वार्षिक प्रतिवेदन

Annual Report

2009-10

परियोजना निदेशालय खुरपका एवं मुंहपका रोग
मुक्तेश्वर, नैनीताल 263 138
(उत्तराखंड)

Project Directorate on Foot and Mouth Disease
Mukteswar, Nainital 263 138
(Uttarakhand)

परियोजना निदेशालय खुरपका एवं मुंहपका रोग

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Front Cover Page

Distribution of Regional centers and Network Units of Project Directorate on FMD and disease lesions in Mithun and Buffalo

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Participants of OIE/ FAO FMD Reference Laboratories Network Meeting, 2009

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Contents

कार्यकारी सारांश

खुरपका मुंहपका (एफ0 एम0 डी0) रोग गो वशीम पशुओं में तीव्र गति से फैलने वाला विषाणु जनित संक्रामक रोग है। यह रोग केवल भारतवर्ष ही नही वरन् समूचे विश्व की अर्थव्यवस्था के लिए महत्वपूर्ण है। एक अनुमान के अनुसार दक्षिण पूर्व एशिया की 96 प्रतिशत जनसंख्या मिश्रित कृषि पर आधारित है, जिसमें 70 प्रतिशत हिस्सेदारी महिलाओं की है।

एफ0 एम0 डी0 केवल पशु उत्पादों को ही कम नहीं करता वरन् निर्यात को भी प्रतिकूल प्रभावित करता है। एफ0 एम0 डी0 मुक्त देश भी इसके प्रभावों से नहीं बच पाते और पशु उत्पादों को एफ0 एम0 डी0 प्रभावित क्षेत्रों या देशों से आयात पर पाबंदी लगा देते है। दक्षिणी पूर्वी एशिया में एफ0 एम0 डी0 एक स्थानिय रोग है जो विशेषकर गौ पशुओं में महत्वपूर्ण है। इस क्षेत्र में गौ पशु ही दूध, मांस एवं कृषि में प्रमुख भूमिका निभाते है।

एफ0 एम0 डी0 भारतवर्ष में 'ओ', 'ए' तथा 'एशिया–वन' सीरोटाइप द्वारा पूरे वर्ष होता है।

ओ0 आई0 ई0 (OIE)ने 70 देशों की पहचान एफ0 एम0 डी0 मुक्त क्षेत्रों के रूप में कर दी है, किन्तु 100 से अधिक देश अभी भी एफ0 एम0 डी0 से ग्रसित है।

सन् 2009 में पराग्वे देश में हुए विश्व एफ0 एम0 डी0 सम्मेलन में यह निष्कर्ष निकला की इस बिमारी का निदान तभी सम्भव है, जब विश्व के सभी देश एकजुट हो कर के एफ0 एम0 डी0 उन्मूलन के लिए संगठित रूप से कार्य करेंगे।

भारतवर्ष अपने पूरे समर्पण से एफ0 एम0 डी0 रोकथाम तथा उन्मूलन में योगदान कर रहा है। भारत में निर्मित तत्कालीन प्रयोग में आने वाली वैक्सीन कारगर तो है, परन्तु, वैश्विक उष्मीकरण के बढ़ने के साथ ही नई वैक्सीन के तकनीकी विकास पर बल दिया जा रहा है, जो न सिर्फ लगातार बदलते विषाणु से रक्षा कर सके वरन् अधिक समय तक पशुओं की रक्षा कर सके।

खुरपका मुंहपका रोग (FMD) निदेशालय भारत में सन 1968 में ए0 आई0 सी0 आर0 पी0 के रूप में स्थापित हुआ था। पिछले चार दशकों के दौरान निदेशालय का व्यापक विस्तार हुआ है तथा सन् 2001 तक 23 प्रादेशिक तथा क्षेत्रीय नेटवर्क यूनिट पूरे देश में एफ0 एम0 डी0 निरीक्षण तथा जॉच के अलावा अन्य नई जॉच की तकनीकों में भी विशेष योगदान दे रहा है। साथ ही साथ निदेशालय एफ0 ए0 ओ0 के दक्षिण पूर्व एशिया के प्रादेशिक निर्देश केन्द्र के रूप में भी कार्य कर रहा है तथा ओ0 आई0 ई0/एफ0 ए0 ओ0 के एफ0 एम0 डी0 वैश्विक नेटवर्क का भी सहयोगी है।

निदेशालय विश्व स्तर पर होने वाली वैक्सीन मिलान परियोजना में भी भाग ले रहा है।

सत्र 2009–10 में भारत में कुल 799 एफ0 एम0 डी0 आउटब्रेक अंकित किये गये, जो कि पिछले सत्र (511) से अधिक है। सबसे अधिक आउटब्रेक पूर्वी भारत से ज्ञात हुए जो कि पिछले सत्र से चार गुना अधिक है तथा पूर्वोत्तर क्षेत्रों में भी आउटब्रेक लगभग दो गुना अधिक ज्ञात हुए है। जबकि दक्षिण भारत में एफ0 एम0 डी0 पिछले सत्र की अपेक्षा कम पायी गयी है। हिमांचल एवं पंजाब में एक– एक तथा तमिलनाडु में शून्य आउटब्रेक ज्ञात हुऐ है। निरीक्षण से ज्ञात हुआ है कि हिमांचल प्रदेश, पंजाब, हरियाण तथा दिल्ली एफ0 एम0 डी0 मुक्त होने की दिशा में अग्रसर है। कुल 799 आउटब्रेकों से 1624 नमूने एकत्रित किये गये। सैंडविच एलाइजा एवं मल्टीप्लेक्स पी0 सी0 आर0 की सामूहिक जॉच में 1067 नमूनों में एफ0 एम0 डी0 विषाणु प्रमाणित किये गये। पूरे देश में हमेशा की तरह टाइप 'ओ' सर्वाधिक रूप से पाया गया, जो कि पिछले वर्षों कि तुलना से भी अधिक है। सीरोटाइप 'ए' उत्तरी तथा मध्य भारत में पाया गया जो कि पिछले कुछ वर्षों से यहॉ अनुपस्थित था।

एकत्रित नमूनों के विषाणुओं का आनुवाशिंकी निर्धारण कर न्यूक्लियोटाइड विभिन्नताओं का निर्धारण किया गया जो वैक्सीन विषाणु की खोज में अहम है। वैक्सीन में प्रयुक्त टाइप 'ए' के विषाणु (IND40/00) का पूर्ण न्यूक्लियोटाइड क्रम ज्ञात किया गया। 7 टाइप 'ओ' को 150 विषाणुओं की सूची में से भविष्य के विकल्प के तौर पर निर्धारित किया गया।

निदेशालय में स्थापित विषाणु सुरक्षा कोष में इस सत्र में 133 नये विषाणुओं को जमा किया गया। इस सत्र में नव निर्मित 'डीवा' किट द्वारा लगभग 21 राज्यों के 335 जिलों से 29763 सीरम नमनों की जॉच की गयी। एफ0 एम0 डी0 की रोकथाम हेतु प्रयोग में आने वाले सभी किटों को निदेशालय सरकारी एवं गैरसरकारी संगठनों को निरन्तर आपूर्ति करता रहा है, एवं इन संगठनों तथा प्रादेशिक केन्द्रों के कार्यकर्ताओं के प्रशिक्षण में मुख्य भूमिका निभाता रहा है।

– बी. पटनायक

1.0 Project Director's Report

Foot and mouth disease (FMD) is a major transboundary animal disease of importance, and remains a major threat to Global and regional food security. It is estimated that about 96% of the total population of South Asia live in the mixed crop-livestock farming zone, and women account for more than 70% of livestock production in the SAARC region. FMD adversely affects production and production efficiency is diminished in terms of quality of dairy products and weight gain ratios. Use of large ruminants for ploughing and traction are also seriously diminished due to the disease. In South Asia, FMD is endemic and predominantly associated with cattle and buffalo that are used for the production of the bulk of the milk and also meat, and currently FMD virus serotypes O, A and Asia1 are prevalent. Seventy countries in the world are officially recognized by the OIE as free from FMD with or without vaccination while more than 100 countries are still either considered as endemically or sporadically infected with the disease. The Global FMD Conference 2009 in Paraguay that was attended by different stake holders recommended a Globally Coordinated Approach to Control FMD. Freedom from FMD is essential for healthier economy and greater food security. There is need of strong commitment of all countries in the SAARC region to control and eradicate the disease In India, there is already an ongoing FMD Control Programme since 2003.

Though India produces suitable FMD vaccine, there is an urgent need in the country for research in vaccine to improve duration of immunity and thermostability to meet challenging environmental conditions. There is also need to develop alternative tests to replace current vaccine potency testing in cattle. So also, all vaccine batches must be tested for DIVA compliance.

The Project Directorate on Foot and Mouth Disease (FMD), the premier Institute for FMD in the country, was established as an All India Coordinated Research Project (AICRP) for FMD in 1968. During the last four decades of its existence the scope of the project was expanded considerably and several milestones were achieved to reach the current status of a Project Directorate in 2001 with 23 Regional Centers and Network Units covering all the major regions of the country. The Project Directorate has developed scientific expertise in conventional as well as in cutting edge areas, in the field of FMD diagnosis, epidemiology and research in the world. The primary mandate of the institute is to carry out research on the epidemiology of FMD in the country. It is also entrusted with the duty of post-vaccinal seromonitoring in FMD control programme currently undergoing in the country.

The institute also functions as the FAO Regional Reference Center for FMD for South Asia, and is also a member of the OIE/FAO Global Network of FMD Reference Laboratories. Recently, a FAO sponsored project entitled "Diagnostic Laboratory Network Coordination for FMD surveillance and vaccine evaluation in South Asia" with ICAR-PDFMD to play the lead role has been agreed in principle by both FAO and ICAR. SAARC has selected India to host the Regional FMD laboratory. The specific objectives of the project are:

- To establish a Regional Laboratory Network and Quality Assurance System.
- To establish agreement on vaccine performance monitoring and vaccine quality standards for the region.

