIE and affiliate laboratories, LabNet and EpiNet leads from the FMD regional roadmaps and PCP Support Officers (PSOs).

Recent achievements of the Network were reviewed, including:

- Publication of the 2020 Annual Network Report summarising the global situation regarding the distribution of FMD in different regions of the world.
 - Circulation of a new simple-to-complete Excel spreadsheet to help with analysis of trends and the preparation of the annual report
- Tools to improve surveillance in endemic pools via the development of cost-effective pipelines to ship samples and translation of the e-learning course for FMD diagnostics into French
- Coordinated work to develop improved methods to assess serological responses that can be used for post-vaccination testing and FMD surveillance in endemic countries
- Two manuscripts have been published in peer-reviewed journals during 2021:
 - History of serotype C and recommendations to prevent re-introduction of the serotype published in *Virus Evolution*
 - FMD Reference materials highlighting current gaps in available reagents published in *Scientific and Technical Review of the OIE*
- Development of a FMDV sequence database (<u>https://www.fmdbase.org/</u>) and work to establish web-based dashboards to facilitate data exchange and analyses (discussed on Day 2)
- Distribution of a survey to collected views on FMDV strain nomenclature
- Other reference laboratory networks are currently being developed by the OIE/FAO using the FMD Reference Laboratories Network as a positive example.

Update on OIE activities on FMD (Min Kyung Park)

Twenty-four applications for disease-free status were received in May 2021; thirteen of these were for FMD. Six FMD applications across three countries were successful; in Brazil, the whole country is now free from FMD covered by different zonal statuses free with and without vaccination with a larger part of the country free with vaccination. Russia has two newly recognised zones free with vaccination while the rest of the country is FMD free without vaccination. The FMD-free with vaccination zone has been extended in Colombia at the border with Venezuela.

A revision of the Terrestrial Animal Health Code Chapter 8.8 Infection of foot and mouth disease has been circulated to members for comment. The introduction of vaccinated animals into an FMD-free zone or country where vaccination is not practiced is under discussion, along with alternative surveillance measures to demonstrate freedom from FMD after an outbreak in a shorter recovery period from six to three months. It is also proposed that the Protection Zone (Article 4.4.6) be used



as a risk management strategy to minimise the impact of disease introduction. Chapter 3.1.8 Foot and mouth disease (infectious with foot and mouth disease virus) in the Manual of Diagnostic Tests and Vaccines for Terrestrial Animal 2019 has two proposed revisions.

Action 021-01 coordinate contributions from the network to the OIE code and OIE Manual – Led by David Lefebvre, Charles Nfon, Labib Bakkali-KassimiKassimi and Sabrina Galdo. The harmonisation of heterologous post-vaccination responses (Paragraph "D. Vaccine matching tests" of Chapter 3.01.08 of the OIE Manual) will be reviewed by the new working group (see Action 021-09)

Update from FAO (Samia Metwally)

Dr Metwally provided thanks to everyone attending the meeting; the network is doing a great job in understanding and controlling FMD. The global action plan for regional epi and lab networks was outlined as a way to manage FMD at the regional level for the next 5 years with reference laboratories and the Network continuing to work as collaborative partners. The Emergency Management Centre for Animal Health (EMC-AH) has been introduced to help countries respond to disease emergencies.

An initiative towards eliminating residual risks associated with FMD, serotype C is underway (under umbrella of GF-TADs) based on compiling evidence that this virus serotype no longer circulates in livestock populations. This initiative (2021-26) will review surveillance evidence that serotype C is extinct and will aim to reduce the risk of FMD-C being released and also adopt a system to respond if this serotype reappears.

Regional roadmap meetings for SADC, W. Africa and Middle East have taken place along with Regional Advisory Group Meetings for E. Africa, W. Eurasia, SADC and W. Africa. Epidemiology and Laboratory Network meetings for W. Africa, W. Eurasia and Middle East have been strengthened by the work of PCP-FMD support officers within countries.

The Global Coordination Committee of FMD (GCC-FMD) met in September 2021 and is working to harmonise an FMD control strategy across the world.

Action 021-02 Plan to improve ongoing connectivity of the Network to regional laboratories (e.g regional roadmaps) and the inclusion of more laboratories in the proficiency testing schemes

Update from WRLFMD (Don King)

The number of samples (n=451) received to the WRLFMD in 2021 has increased compared to 2020 when overseas shipments were adversely affected by COVID-19. A coordinated approach by Valérie Mioulet, Kees van Maanen and Paolo Motta to target priority FMD endemic countries was acknowledged as responsible for a larger number of samples received by the WRLFMD this year. Shipments include those from the DR Congo (where samples have been received for the first time in 10 years) and Nigeria where large batches of samples have been collected. Real-time exchange of viral sequences for FMD outbreaks in Israel, Iran, Malawi, Mauritius, Namibia and Turkey has benefited from the connections established within this Network.

Headline events in 2021 include:

• The identification of serotype O (O/EA-2 topotype) in Namibia, where cases in July represent the continued spread of this lineage that originates from East Africa. Together with cases due



to the O/EA-2 topotype that have also been detected recently in Zambia, these outbreaks pose new threats to Southern Africa where serotype O is not normally resident.

- Increasing dominance of the O/ME-SA/Ind-2001e lineage in Pool 1 where it appears to be supplanting previously circulating serotype O lineages (including O/SEA/Mya-08 and O/ME-SA/PanAsia). This lineage continues to be detected in Pakistan (where it was first detected in 2019) and there has also been a second detection of the O/ME-SA/Ind-2001e lineage in Mauritius in 2021 that appears to be distinct to viruses that caused outbreaks in 2016.
- Detection of SAT 2 (topotype I) in KwaZulu-Natal in May 2021 that represents an FMD outbreak that has occurred away from the usual areas of high concern in southern Africa.
- FMD viruses detected in animals imported from East Africa to Bahrain were characterised as O/EA-3 and A/AFRICA/G-1 highlighting the ease by which FMD viruses can be imported into new areas via the trade in live animals.

A new research paper (Di Nardo et al., 2021 – *Molecular Biology and Evolution*) from the WRLFMD and collaborators has studied VP1 sequences collected over a 20-year period for lineages endemic in the Middle-East. These results help us to understand how FMD is maintained (as periodic waves) in endemic countries and identifies important viral reservoirs for serotypes O, A and Asia-1 in Afghanistan, Pakistan and Iran.

Vaccine matching data at WRLFMD have been generated for 27 field strains against the vaccines from MSD and BI and now also includes O1 Campos from Biogenesis Bago. Vaccine selection for endemic settings is assisted by continued work to measure heterologous antibody responses – where the use of regional reference antigens is increasingly important.

Pool 1: Southeast Asia (Kingkarn Boonsuya Seeyo)

Samples have been received from Thailand (n=75 total; 17 serotype O and 34 serotype A isolates) and Lao PDR (n = 20 no virus detected). Sequencing characterises the FMD viruses collected from Thailand as belonging to the O/ME-SA/Ind2001e and A/ASIA/Sea-97 lineages. A recent NSP serosurvey in Thailand (n=2,866 sera) has highlighted a seroprevalence of 30.08%. Vaccine matching has been undertaken using LBPE and VNT; where serotype O (O/ME-SA/Ind-2001e lineage) were well-matched against the O/189/87 FMD vaccine from Thailand. Testing of two Thai vaccine strains for serotype A shows that A/Sakolnakorn/97 is well matched for most A/ASIA/Sea-97 strains (6/7 for VNT and LBBE), while the A/Lopburi/2012 responses are less well-matched (only 1/7 generated an r1 value >0.3 using VNT). A renovation project of the BSL-3 facilities at Pakchong is underway and will be ongoing until 2023.

Pool 1: East Asia and China (Wen Dang)

A total of three official FMD outbreaks have been reported in three different provinces in China during 2021. Samples from confirmed cases have been supplemented by other samples from clinical or suspect FMD cases in the country (17 samples in total). National surveillance for FMD has also continued using real-time RT-PCR (n= 1801 lymph node and OPF samples), LPBE and 3ABC ELISA for samples collected in slaughterhouses located in 12 Chinese provinces and from border areas in the southwest of the country. FMD positive samples (n=33, VP1) have been sequenced – representing from serotype O (O/CATHAY and O/ME-SA/Ind-2001e lineages) with close relationships to FMD viruses circulating in China and southeast Asia in recent years. The O/ME-SA/PanAsia strain has not been identified since 2019 and there have been no outbreaks caused by O/SEA/Mya-98, but the



lineage is believed to still be circulating in the field, especially in pigs. Vaccine matching (using VNT) shows a good antigenic match for the Re-O vaccine strain against O/ME-SA/Ind-2001e, O/SEA/Mya-98 and O/CATHAY field strains, in contrast to the O/BY/2010 vaccine strains that generated poor results for Mya-98 and CATHAY lineages. In addition, in vivo potency studies have been conducted in pigs to demonstrate protection for the Re-O (and alternative Re-O/17002) vaccine against a recent O/SEA/Mya-98 field isolate.