- To develop a Regional roadmap towards long term FMD progressive control in the SAARC region.

The institute also participates in Global vaccine matching exercise. Recently a collaborative research programme under GFRA (Global FMD Research Alliance) with USDA (PIADC) on "Effective Molecular vaccines against FMD" has been initiated.

During this period, a total of 799 outbreaks were recorded/reported as against 511, during the year 2008-09 (Table 1). Maximum outbreaks were recorded in Eastern Region with four fold rise compared to last year. Similarly North Eastern region witnessed two fold rise in number of outbreaks. Drastic reduction in outbreaks was noticed in Southern region. This was mostly due to the effect of immunity following immediate past infection and high level of virus circulation in the population. No FMD cases were reported in Tamilnadu, whereas Himachal Pradesh and Punjab recorded a single case of FMD each. Though there is seasonal variation in occurrence of FMD in different parts of the country during 2009-10, maximum incidence of disease was reported in the month of March and August to December (Figure 1). The Northern states of Himachal Pradesh, Punjab, Haryana and Delhi hold promise to become "Disease Free Zone" with a little more effort.

Table 1. Number of FMD outbreaks in different years in different geographical region of the country

Year	North	West	South	East	Central	North East	Total
2005-06	314	59	2117	355	52	65	2962
2006-07	18	29	697	611	32	80	1467
2007-08	42	42	631	353	41	102	1211
2008-09	42	19	263	102	33	52	511
2009-10	61	33	85	498	20	102	799

A total of 1624 clinical specimens were collected from 799 outbreaks and some of these were duplicate samples from the same outbreak collected at different times. The details of the virus serotype confirmation are shown in Table 2. As usual there is dominance of type O virus in causing FMD in the country. Using sandwich ELISA and multiplex PCR, virus could be diagnosed in 1067 samples (Table 2).

During 2009-10, in all the geographical regions, serotype O was most prevalent. Incidence of serotype O

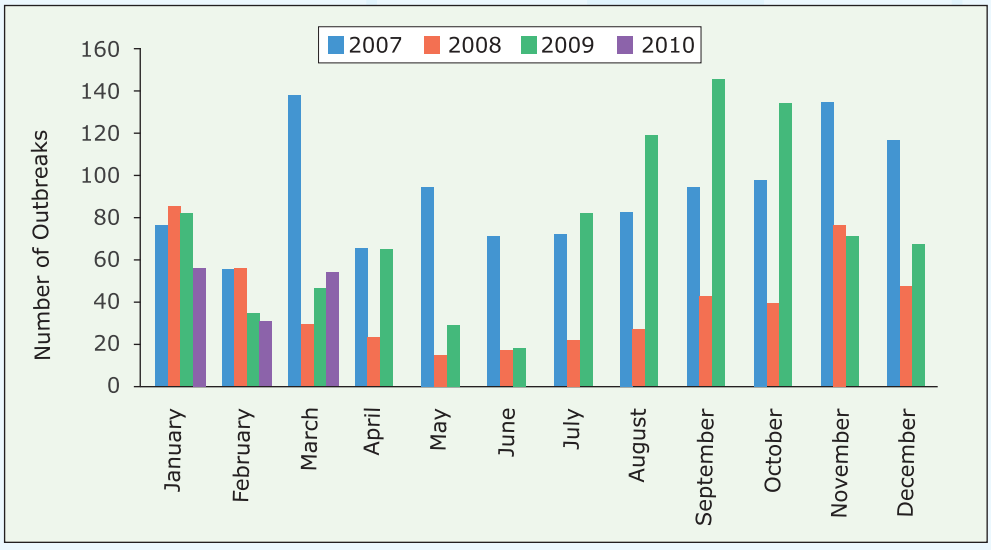


Fig 1. Month wise occurrence of FMD outbreaks. During 2009-10, FMD outbreaks occur round the year also with maximum incidence in the months of August to October and also in March

Table 2. Number of FMD outbreaks diagnosed during the period and serotypes involved

			Serotype				
			O	A	Asia1	Total	NVD
Number of tissue samples	2007-08	2258	1042	136	91	1269	982*
	2008-09	640	334	26	16	376	264*
	2009-10	1624	991	38	38	1067	557*
Number of Outbreaks	2007-08	1211	721	65	56	842	369
	2008-09	511	198	19	24	241	270
	2009-10	799	568	33	9	610	189

*Many of them are duplicate samples collected from same outbreak at different times

increased over the previous year. Increase in serotype O incidence was noticed in all regions except Northern and Western regions. Serotype A which was absent in Northern and Central regions last year appeared this year. Serotype Asia 1 was absent in Southern, Central and North Eastern regions. Incidence of Asia1 serotype was increased in Western region but came down drastically in Central region (Table 3). Though majority of the outbreaks involved cattle, disease was also reported in buffalo, pig, sheep and goats.

Compared to recent past, this year saw a major shift in genetic lineage of serotype O viruses circulating in India. The 'Ind2001' lineage viruses gained upper hand after a gap of 8 years and outcompeted PanAsia II lineage. The reemergence of this group has been tracked back to 2008, during which it caused sporadic outbreaks in Northern India. Soon it spread to majority of states in Eastern, Central and reached Southern most state of Kerala at the end of 2009, traveling through Andhra Pradesh and Karnataka. Nevertheless, the disease due to PanAsia 2, PanAsia1 and Branch C-II (a group divergent from the three lineages) viruses were not completely absent. In type A, all the isolates were found to cluster within genotype VII but grouped both in the nondeletion and the VP3[59] deletion lineages. Precisely, the viruses from Punjab and Uttarakhand shared ancestry and clustered in the same VP3[59] deletion lineage, VIIf, where as viruses from two different outbreaks in Orissa and Andhra Pradesh clustered together and

Table 3. Distribution/incidence/percent of FMD virus serotypes in different geographical regions as revealed by serotyping of clinical materials diagnosed during 2008-09 (numerator) and 2009-10 (denominator)

Geographical region	Serotypes			Number of clinical materials serotyped	
	O	A	Asia-1	2008-09	2009-10
North	**12/37(**80%/66.1%)	**0/13** (0%/23.2%)	03/6(20%/10.7%)	15	56
West	**18/40** (69.3%/57%)	**02/02** (7.7%/3%)	06/28(23%/40%)	26	70
South	**80/127** (86%/91.4%)	**13/12** (14%/8.6%)	**0/0** (0%/0%)	93	139
East	**104/619(**91.2%/99.04%)	**04/5** (3.5%/0.8%)	**06/01** (5.3%/0.16%)	114	625
North East	**60/114** (88.2%/100%)	**07/0** (10.3%/0%)	**01/0** (1.5%/0%)	68	114
Central	**60/42** (70.6%/93.3%)	**0/03** (0%/6.4%)	**25/0** (29.4%/0%)	85	45
Total	**334/979(83.3%/93.3%)**	**26/35(6.5%/3.3%)**	**41/35(10.2%/3.3%)**	**401**	**1049**

revealed no deletion in the VP3 coding region. Hence it appears both deletion and non deletion mutants belonging to genotype VII are co-circulating in the country in recent times. During the period, outbreak due to Asia1 serotype was recorded in Gujarat, Madhya Pradesh, Maharashtra, Uttar Pradesh and West Bengal. The Asia1 field isolates were grouped with lineage C reiterating the supremacy of this lineage in the country since 2005.

Studies on antigenic relationship of the field outbreak strains with in-use vaccine strains (Vaccine matching) is a regular exercise to monitor antigenic variation, if any, occurring in the field. This year, a total of 33 virus isolates including 20 type O, 7 type Asia1 and 6 type A field isolates were subjected to one-way antigenic relationship study. All of them showed close antigenic match (r-0.30) with respective vaccine strain indicating optimal antigenic coverage.

The transmission of FMD by contaminated semen of affected/convalescent bovine bulls has been a major concern for its use in artificial insemination (AI). The major risk lies with semen collected from the bulls before the onset of clinical disease. The detection and identification of FMD virus in semen prior to use in AI is crucial. Cytotoxicity of semen is a major limiting factor for isolation of FMDV from semen and genomic diagnosis. A modified RNA extraction method was adapted for RNA isolation in order to increase the efficiency of PCR diagnosis. This test system was developed to take care of enzyme inhibitors in semen and validated. Fifteen out of 32 semen samples tested were found positive for FMDV serotype O. This testing facility for semen is being extended to all germplasm/semen collection centers in the country.

The complete molecular characterization of in use new vaccine strain of serotype A (IND 40/00) was carried out. The sequence and phylogenetic analysis exhibited that the IND40/00 belongs to genotype VII and is similar to IND81/00. Moreover IND40/00 shares common ancestry with Middle East virus sequences with atleast 11% (average) nucleotide divergence from Middle East isolates.

Seven FMDV type O virus from a pool of nearly 150 previously characterized FMDV type O field isolates were short listed from the repository for evaluation as alternate candidate vaccine strains based on the criteria of infectivity titer in cell culture and genetic relationship with presently circulating virus groups/lineages. This work was undertaken in collaboration with FMD vaccine industry with the objective to have a panel of strains that can be used as vaccine strain when the need arises. Greater participation of Indian FMD vaccine industry is solicited in such exercises.

The National FMD Virus Repository is upgraded annually with inclusion of latest/new virus isolates. This year, a total of 133 virus isolates including 97 type O, 12 type Asia1 and 24 type A field isolates were added to the repository. At present this national repository contains a total of 1687 (1096-O, 317-Asia 1, 259-A, 15-C) well catalogued field isolates.

We have developed a recombinant nonstructural protein (3AB3) based ELISA test for differentiation of FMD infected from vaccinated animals (rDIVA-FMD). This DIVA kit is first of its kind for any animal disease in India and at least 4 times cheaper than the imported kits and is in extensive use in the country. A total of 29,763 random serum samples collected at the rate of 100 per district from 335 districts covering 21 different states of the country were tested in DIVA ELISA in an exercise to estimate FMD prevalence in the country. This revealed 27.9 % (average) of the bovine population in the country to be FMD infected during 2008-2010, which might fluctuate consequent upon inclusion of data from bigger states like Uttar Pradesh. This DIVA kit is now being used by FMD vaccine industry to identify FMD negative animals use in Quality Control of the vaccine.