Pool 1: East Asia and South Korea (Jong-Hyeon Park)

There have been no FMD outbreaks reported in the Republic of Korea during 2021 and the last FMD virus lineage detected was O/ME-SA/Ind-2001e in January 2019. Surveillance activities continue involving the testing of samples from quarantine inspection (n=219 rRT-PCR), Animal disease control (n=160 rRT-PCR) and NSP antibody sero-surveillance (n=449,002). The vaccination policy in the Republic of South Korea uses commercial vaccines from three international suppliers (BI, ARRIAH and Biogenesis) covering serotypes O and A. Sera (n=448,458) have been tested for post-vaccination monitoring purposes using serotype O ELISA demonstrating that antibody levels in cattle, breeding pigs and fattening pigs are all >80% at a population level. Vaccine matching (using VNT) has been performed for four established FMD vaccine strains (O1/Manisa, O/3039, O/Primorsky/2014 and O1/Campos) as two FMD vaccine candidates from South Korea (Om-O/PanAsia-2 and O/BE/SKR/2017) against representative field isolates from four different lineages (O/ME-SA/PanAsia, O/ME-SA/Ind-2001d, O/SEA/Mya-98 and O/CATHAY). The best antigenic match for all eleven isolates was demonstrated for the O1/Manisa vaccine. For serotype A, the A/Zabaikalsky and A22 vaccines provide best coverage for A/ASIA/Sea-97 in contrast to the A24/Cruzeiro that was not matched to any of these field isolates. In vivo potency studies provide confidence that vaccines are fit for use, where new smallscale studies in pigs have indicated that three serotype O vaccines (O1/Manisa+O/3039, O/Primorsky/2014+O/SEA/Mya-98 and O1 Campos) when given as two doses provide protection for challenge with an O/CATHAY field isolate. Similar small-scale studies in pigs have also investigated the cross-protection provided by serotype A vaccines.

Pool 1: Russia (Svetlana Fomina)

The Russian Federation has three zones recognized as FMD free by the OIE, comprising a large zone that is FMD-free without vaccination and two additions zones that are FMD free with vaccination (13 areas in the southwest bordering the Black and Caspian seas and the Island of Sakhalin). Application to the OIE for FMD-free status for a further 4 zones along the southern border will be made in 2022. In 2021, 131 samples from cattle and pigs with suspected FMD were tested using RT-PCR, but no FMD virus was detected. A further 444 samples collected from wildlife located in different regions for surveillance purposes yielded RT-PCR negative results. Sero-surveillance using LPBE has been undertaken in non-vaccinated populations (n=24,150 sera) to demonstrate absence of FMD circulation and the LPBE has also been used in FMD vaccination animals (n=19,879 sera) to assist in post-vaccination monitoring. In addition, the NSP ELISA has been used to test 63,706 sera from cattle during 2021. The O/ME-SA/Ind-2001e lineage has been identified as a new risk for Russia, based on the circulation of this lineage in neighbouring counties (such as Mongolia).

Pool 2: India (Rabindra Prasad Singh)

The ICAR-DFMD laboratory has moved recently from Mukteswar to Bhubaneswar. Data presented in the talk covered samples that were tested during 2020, where 215 samples were received associated



with 94 serotype O outbreaks, 28 serotype A outbreaks and 9 serotype Asia-1 outbreaks in the country. During 2020, FMD sero-surveillance in India encompassed 28,284 samples which exhibited 16.2% positivity when tested by a 3AB3 NSP ELISA. New on-going surveillance for 2021 will also accommodate probang sampling in potential disease-free zones. New sequence data generated in 2020 were characterised as serotype O (O/ME-SA/Ind2001e) and Asia-1 G-IX (BD-18). The O/ME-SA/SA-2018 lineage (reported at last year's meeting) needs to be closely monitored in the region. The first detection of Asia-1/GX-IX (BD-18) in January 2020 can be interpreted as an incursion from Bangladesh or in-situ evolution within India from the prevalent Asia-1/G-VIII virus. A trivalent vaccine has been used in India since 2003 and vaccine matching is regularly carried out using monovalent BVS using 2D-VNT. Recent results show that serotypes O and Asia are well matched against respective field isolates, but the serotype A component only matches 56% of isolates collected since 2007.

Pool 3: Turkey (Can Çokçalişkan)

During 2021, 109 samples have been received from field cases of FMD in Turkey (60 typed as O and 3 FMDV-positive by RT-PCR but not serotyped), plus a further 25 samples from Iran (14 typed as O, 17 typed as A and 3 typed as Asia-1). Sero-surveillance activities in Turkey have included testing of sera (n=4596) to support the continued FMD-free (with vaccination status) in Turkish Thrace, NSP testing to support epidemiological investigations in Anatolia (n=58,279) and post vaccination monitoring. FMDV positive samples (n=35) have been sequenced to show that all the Turkish samples are serotype O within two sub-lineages (QOM-15 and ANT-10), while Iranian isolates have been characterised as O/ME-SA/PanAsia-2/ANT-10, A/ASIA/Iran-05/FAR-11 and Asia 1/Sindh-08. NB: serotype A and Asia 1 have not been detected in Turkey since 2018 and 2015, respectively. There are plans to include a new strain in the vaccine (representing an A/IRN/21 (A05-FAR-11) isolate) to compensate for the recent poor r-values generated by vaccine matching by the ŞAP Institute.

Pool 4: Kenya – East Africa (Abraham Sangula)

Ninety-eight samples were submitted from field cases of FMD in cattle during 2021 (37 typed as O, 7 as A, 1 as SAT1, 17 as SAT2). Approximately 1300 sera have been tested by VNT; where 74% were antibody positive, and approximately 200 samples have been tested using NSP ELISA with two-thirds positive for FMDV-specific antibodies. VP1 sequence data for samples submitted to the WRLFMD identified 13 serotype O, 1 A and 1 SAT1. The FMD virus lineages represented are: O/EA2, A/AFRICA/G-I, SAT1/I and SAT 2. Homologous in-vivo potency tests have been recently undertaken using the KEVEVAPI vaccine components (O K77/78, A K5/80 and SAT2 K52/84) generating >6 PD50 results for all experiments.

Pool 4: Ethiopia – East Africa (Daniel Gizaw)

East Africa is endemic for FMD and the disease is a major obstacle to agricultural development, livestock production and animal export. In Ethiopia, the most common serotypes are O (in the central and south-east of the country) and SAT 2 (in central and western areas). Due to COVID-19, there has been limited outbreak surveillance in the country; however, 112 samples have been tested by NAHDIC - 22 serotype O, 7 SAT1 and 24 SAT2. A further 8 samples could not be serotyped. Surveillance testing of 4695 small ruminant and bovine samples has also been undertaken, of which 849 were antibody positive and 3846 negative. Export testing has also been performed for small ruminants, where 18.1% were positive for NSP antibodies.


Action 021-03 Organise a shipment of samples to WRLFMD to further investigate the re-identification of SAT 1 in East Africa for the first time since 2007.

Pool 5: Nigeria – West Africa (Wungak Yiltawe)

There have not been many outbreaks this year but serotype O and SAT2 have been detected from the 24 suspect samples collected in the field. 8 samples were not able to be serotyped because of the antigen ELISA used but SAT1 is suspected, and these samples will be sent to WRLFMD or NCFAD, Canada for further analysis. More than 2000 samples have been received for surveillance purposes from cattle, sheep, goats and pigs where 400 samples have been identified as antibody positive. Large batches of samples have been sent recently to NCFAD, Canada and WRLFMD and results will be reported shortly.

Action 021-04 Samples to be shipped from NVRI for sequence analysis as soon as possible.

Action 021-05 WRLFMD or Sciensano to provide virus for monoclonal antibody screening to IZSLER so that the antigen ELISA kit can be upgraded.

Pool 4-6: Sub-Saharan Africa (Livio Heath)

From South Africa, forty-two clinical samples have been received during 2021, of which 2 have been serotyped as SAT 2 and 15 as SAT 3. A further 8 samples relating to illegal transborder animal movements were received from Eswatini, but no virus was detected. One SAT 2 and three SAT 3 viruses from the South African outbreaks have been sequenced. The FMD virus causing the SAT 2 outbreak in KwaZulu-Natal was closely related to an isolate collected in the north of Limpopo from 2019. The SAT 3 outbreaks are located in the same area in Limpopo that was affected during 2015. Surveillance activities using SPCE and RT-PCR have been undertaken for different purposes: (i) continued country-level survey, (ii) to support investigations into the field outbreaks of SAT 2 and SAT 3, and (iii) for animal movements in South Africa. A small number of additional samples have been tested for clients in Eswatini, Thailand, India and Zimbabwe.

Since 2015, 10 FMD outbreak events have been reported in South Africa. Buffalo are not implicated in any of the outbreaks identified in recent years and there is evidence to suggest the FMD viruses are being maintained within the domesticated cattle population, where many of the outbreaks have been characterised by mild clinical signs or sub-clinical infection. Vaccine matching using SPCE has been undertaken against the FMD vaccine strain SAR 3/04/2 that was developed 5 years ago at OVI.

Brief comments from questions:

The observation of mild clinical signs – may be related to the long-term circulation of FMD in cattle populations.