Three diagnostic kits were produced and supplied to all across the country including FMD vaccine industry. This Directorate extending full diagnostic (pre- and post vaccinal immune response) support to the FMD Control Programme being run by the Department of Animal Husbandry, Dairying and Fisheries, GOI in selected 54 districts of the country. Eight Regional centers and 02 Network units, and the Central FMD Laboratory participate in this programme. The institute produces and supplies LPB ELISA kits for this programme alongwith bench training on its application. Level of Protective antibody response following each round of vaccination is monitored by the institute and fed back to the Government for effective implementation of the Control Programme. Gradual increase in protective antibody response was observed subsequent to phase 1 vaccination. After phase 8 vaccination, 79.5, 71.6 and 66.2 percent of animals tested were having protective antibody level (log_{10} 1.8 and above) against serotypes O, A and Asia-1, respectively.

Regular training and refresher courses for the scientific staff of Regional Centers and Network units were conducted on use/application of virus typing ELISA, LPB-ELISA and DIVA ELISA. Overall performance of the regional centers and network units were monitored periodically and any technical difficulties faced by the centers were removed instantly through electronic guidance. Sufficient fund was provided to all the centers and network units of the AICRP to carry out the technical programmes. Achievements of the scientists of the Project Directorate during the year have been excellent and deserve appreciation.

Collaboration with USDA (PIADC) under GFRA on "Effective Molecular vaccines against FMD" has been initiated. IVRI, Bangalore campus also participates in this programme. The overall objective of this project is to develop and evaluate a hAd-5 vector platform to generate candidate vaccines containing the P1 genomic region of the Indian vaccine strains (serotypes O, A, and Asia1) and any other widely antigenic reactive Indian field strains, and test its efficacy in cattle and buffalo.

The Project Directorate on FMD, a Regional FAO Reference Laboratory and a partner in OIE/FAO Global Network of FMD Reference Laboratories, participated in 2008 Global Inter-Laboratory Comparative Testing Exercise for FMD vaccine matching between members and observers within the OIE/FAO Network of FMD Reference Laboratories. This was organized by the European Community Reference Laboratory for FMD and the OIE and FAO-World Reference Laboratory (WRL) for FMD at the Pirbright Laboratory of the Institute for Animal Health (IAH-P) as per the discussion during Network Meeting held in Botswana in June 2007. This is the first step taken by the OIE/FAO Network in working towards establishing equivalence in the vaccine matching methods that are done in different laboratories. The LPB ELISA test developed by the institute did extremely well in this inter laboratory vaccine matching exercise.

I am happy to share that progress has been made towards establishment of International Center for FMD with state-of-the-art features of bio-safety and bio-containment (BSL 3+) during the 11th Plan period. The details of laboratory layout have been finalized by the Project Technical Committee constituted by ICAR, and the National Dairy Development Board (NDDB) has agreed to undertake this specialized job on turnkey basis. Finally, I hereby thank all my fellow colleagues, administrative, accounts and laboratory staff of the institute for their sincere efforts to accomplish the tasks assigned to the Institute.

- B. Pattnaik

2.0 Mandate, Objectives, Technical Programme

Mandate

Active epidemiological surveillance through regularly monitoring antigenicity and genomic make up of the FMD virus strains responsible for disease outbreaks, and also to provide training in diagnosis and epidemiology.

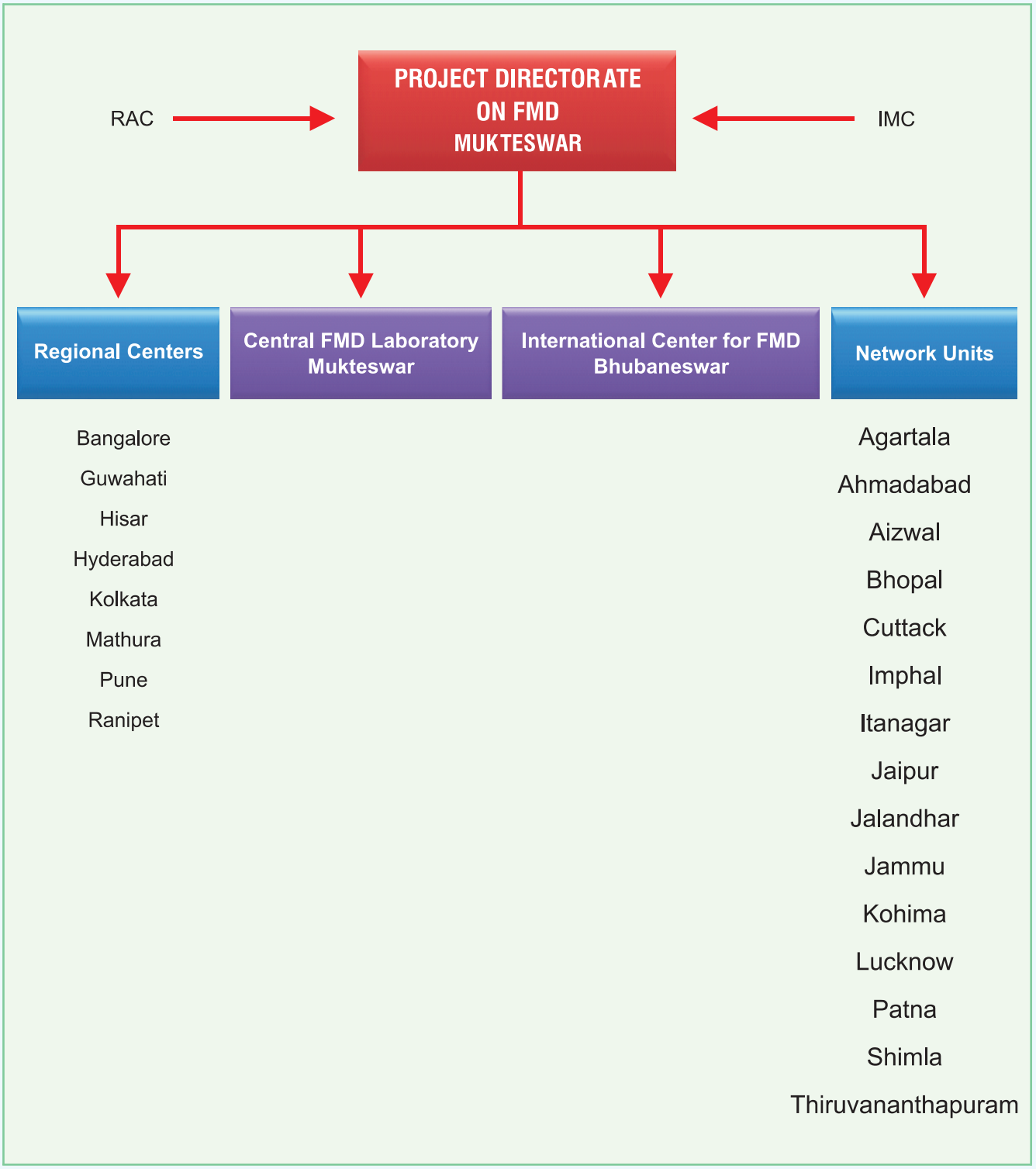
Objectives

1. To conduct systematic epidemiological and molecular epidemiological studies on Foot-and- Mouth Disease (FMD), and also to study carrier status of the infection and latency of the virus.
2. Antigenic and molecular characterization and cataloguing of FMD virus strains isolated from outbreaks, and monitoring suitability of the vaccine strains in use along with maintenance of National Repository of FMD Virus.
3. Production, standardization and supply of diagnostic reagents for FMD virus serotyping and post-vaccinal sero-conversion. Maintenance and supply of most appropriate vaccine strain to the FMD vaccine manufacturers.
4. Development of newer diagnostic techniques using cutting-edge technologies in molecular biology and hybridoma.
5. Analysis of economic impact of FMD on livestock industry
6. To act as referral laboratory for FMD in South Asia.

Technical Programme

1. To continue the production and standardization of type-specific anti-146S sera, antigen and other reagents used in sandwich and LPB ELISA for supply to the regional Centers and network units to ensure uniformity of results.
2. To continue to carryout typing of the clinical samples received from regional centers and network unit and also from other sources for confirmation/diagnosis.
3. To study the molecular epidemiology of FMD in India through ID gene sequencing.
4. To continue to carryout thorough antigenic and molecular characterization of field isolates.
5. To continue to carryout vaccine matching exercise
6. To continue the maintenance of Repository of important virus strains (Involving use of cryogenics) at PD FMD.
7. To continue to develop and standardize the advanced laboratory techniques in compliance with the International standards and pass them on to the concerned Centres/ Users/Stakeholders (subject to technical competence and facilities available) with proforma details to facilitate and ensure their uniform application.
8. To organize skill orientation programme of the scientific staff of the project for keeping them abreast with the latest knowledge and expertise from time to time through short-term training courses
9. Participation in FMD Control Programme with vital contribution in assessing antibody response following vaccination that indicates individual and herd immunity level.
10. National FMD Serosurveillance

3.0 Oraganizational Set-up



4.0 Staff Position

S. No.	Name of the scientist/staff	Designation	Month of Joining	Month of Leaving
		Scientific staff		
1	Dr. B. Pattnaik	Project Director	December 2006	Continuing
2	Dr. A. Sanyal	Pr. Scientist	April 2009	Continuing
3	Dr. B. B. Dash	Sr. Scientist	August 2009	Continuing
4	Dr. J. K. Mohapatra	Scientist	February 2006	Continuing
5	Dr. R. P. Tamilselvan	Scientist	May 2007	Continuing
6	Dr. S. Saravanan	Scientist	May 2007	Continuing
7	Dr. Sachin S Pawar	Scientist	May 2008	Continuing
8	Dr. K. Muniswamy	Scientist	June 2008	Continuing
9	Dr. P. Rameshkumar	Scientist	June 2008	Continuing
10	Dr. S. D. Narnaware	Scientist	October 2009	December 2009
11	Dr. G. K. Sharma	Scientist	December 2009	Continuing
12	Dr. Manoranjan Rout	Scientist	March 2010	Continuing
		Administrative staff		
1	Shri. D. N. Joshi	AAO	January 2009	Continuing
2	Shri. Raja Ram	A & AFO	August, 2008	Continuing
		Technical staff		
1	Shri. M. C. Meena	T-5 (Lab)	January 2007	Continuing
2	Shri. A. K. D. Bhatt	T-3 (Stockman)	April 1999	Continuing
3	Shri Nayan Sanjeev	T-1 (Lab)	October 2005	Continuing
		Supporting staff		
1	Shri J. P. Bhan	S. S. Gr. IV	February 2008	Continuing

5.0 Epidemiology Report

Table. 4 FMD outbreaks reported/recorded during 2009-10 and serotype involved

States	Reporting Centre/Unit	No. of outbreaks recorded	No. of Samples tested	Serotyping Results O	A	Asia1	NVD
		Southern Region					
Tamil Nadu	Ranipet	Nil					
Andhra Pradesh	Hyderabad	31	60	10	2	-	48
Karnataka	Bangalore	29(158)	80	50	-	-	30
Kerala	Thiruvanthapuram	25	165	62	6	-	97
	Total	**85**(158)	**305**	**122**	**8**	**-**	**175**
		Central Region					
Madhya Pradesh	Bhopal	20	45	42	-	3	-
Total	20	45	42	-	3	-	
		Western Region					
Gujarat	Ahmedabad	20	104	30	-	27	47
Rajasthan	Jaipur	(35)	8	-	-	-	8
Maharashtra	Pune	13	20	10	2	1	7
	Total	**33**(35)	**132**	**40**	**2**	**28**	**62**
		Northern Region					
J &K	Jammu	**(13)**	Clinical materials could not be collected				
Haryana	Hisar	2	7	2	4	-	1
Himachal Pradesh	Shimla	1	1	1	-	-	-
Punjab	Jalandhar	1	2	-	2	-	-
Uttar Pradesh	Mathura	49	63	32	6	5	20
UP	IVRI	5	28	10	1	-	17
UK	PDFMD	3	13	3	4	-	6
	Total	**61**(13)	**114**	**48**	**17**	**5**	**44**
		Eastern Region					
Orissa	Cuttack	44	51	9	4	-	38
Bihar	Patna	158	274	219	-	-	55
West Bengal	Kolkata	294	501	379	1	1	120
Jharkhand	Guwahati	2	6	6	-	-	-
	Total	**498**	**832**	**613**	**5**	**1**	**213**
		North Eastern Region					
Assam	Guwahati	45	48	33	-	-	15
Meghalaya		5	6	6	-	-	-
Sikkim		1	1	1	-	-	-
Manipur		3	6	6	-	-	-
Mizoram	Aizwal	1	1	1	-	-	-
Tripura	Agartala	29	26	19	-	-	7
Arunachal	Itanagar	18	108	43	-	-	65
	Total	**102**	**196**	**109**	**-**	**-**	**87**
	Grand Total	**799**(206)	**1624**	**974[17]**	**32[6]**	**37[1]**	**581**
				991	**38**	**38**	**557**

Outbreaks reported based only on clinical symptoms are in bracket.
Samples diagnosed by multiplex PCR are highlighted in red.

5.1 Southern Region

Tamilnadu (Ranipet): No outbreaks was recorded during the period under report

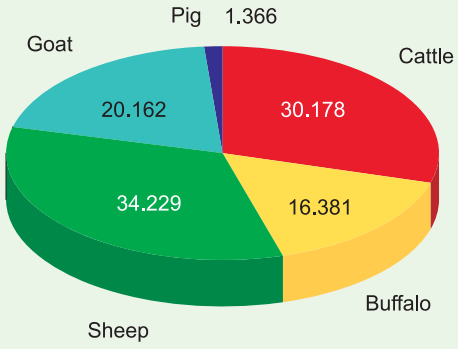
Andhra Pradesh (Hyderabad): During the year under report 31 outbreaks of Foot and Mouth Disease were recorded. Highest numbers of outbreaks (12) were reported during the month of October followed by 5 outbreaks in the month of March and 3 outbreaks each in the month of July, November and December. Maximum number of outbreaks were reported from Srikakulam district (12) followed by Vizianagaram (6) and Kurnool (3). Involvement of serotype O in 10 outbreaks and type A in 2 outbreaks were identified.

Karnataka (Bangalore): During the year under report 187 outbreaks were reported in the state, out of which 29 outbreaks were investigated. The rest of the outbreaks (158) were reported based on clinical signs and from these outbreaks samples could not be collected. Involvement of FMD virus type O could be identified in 22 outbreaks and rest of the outbreaks went undiagnosed. Sheep were the main species affected followed by cattle, Buffalo, Goat and pigs. The highest number of outbreaks were reported from Bagalkot (38) followed by Hassan (28), Gulbarga (21), Chikkaballapur (15), Bangalore rural (11), Mandya (11), Koppal (11), Raichur (10), Bangalore urban (5), Belgaum (5), Bijapur (5) and Kodagu (5).

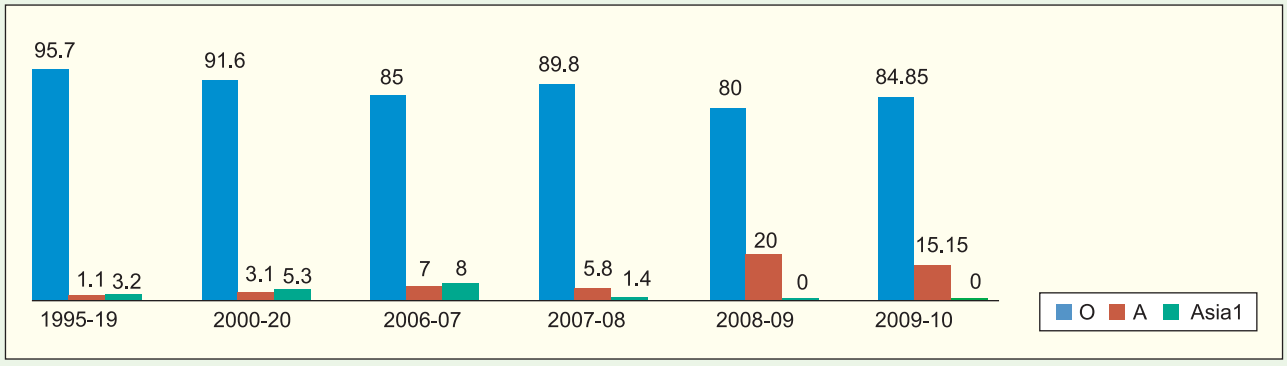
Kerala (Thiruvananthapuram): A total of 25 outbreaks were recorded in the state in all districts the state except Kazargode and Idukki. When compared to last year, though the number of outbreaks had gone down, the spread of the disease and morbidity rates were on the rise. More numbers of outbreaks were recorded during

Scenario of FMD in Southern Region

Susceptible animal population
(in millions)



Serotype-wise prevalence



November to January and this could be due to the increased animal movement from other state for slaughter purpose in the festival months. Random serosurveillance showed lower level of herd immunity. Cattle, goats and pigs were the species affected. Serotype A was identified in 6 outbreaks and rest of the outbreaks were caused by serotype O.

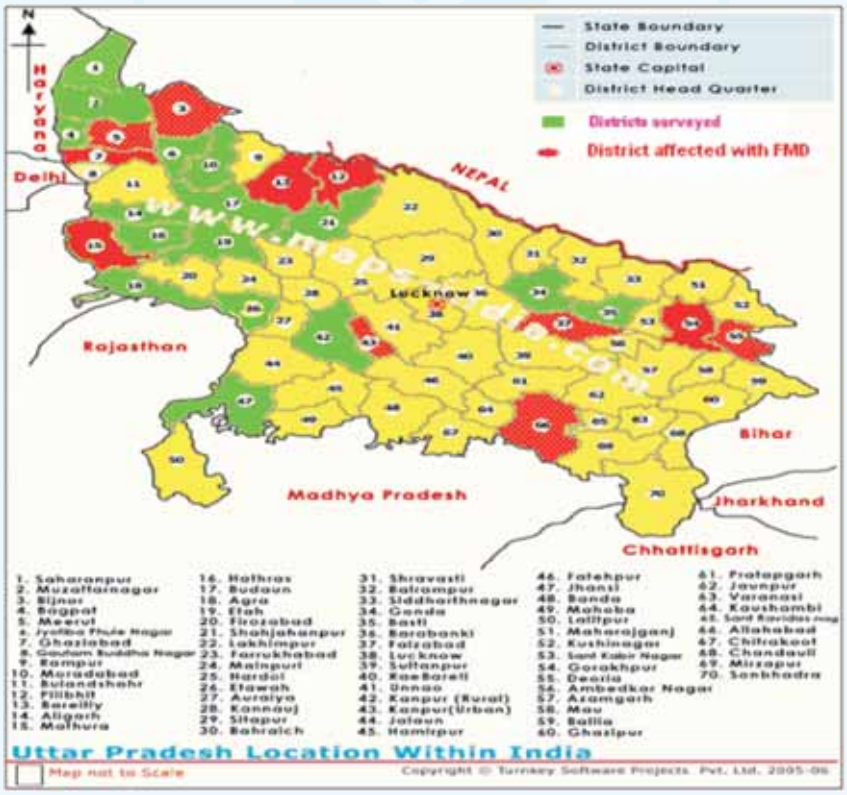
5.2 Northern Region

Jammu and Kashmir (Jammu): A Total 13 outbreaks (6 from Kashmir division and 7 from Ladakh) were reported in the J&K State. All outbreaks have been reported in bovines only. Maximum outbreaks have been reported from Kargil (6) and Baramulla (2) followed by Srinagar (1), Pulwama (1), Budgam (1) and Leh (1). Outbreaks occured in May, October, November and March. Clinical materials could not be collected from any of the outbreaks as no timely reporting of outbreak was received from the concern.