Status of locally produced vaccine? -in process of registering and hope to be in commercial use by mid-2023.

Pool 4-6: Sub Saharan Africa (Elliot Fana)

During 2021 the RRLSSA has received 49 samples collected from suspect FMD cases in 5 countries: from Namibia (serotype O and SAT2), Botswana (FMDV negative), Zambia (serotype O and SAT 2) and from Malawi (serotype SAT1). VP1 sequencing has confirmed SAT1 in samples from Malawi and O/EA-



2 and SAT2 in samples from Namibia as well as serotype O/EA-2 in samples from Zambia. Cases due to O/EA-2 in Zambia and Namibia represent the recent spread of this topotype from East Africa and pose new threats for onward spread south into Botswana, Zimbabwe and South Africa. Serotype O vaccines are not widely used in Pool 6. In order to anticipate future demand for a regionally relevant serotype O vaccine, the RRLSSA has generated new vaccine matching data (using VNT) for O-Manisa and O-3039 that indicates that these two vaccine strains are antigenically matched to the field isolate from Namibia.

Pool 7: South America (Maristela Pituco)

No suspect FMD samples have been received during 2021; however, testing has been performed for differential diagnosis of other endemic diseases (VS, SVV, bluetongue and poxvirus infection). A collaborative research project with NCFAD, Canada has completed the complete genome sequencing of 95 historical South American serotype O and A strains collected during 1950-2018. Together with other data, these sequences demonstrate that the North Andean region is an isolated ecosystem, and there is no evidence that FMD viruses (from serotype O or A) from this region have spread to other parts of South America.

With the exception of Venezuela, other countries in the South American region remain FMD-free (representing 95.9% of the total cattle population). PANAFTOSA is working with Venezuela to improve surveillance and to strengthen vaccination strategies to improve immunity. Proficiency schemes were organised for 22 laboratories from 17 countries this year to strengthen laboratory quality.

Question relating to differential diagnosis – what is the presence of Seneca Valley Virus infection in the pig population? For the COSALFA 2020, two countries reported the detection of Seneca Valley Virus as part of routine surveillance for FMD (and its differential diagnoses): Brazil with 86 farms affected and Colombia with 9 farms. In total, 1884 suspicions of FMD were investigated and ruled out by the countries of the Region in 2020. <u>https://www.paho.org/sites/default/files/informe_situacionpaises-2020-borrador_1.pdf</u>

Pool 7: South America (Sabrina Galdo)

Twenty-two samples relating to FMD 6 suspect cases have been received this year, all were negative for FMDV. Surveillance of more than 9,000 samples has been carried out to confirm absence of virus circulation and all were FMDV negative. Sequencing of 19 type O and 4 type A FMD viruses has been undertaken as part of a collaboration with Vietnam and 1 type O and 3 type A as part of a project with South Korea. Vaccine matching tests demonstrate good antigenic match for a range of serotype O field isolates (from O/CATHAY, O/ME-SA/Ind-2001, O/ME-SA/PanAsia and O/SEA/Mya-98 lineages) against the O1 CAMPOS vaccine strain, but a poor match for monovalent A24 Cruzeiro for the strains circulating in South Korea and Vietnam. However, the trivalent vaccine O1 CAMPOS/A24 Cruzeiro/A Argentina 2001 provided better results for these serotype A viruses. Three in-vivo potency studies have been undertaken using the trivalent vaccine (O1 Campos, A24 Cruzeiro, A/Argentine 2001) with challenge by serotype O isolates from South Korea which all yielded with good results (all >6PD50).

The only risk currently identified is in relation to trans-pool movements in other regions so evaluating the optimisation of antigen bank is a priority.

Action 021-06 Potency tests are expensive with ethical constraints and are being undertaken by a number of partner organisations. From a network perspective, we should consider how we can



coordinate these studies and ensure that data are rapidly communicated to support FMD control initiatives. WRLFMD to explore whether this information can be included as a new webpage on the Network website.

Update from SCIENSANO (David Lefebvre)

No suspect cases but 10 samples from 4 herds were received by Sciensano for exclusion diagnostics no FMD virus detected. Further samples were received for import, export drug testing and vaccine control, all were FMDV negative. Participation in three proficiency testing schemes from ANSES, TPI and BVI this year gives the opportunity in the absence of samples to keep sequencing knowledge up to date. No vaccine matching performed this year, but small-scale training is provided to students.