Punjab (Jalandhar): A single case of FMD was recorded in Gurukripa Farm in Village Langyana of District Moga. The farm had about 135 cattle and 12 buffaloes. FMD serotype A was diagnosed. The village has a livestock market for trading unproductive animals which are also brought from neighbouring states and the virus had probably spread from these animals. The isolates (IND 749/09 and IND 898/09) clustered in lineage VIIf of VP3[59] deletion group. Last year (2008-09), the state has not recorded any outbreak.

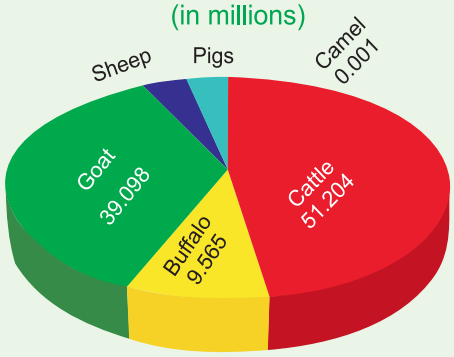
Haryana (Hisar): Two FMD outbreaks were recorded. One outbreak recorded in the month of January from Kurukshetra and another from Hisar district in March 2010. Only cross-bred cattle were affected, particularly, Holstein Frisian. Buffaloes and indigenous cattle remained unaffected in both these outbreaks. The outbreak in Kurukshetra was due to FMDV serotype A and FMDV type O was responsible for outbreak in Hisar.

Uttar Pradesh (Mathura): During the period under report, a total of 49 outbreaks

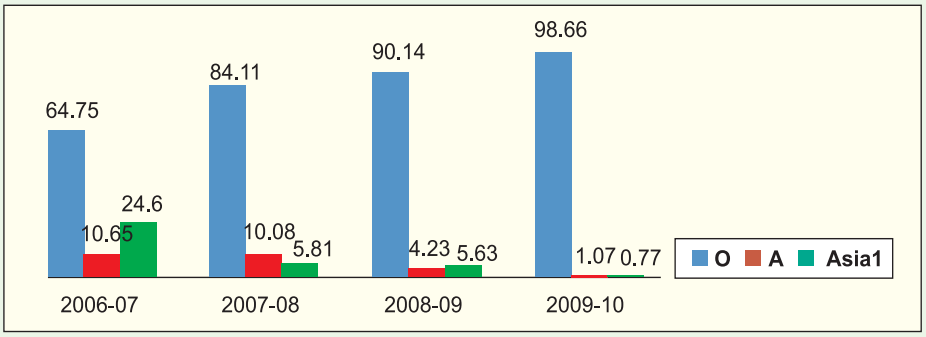


Scenario of FMD in Estern State

Susceptible animal population
(in millions)



Serotype-wise prevalence



which involved cattle and buffaloes were recorded. Maximum outbreaks occurred in the month of October, November and December. Outbreaks were recorded in districts Allahabad, Bareilly, Bijnore, Deoria, Faizabad, Gorakhpur, Kanpur, Pilibhit, Agra, Gaziabad, Mathura, Meerut and Muzaffarnagar. All the three serotypes were detected. Serotype O was involved in maximum numbers of outbreaks (32) followed by A (6) and Asia1 (5).

Himachal Pradesh (Shimla): A single sporadic outbreak of FMD was in District Solan and serotype O was responsible. For last two years, no outbreak was recorded in HP.

5.3 Western region

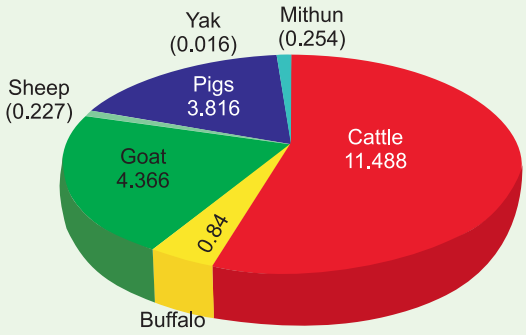
Gujarat (Ahmedabad): During the year, a total 20 outbreaks of FMD were recorded. Most of the outbreaks were recorded during the winter period of year. Five outbreaks were reported during February-2010. Ahmedabd, Rajkot and Vadodara region recorded 6, 9 and 5 outbreaks, respectively. (Outbreak wise diagnosis not provided in the report)

Maharashtra (Pune): During the year, a total 13 outbreaks of FMD were recorded. Maximum numbers of outbreaks were recorded in the month of December (5) followed by March (3) and October (2). All the three serotypes were detected. Typo O was responsible for 10 outbreaks, type A and Asia1 were responsible for 2 and 1 outbreaks, respectively.

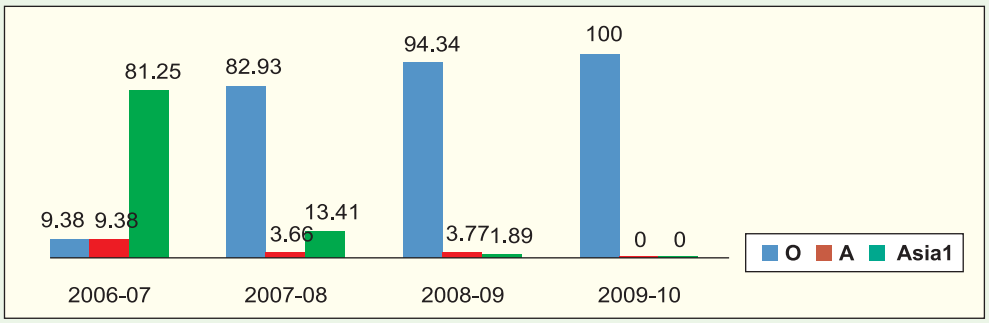
Rajasthan (Jaipur): During the year under report 35 outbreaks were reported/recorded in the state based on clinical signs. Only eight samples could be collected and all of them were found negative for FMDV.

Scenario of FMD in North Estern State

Susceptible animal population
(in millions)



Serotype-wise prevalence



5.4 Eastern Region

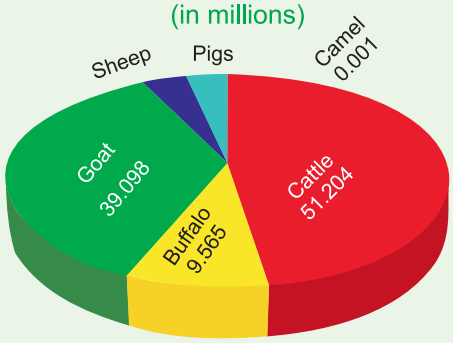
West Bengal (Kolkata): A total of 294 FMD outbreaks were reported/recorded during the period in the state. The highest number of FMD outbreaks were in Hooghly district (56) followed by North 24 Parganas (39), Jalpaiguri (29), Nadia (24), Howrah (22) and Coochbehar (22). Involvement of FMD virus O type was maximum and detected in 216 outbreaks. FMDV types A and Asia 1 were diagnosed in 1 outbreak each. Maximum numbers of outbreaks were reported during the month of September'09 (78) followed by August'09 (58), October'09 (43) and April'09(26).

Orissa (Cuttack): A total of 44 outbreaks were recorded/reported in the state during the period. Only ten outbreaks could be diagnosed and of which 7 were due to type O and 3 were caused by serotype A. Mostly Cattle consisting of local non-descript, crossbred Jersey and Haryana were affected. No disease was reported in Sheep, Goats and Pigs. Maximum number of outbreaks was reported from Cuttack (10) district. Highest numbers of outbreak was recorded in the month of August (9) followed by September (6) and November (6).

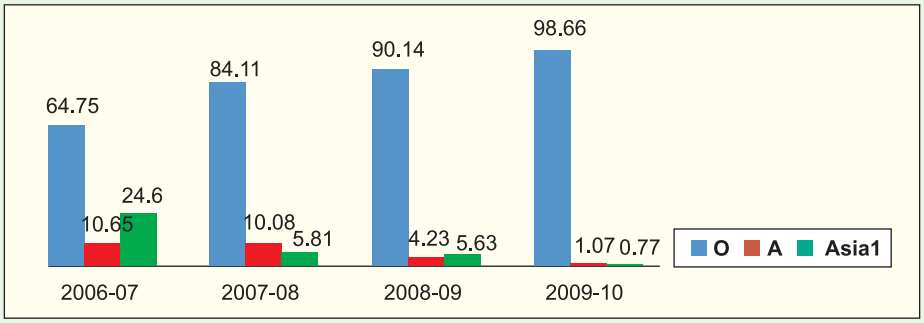
Bihar (Patna): During the period under report, a total of 158 outbreaks of FMD were recorded, of which 137 outbreaks were diagnosed to be due to serotype O. Serotype A and Asia 1 were not detected during the period. Highest number of outbreaks were recorded in the months of July (40), August (27), September (20), November (15), October (13) and December (11). Maximum number of the outbreaks were recorded in Patna (55), followed by Madhubani district (22), Madhepura (21), Supaul (15) and Nalanda (10).

Scenario of FMD in Estern State

Susceptible animal population
(in millions)



Serotype-wise prevalence



5.5 North Eastern Region

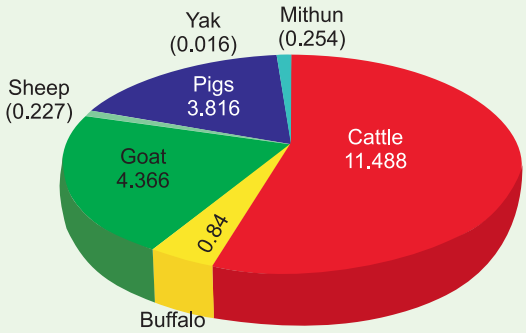
Assam (Guwahati): A total of 61 outbreaks of FMD were recorded in Assam and other N. E. States. The highest number (45) of outbreaks was in Assam followed by 8 outbreaks in Arunachal Pradesh, 5 outbreaks in Meghalaya and 1 outbreak each in Mizoram, Manipur, and Sikkim. In Assam, highest number (22) of outbreaks was recorded in Kamrup district. Cattle were affected in all the out breaks and in only two outbreaks, pigs were also affected along with cattle. FMDV serotype O was diagnosed as causative agent for 30 outbreaks in Assam, 8 in Arunachal, 5 in Meghalaya, 1 each in Sikkim, Mizoram and Manipur. The highest number of out breaks was recorded in the months of September 09 (10 outbreaks) like the previous year (2008-09) followed by August and February 10

Mizoram (Aizwal): During the period under report 9 FMD outbreak/incidence were recorded from three districts of Mizoram (Aizawl, Serchhip and Mamit). The outbreaks were recorded in the month of June, July and January. In two outbreaks which occurred during June and July 2009, both species of porcine and bovine including Mithun were affected leading to high mortality among Mithun population. Involvement of FMDV serotype O in eight outbreaks was identified.

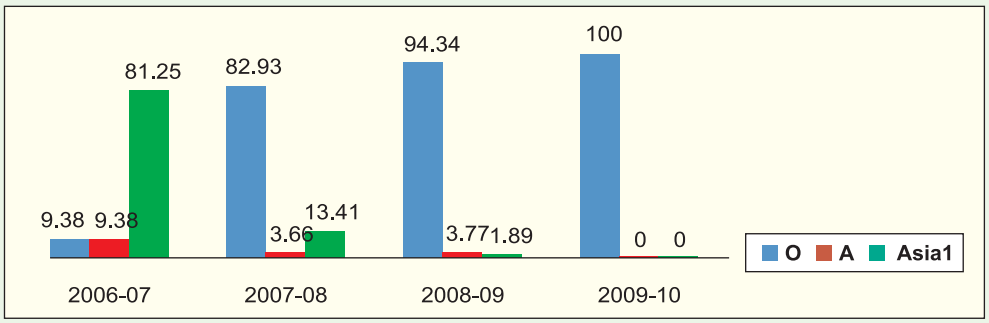
Tripura (Agartala): A total of 27 outbreaks were recorded in the state which involved cattle and Pigs. Twelve outbreaks could be diagnosed using sandwich ELISA and 3 outbreaks were diagnosed retrospectively using LPB ELISA. All the fifteen outbreaks were caused by FMDV serotype O virus. The occurrence of FMD was recorded in almost every month except the month of November, December and March.The outbreaks were recorded in all the 4 districts of the State of Tripura. FMD outbreaks involving

Scenario of FMD in North Estern State

Susceptible animal population
(in millions)



Serotype-wise prevalence



type O virus have been recorded constantly in the State since 1986 but during the year, 2008-09 type A was also detected

Arunachal Pradesh (Itanagar): A total of 18 outbreaks of FMD were recorded during the period under report. Besides Cattle, Mithun and Yak species were also involved. Highest number of outbreaks was recorded in the months of November (6), July (4) and June (2). Maximum number of the outbreaks were recorded in P.Pare (7), followed by E.Siang (3) and L. Subansiri (2). Serotype O was diagnosed to be causative agent in 13 outbreaks.

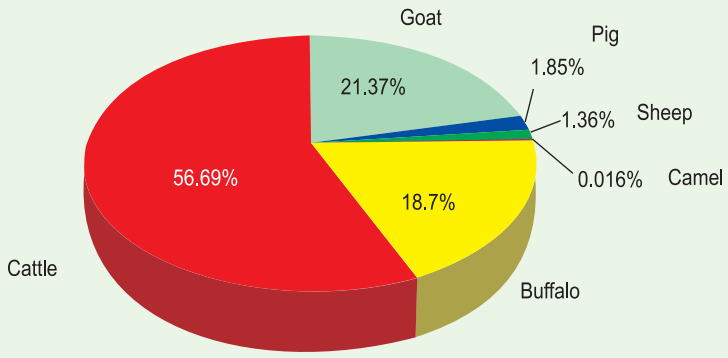
Manipur (Imphal): During the year, 3 outbreaks of FMD in two neighbouring districts of the state at three small villages (Liwachangning, Sarei village of Chandel district and Sugnu Ward No.4 and 5 of Thoubal district) were recorded. Involvement of serotype O could be diagnosed in two outbreaks. The outbreak occurred during the first week of January, 2010 and affected only Cattle. The source of infection was speculated to be the illegal import of buffaloes from Myanmar during Christmas festival in the month of December, 2009. There was absence of serotypes of A and Asia 1 in North Eastern region during the period.

5.6 Central Region

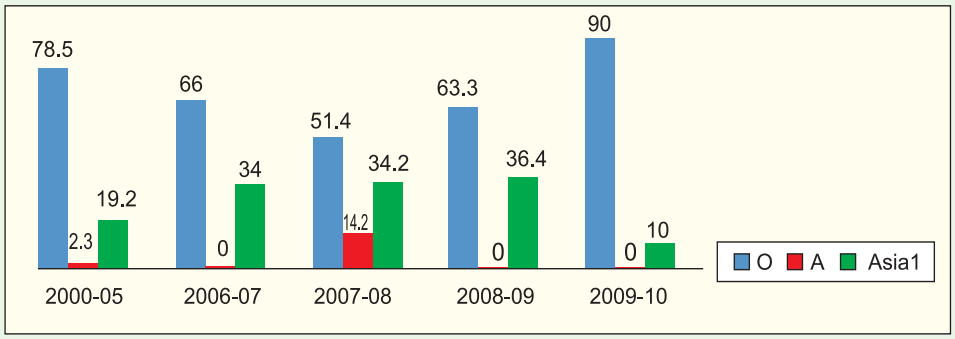
Madhya Pradesh (Bhopal): A total of 20 outbreaks reported in Madhya Pradesh and of which 4 outbreaks were reported from Shivpuri, 3 outbreaks were reported from Seoni, 2 from Balaghat, Chindwara, Indor, Mandla and Ujjain each, 1 each from Betul, Rewa and Vidisha district. Eighteen outbreaks were due to type O and 2 were due to type Asia1. Maximum number of outbreaks were reported in the month of March (8), followed by February (7) and October (4).

Scenario of FMD in Central Region

Susceptible animal population
(in millions)



Serotype-wise prevalence



6.0 Virology and Molecular Epidemiology

6.1 Processing of field samples

A total of 1155 clinical materials from 549 outbreaks were received from centres and network units of the project during the year 2009-10. Maximum number of clinical samples were received from West Bengal followed Bihar and Assam. A large proportion of samples collected from Bovine species. The samples were processed using chloroform and made in to suspension in PBS. The processed materials were subjected to sandwich ELISA, and ELISA negative samples (39) were tested using multiplex PCR for virus diagnosis. FMDV serotypes could be identified in 445 outbreaks (Table 5). FMDV type O could be found in maximum numbers of outbreaks (423) and Serotype A and Asia1 were detected in 15 and 7 outbreaks, respectively.

6.2 Genetic and antigenic characterization of field isolates

6.2.1 Type O FMD Virus

Molecular Epidemiology

Genetic characterization of FMDV serotype O has been one of the fundamental objectives in the molecular epidemiology of FMD virus due to its wide prevelence. This has resulted in establishing of genetic identity of the virus, tracing the movement of virus across the state boundaries, reemergence of older strains, and introduction of new strains, if any. Compared to recent past, the year 2009-2010 saw a major shift in genetic lineage of serotype O viruses circulating in India. The viruses of Ind2001 lineage gained upper hand after a gap of 8 years and outcompeted PanAsia II lineage. Reemergence of this group has been tracked back to 2008, during which it caused sporadic outbreaks in Northern India. Soon it spread to majority of states in Eastern, Central and reached Southern state of Kerala at the end of 2009, traveling through Andhra Pradesh, Karnataka (Fig.2). Nevertheless the disease due to PanAsia 2 (Fig.3), PanAsia1 (Fig.4) and Branch C-II (Fig.5) viruses were not completely absent.

During the period under report, a total of 116 type O virus isolates were subjected to 1D genomic region sequencing either directly from tongue epithelium or cell culture supernatant. The viruses selected covered 15 of the 18 states from which type O samples have been received at Central FMD Laboratory. The states covered included Kerala, Karnataka, Andhra Pradesh, Orissa, Jharkhand, Bihar, Uttar Pradesh, Uttarakhand, Gujarat, West Bengal, Assam, Arunachal Pradesh, Meghalaya, Manipur and Tripura. Results from the phylogenetic analysis shows that Ind2001 viruses, which reemerged in late part of year 2008, spread to majority of states in Northern, Eastern, North-Eastern and Southern India. Fourteen states out of 18 states where disease due to type O was experienced were traced back to this lineage. These viruses diverged (genetically) from Ind2001 viruses isolated in 2001 by 7.3% and 13.1% compared to the in-use vaccine virus (O/IND/R2/1975). Analysis of complete capsid coding region sequenced directly from tongue epithelium showed that the viruses isolated from Thrissur, Kerala (IND830/2009) had unique amino acid changes at position 61 (Asparagine to Serine), 108 (Threonine to Isoleucine), 159 (Serine to Proline), 376 (Valine to Threonine), 662 (Glycine to Serine), 663 (Serine to Aspartic Acid), 720 (Asparagine to Serine). Though none of these positions fall in 5 known antigenic sites of FMD,

Table 5. Details of the field materials received at PD FMD collected during the year 2009-10