WEDNESDAY 24th NOVEMBER 2021, DAY 2

Update from CSIRO (Wilna Vosloo)

FMD activities include two research collaborations with FLI, Germany. The first project investigated the safe transport of infected epithelium samples, and whether FMDV will be inactivated using commercially available buffers and if virus can then be recovered for further investigation. McIlvane buffer was shown to be the simplest, cheapest and most effective option for on-farm inactivation and all buffers preserved RNA for at least 48 hours. The second project aimed to understand FMD virus survival confirming that temperature, rather than humidity impacts upon FMDV survival. FMD viruses were rapidly inactivated at temperatures > 20°C but could survive for up to 10 days at 10°C. A further project (with MSD) has adopted a systems-immunology approach to study early immune response to FMD vaccines in pigs by comparing intramuscular vaccine with intradermal vaccines administered using an IDAL device. This presentation also highlighted a number of in vivo studies that have been conducted recently to assess FMD vaccine performance (see Action 021-06).

Update from ANSES (Souheyla Benfrid)

During 2021, samples have been tested from Burkina Faso, Niger and Mauritius. Samples from Burkina Faso comprised 12 LFDs collected during 2020, from which serotype SAT 2 FMD viruses were detected (sequenced as SAT 2/VII/Lib-12), while samples from Niger included 7 LFDs and 59 swabs which yielded 15 serotype A and SAT 2 viruses (sequenced as A/AFRICA/G-IV and SAT2/VII, respectively). An outbreak in Mauritius (Rodrigues Island) this year resulted in the receipt of two Epi and 16 blood samples and the detection of 2 serotype O samples; sera from this outbreak were received for NSP and ELISA testing, 4 were NSP and O positive (unvaccinated) and 31 were type O positive by NSP negative (17 of which were vaccinated and 14 unvaccinated). Sequencing confirmed the reappearance of O/ME-SA/Ind-2001e in Mauritius.

Other activities include the validation of a new Triplex one-step real time RT-PCR for detection of FMDV and ongoing revision experiments for a Duplex one-step rtRT-PCR for detection of viruses from the A/AFRICA/G-IV lineage. ANSES organised an FMD Proficiency Testing Scheme for 42 laboratories from 40 countries.

Update from IZSLER (Santina Grazioli)

No clinical samples have been received this year.



IZSLER has produced and distributed 409 ready-to-use ELISA kits to 37 countries which represents an 80% decline based on previous years; this reduction in requests may be primarily linked to shipping issues this year. Two mulitplex lateral flow devices have been developed in collaboration with the University of Turin for identification and serotyping of FMDV in the field (One for EurAsian serotypes and one that detects PanFMDV, SAT 1, SAT 2). Validation using clinical samples gave diagnostic sensitivity results to antigen ELISA testing. Extensive field validation on clinical samples is now needed.

IZSLER has participated in an interlaboratory study (coordinated by WRLFMD) that aims to harmonise and calibrate VNT methods used for post vaccination monitoring. IZSLER provided support for the AgResults Foot and Mouth Disease Challenge Project focusing on the antigen profiling of 67 FMDV isolates for use as candidate reference antigens from Pool 4 of the EA region. Furthermore, a field trial to assess the effectiveness of multivalent FMD vaccines (O, A and Asia 1) used in Transcaucasian countries has been performed where 180 sera from Azerbaijan have been tested – where early results suggest that the vaccine is not expected to induce protective responses in the vaccinated population, even after a second administration of the vaccine.

Update from NCFAD (Charles Nfon)

This presentation provided an overview of data generated from an on-going collaboration with NVRI, Nigeria. A large batch of tissue samples (n=177) have been sent to Canada from which serotype O (n=103), serotype A (n=22) and SAT 2(n=24) have been detected. Sequence data confirm the presence of FMD viruses from the O/EA-3, A/AFRICA/G-IV and SAT 2/VII lineages. For serological surveillance, 1060 samples from multiple states in Nigeria have been received to add knowledge to the seroprevalence of FMD, 708 are still to be tested with ~85% positivity identified so far.

Update from FADDL/APHIS, Plum Island (Robin Holland)

Diagnostic testing is maintained for vesicular diseases and 1,738 samples have been tested for FMD by RT-PCR, the majority of which have been submitted for swine vesicular investigations in the US (many of which are associated with Seneca virus A infection). All FMDV testing as part of the passive surveillance programme in the US has yielded negative results. Capability for sequencing FMDV is maintained although none has been performed in 2021. A desktop exercise to plan for the event of an FMD outbreak has recently been undertaken.

Introduction from WBVR, The Netherlands (Phaedra Eble)

This presentation provided a brief history of WBVR and introduced the research team and the laboratory tests that are all ISO17025 accredited (RT-PCR, VI, Ag ELISA, VNT, Ab ELISA and NSP ELISA). A lot of epidemiological/transmission studies have been carried out to quantify the effect of emergency vaccination on dampening down an epidemic and this information is used in Dutch contingency plans, EU regulations and OIE standards.

Contract research for third parties involves animal experiments to test FMD Vaccines, including efficacy experiments, serological tests and challenge experiments in small and large animals. In the last five years 90 studies have been undertaken and this has resulted in a very valuable serum collection. As an FAO Lab, it is anticipated that WBVR can assist with post-vaccination monitoring, vaccine quality control, sharing information on the relationship between homologous and heterologous potency and also contribute llama FMDV-specific antibodies from a large collection.



Discussion: Tools to disseminate and display FMD data

Update on FMDV genotyping dashboard (Antonello Di Nardo/Uli Mueller)

The Network of OIE and FAO Reference Laboratories provides critical surveillance information about the circulation of FMDV lineages. This presentation highlighted new tools that have been recently developed by WRLFMD to disseminate FMDV sequence data and provide tools for viral genotyping. The core capability for these tools centre on a new FMDV sequence database (www.fmdbase.org) which displays and allows for downloading of publicly available (n=12,146) FMDV sequences. Unpublished FMDV sequence data can also be retrieved (n=16,764) from the database. A new initiative in partnership with Epi-Interactive (New Zealand) and funded by EuFMD is developing a FMDV Genotyping dashboard to allow users to generate custom phylogenetic trees and reports for inputted sequences. A prototype version of this tool was demonstrated by Dr Mueller.

Update on FMD Surveillance dashboard (Melissa McLaws/Shankar Yadav)

This second presentation provided an overview of new dashboard tools that have been established to help present FMD data to the wider community. This work builds on the quarterly reports from WRLFMD/EuFMD to (i) visualise FMD surveillance information, (ii) track progress along the PCP, (iii) identify gaps (i.e., monitoring of specific lineages) and (iv) promote and facilitate the sharing of data within the regional roadmaps. Three prototype dashboards have been established using Tableau Public (free software) for the display of (i) PCP information, (ii) FMD surveillance information and (iii) FMD vaccination data. It is planned that these systems will be interoperable with the Network website and other systems such as EMPRES-i. These systems were demonstrated, and feedback is requested - contact: melissa.mclaws@fao.org.

Action 021-07 Please complete a survey to share your opinions on the EuFMD FAST Report: https//www.surveymonkey.com/r/XHVLHNS.

Discussion:

[1] *Will the dashboards accommodate different FMDV lineages?* – MM: Yes, this is certainly an ambition of these tools – AdN: Scope to establish a single portal that uses the epidemiological/sequence data and connects to systems such as NextStrain (<u>https://nextstrain.org/</u>)

[2] What type of data will be included in the FMD vaccine dashboards? - MM: Initially this could accommodate vaccination questionnaires used at the regional roadmap meetings – but it is also imagined that vaccine matching data will also be very valuable.

Discussion: Vaccine Selection and post vaccination

Priorities for collaborative vaccine matching studies within the Network (Aldo Dekker)

The presentation reviewed approaches used to select FMD vaccines and predict their ability to protect animals from disease in the field, including vaccine matching (r1-value determination) and heterologous testing by VNT (where access to the vaccine strain is not required). The use of these methods can be complicated by relationship between VN titres and levels of protection which is not always the same within an FMDV serotype. Specific weaknesses of current approaches include (i) inherent variability of VNT methods in different laboratories and differences between results generated by VNT and LPBE, (ii) the use of pooled bovine vaccinal sera (BVS) that does not provide



adequate information on variance of the calculated r1-values, (iii) uncertainty and variability in the homologous potency of vaccines that are used to generate the BVS, and (iv) the complication of defining vaccine performance when multivalent vaccines are used. Immediate priorities include collection of standard reference sera for FMD vaccines (from standardized conditions) and the collection of more data from PVM studies. It was also suggested that better precision in r-values might be achieved by using five separate BVS (as opposed to pooled BVS) perhaps using a 1D assay format.

Alternative tests for vaccine evaluation (Amin Asfor)

An on-going collaborative OIE Twinning project with AU-PANVAC and WRLFMD aims to establish new immunoassay formats to evaluate the quality of FMD vaccines used in East Africa. New assays including total IgG, isotype and avidity formats have adapted approaches that have been successfully used in South America. Compared to VNT, these assay formats have the benefit that they do not require cell culture facilities and can potentially be used in low-containment laboratories. Sera from FLI (Brehm et al., 2008) has been used evaluate candidate assays; where VNT was found to be the best indicator of protection after vaccination. In this preliminary study, all ELISA formats provided a poorer indication of protection and feedback from other laboratories is sought to see whether similar results have been generated with these assays.