S. No.	Centre	Samples Received	Virus types detected at PD on FMD				Source host species for FMD positive samples					
			O	A	Asia1	NVD	Cattle	Buffalo	Sheep\ Goat	Pig	Wild life	NA
1	Agartala	8	5	-	-	3	7	-	-	1	-	-
2	Ahmedabad	17	5	-	6	6	8	-	-	-	-	9
3	Bangalore	27	19	-	-	8	22	3	1	-	1	-
4	Bhubaneswar	23	12	2	-	9	18	-	-	-	-	5
5	Guwahati	47	33	-	-	14	43	-	-	-	-	4
6	Hyderabad	16	10	4	-	2	16	-	-	-	-	-
7	Haryana	2	-	2	-	-	2	-	-	-	-	-
8	Itanagar	18	13	-	-	5	11	-	-	-	7	-
9	Imphal	2	2	-	-	-	2	-	-	-	-	-
10	Jaipur	3	-	-	-	3	3	-	-	-	-	-
11	Jalandhar	2	-	2	-	-	2					
12	Kolkata	262	235	-	-	27	257	1		3	-	1
13	Kohima											
14	Mathura	15	9	3	1	2	15	-	-	-	-	-
15	Patna	76	60	-	-	16	76	-	-	-	-	-
17	Ranipet											
18	Shimla	2	2	-	-	-	2	-	-	-	-	-
19	Thiruvananth-apuram	21	13	-	-	8	11	-	-	-	-	10
20	CADRAD&UP	6	4	1	-	1	6	-	-	-	-	-
21	UK	2	1	1	-	-	2	-	-	-	-	-
	Sub Total	**549**	**423**	**15**	**7**	**104**	503	4	1	4	8	29
	Total			**445**								

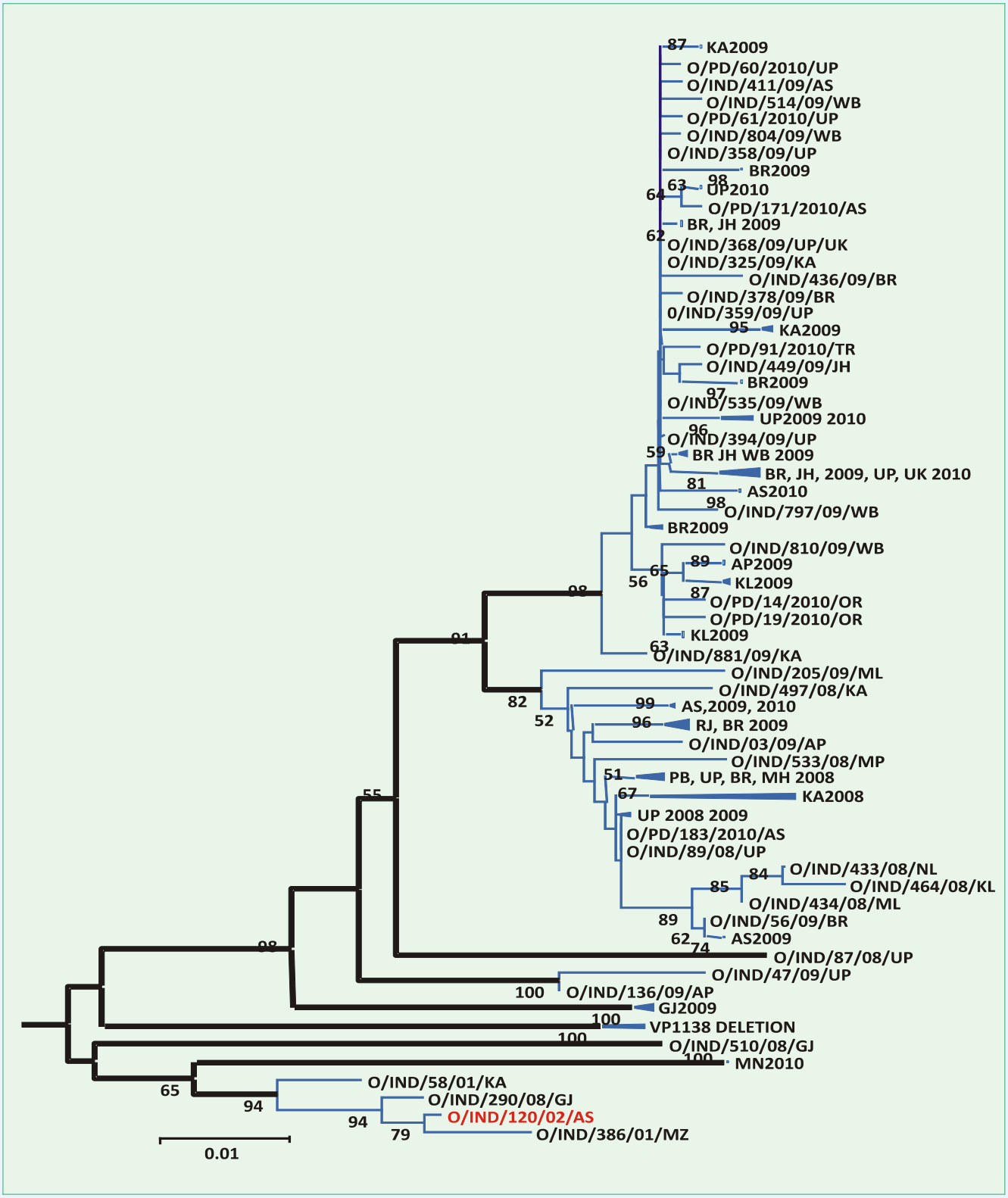


Figure 2. 1D genomic region based phylogenetic tree showing relationship among Ind2001 viruses with other Serotype O virus lineages circulating in India during 2008-2010. Candidate vaccine strain (IND120/2002) is underlined. Tree indicates that viruses evolved over a period of time by selection in to competing population and regained dominance by multiple passages in naive susceptible population

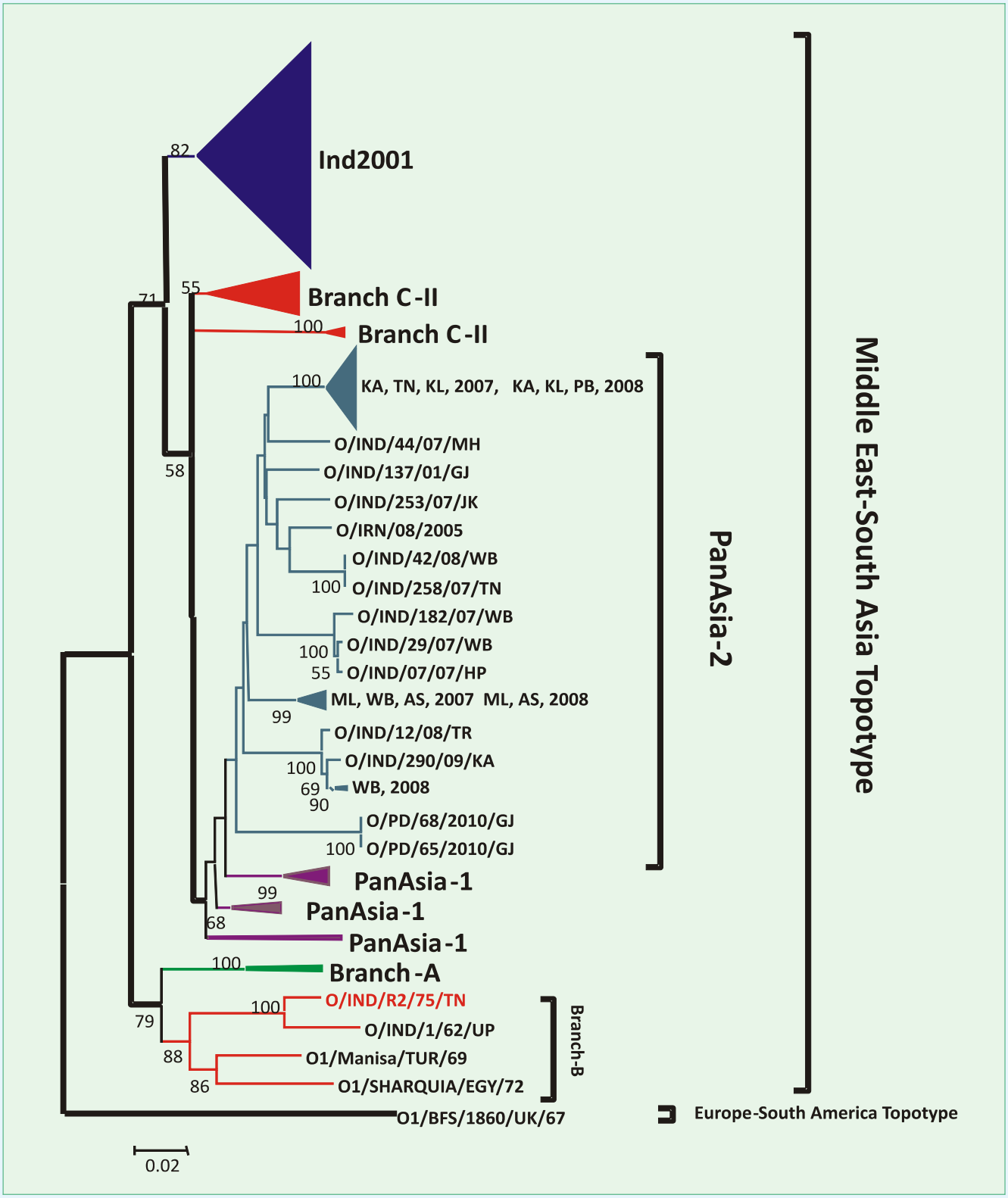


Figure 3. 1D genomic region based phylogenetic tree showing relationship among PanAsia2 viruses with other Serotype O virus lineages circulating in India during 2008-2010. The tree indicates that vruses of PA2 lineage have been still persists in southern peninsular region of Karnataka. However the viruses are new introduction in the Gujrat state