Question: Does recombinant integrin used in these assays differentiate between 146S and 12S FMDV components? – AA: No this is not the case – and the presence of 12S particles is an important confounder with these assays.

Regional reference antigens for heterologous testing: an update (Don King)

In many FMD endemic regions, vaccines can be sourced from a wide range of different suppliers (containing different vaccine strains and formulated at different potencies). Quality assessment of FMD vaccines often focuses only on the homologous responses (according to the OIE Manual), while performance of the vaccine in the field is dependent upon heterologous antibody responses that can protect against the virus strains that are circulating. Vaccine matching remains as an important tool but does not assess the responses of multivalent vaccines and is limited by access to commercial live vaccine strains and relevant BVS. In 2019, the Network proposed to support heterologous testing of final formulated vaccine products (as supplied to customers) using regionally relevant reference FMDV antigens. This heterologous system has been recently developed for FMD viruses circulating in East Africa (http://www.wrlfmd.org/node/2096/) where 16 FMD virus reference antigens (from 4 serotypes) have been used. In partnership with IZSLER, the WRLFMD has generated antigenic profiles for these viruses using VNT and binding of monoclonal antibodies. This simple system can be used to directly compare post-vaccination responses from different vaccines (even after booster vaccination) and has the benefit that access to the vaccine strains from the companies is not required. In order to further develop these tools, it is recommended that the Network partners work to identify candidate antigens that might be used (and made widely available) in each of the endemic pools and support work to collect representative sera collected after vaccination with a wide range of different vaccines. Although further work is required to define serological cut-offs and to address inter-laboratory variability of the VNT, the approach is broadly supported by the SCAD committee of the OIE and may be suitable for inclusion into the OIE Manual at a future point.

Questions: FAO is requesting that vaccine companies supply relevant sera (at small volumes) that can be used to help standardise testing in the countries where the vaccine is used.



Action 021-08: FAO will also explore whether larger volumes of post-vaccination sera might be supplied to Network laboratories for use as reference sera.

Action 021-09: It is proposed that the Network re-establish a working-group to cover vaccine selection and quality and the harmonisation of heterologous post-vaccination responses. Proposed members include: Aldo, Amin, Charles, Livio, Maristela, Phaedra, Sabrina, Santina, Samia (+ Anna Ludi when she returns from maternity leave).



Global and Regional FMD Risks: Review of regional and FMDV lineage distribution information and maps in Annual Report

Conjectured relative prevalence of circulating FMD viral lineages in each Pool*. For each of the regions, data represent the relative importance of each viral lineage [prevalence score estimated as a percentage (%) of total FMD cases that occur in domesticated hosts].

* - these are viral ecosystems not necessarily FAO or OIE recognised regions

Lineage	Southeast / Central / East Asia [Pool 1]	South Asia [Pool 2]	West Eurasia & Middle East [Pool 3]	North Africa	Eastern Africa [Pool 4]	West / Central Africa [Pool 5]	Southern Africa [Pool 6]	South America [Pool 7]
O ME-SA PanAsia-2			35					
O ME-SA PanAsia	10							
O SEA Mya-98	33							
O ME-SA Ind2001	20 (40)	80 (need to accommodate SA-2018) - 6	7	10 (2)				
O EA or O WA			3	55	55 (+0.5)	70 (65) 0-EA-3>0/WA	16 (O/EA-2)	•
O EURO-SA								80 (90)
O CATHAY	10.5							
A ASIA Sea-97	26 (18)							
A ASIA Iran-05	0		27 (22) 🚺					
A ASIA G-VII		16 (10)	15 (10)					
A AFRICA				25 (33)	22	15 (17)		
A EURO-SA								20 (10)
Asia-1	0.5 (0)	4	12.5					
SAT 1				0	8	3	27 (16) 📘	
SAT 2			0.5	10	14	10 (15)	57 (52)	
SAT 3					1 (0.5)		16	

1 Proposed new changes (new values in)

Recent epidemiological changes that were discussed at the meeting include:

- Increasing dominance of the O/ME-SA/Ind-2001e lineage in Southeast and East Asia (Pool 1)
- Continued absence of serotype Asia 1 in Pool 1
- Presence of a new lineage called O/ME-SA/SA-2018 in Pool 2
- Decreasing threats posed by A/ASIA/G-VII in Pool 3 (although this lineage is still present)
- Situation in North Africa where O/ME-SA/Ind-2001d has not been detected for >5 year
- Emergence of the O/EA-2 topotype in Pool 6 (Zambia and Namibia)

It is also proposed that the FMD viral lineages listed in this table will be modified to separate the African serotype O and A topotypes/clades (pending).