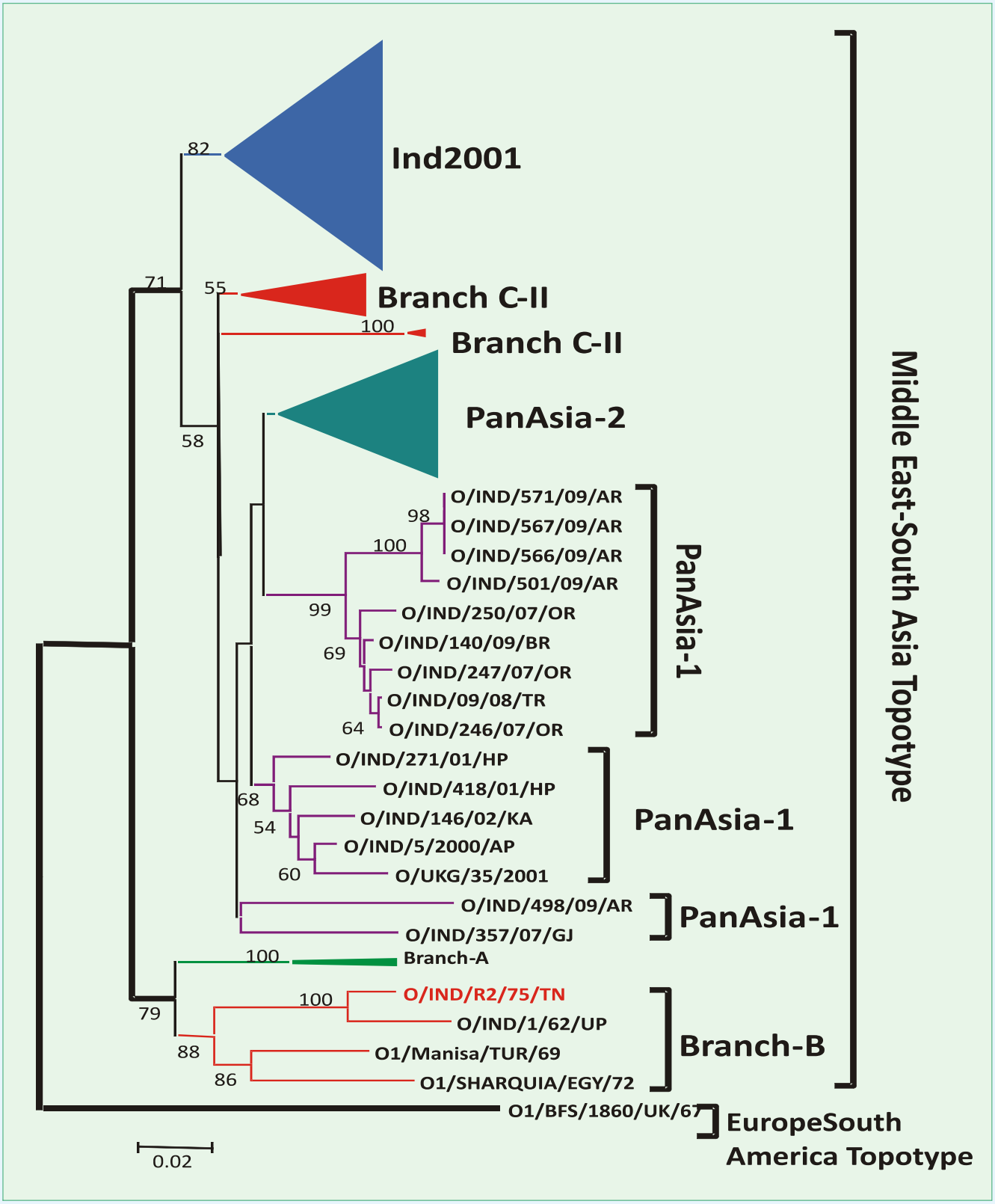


Figure 4. 1D genomic region based phylogenetic tree showing relationship among PanAsia1 viruses with other Serotype O virus lineages circulating in India during 2008-2010. Though PanAsia1 viruses have already been recovered from Arunachal Pradesh viruses responsible for current outbreaksa are of different origin

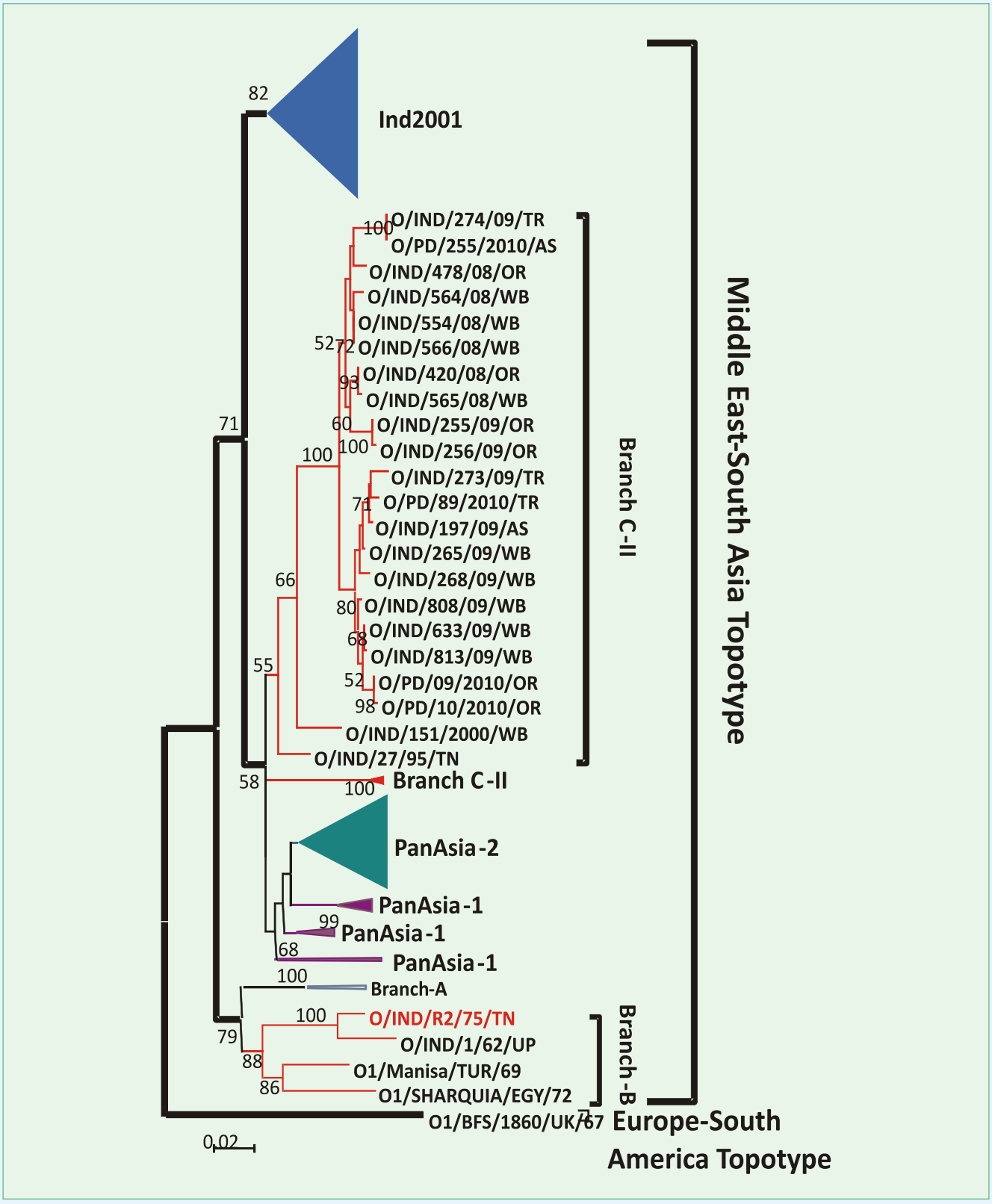


Figure 5. 1D genomic region based phylogenetic tree showing relationship among Branch C-II viruses with other Serotype O virus lineages circulating in India during 2008-2010. The viruses belonging to Branch cII lineage were already persist in the eastern India continue to involve in outbreak in the current year also

position 662 (Glycine to Serine) has been implicated in ($VP1_{139}$) neutralization resistance [E R Rojas, E Carrillo, M Schiappacassi, and R Campos (1992). Modification of foot-and-mouth disease virus O1 Caseros after serial passages in the presence of antiviral polyclonal sera. J. Virol. June 1992 66: 3368-3372]. Nevertheless these viruses are antigenically covered by the currently used vaccine virus as estimated in 2D-MNT (r=0.69). However, even after the reemergence and dominance by Ind2001 lineage, disease due to other lineages was not completely absent. Next to Ind2001 lineage, disease due to Branch-C II lineage (Fig 4) viruses has been recorded in Tripura, Assam, West Bengal and Orissa during 2009-2010. PanAsia II was responsible for sporadic cases in Karnataka and Gujarat. There were outbreaks of FMD in Mithun in Arunachal Pradesh due to PanAsia I; this lineage also caused outbreaks in Bihar. This indicates complex epidemiological scenario of FMD virus in India, which is not surprising because of large population of susceptible animals of different species, sparse vaccination and unrestricted movement of animal. However, prompt surveillance system available and in place in our country, is able to detect the introduction of exotic strains of virus immediately, which is assuming importance since there is more number of FMD outbreaks due to serotype O, Topotype Southeast-Asia, Mya-98 lineage has been recorded in Far East FMD previously free countries such as Japan and Republic of Korea in early part of 2010 [Paton DJ, King DP, Knowles NJ, Hammond J. (2010). Recent spread of foot-and-mouth disease in the Far East. Vet Rec. 2010 May 1; 166(18):569-70].

Antigenic Characterization

During the year 2009-2010, 20 viruses from Assam, Rajasthan, Uttar Pradesh, Arunachal Pradesh, West Bengal, Andhra Pradesh, Kerala, Orissa and Manipur were subjected to antigenic analysis (Fig. 6). All the 20 viruses were found to be antigenically related to currently used vaccine strain (O/IND/R2/1975). Among various lineages, r value ranged from 0.41 to 1.0. The PanAsia 2 viruses isolated in 2008 from Assam had an r-value of 1.0. The vale of 1.0 was obtained also for PanAsia 1 viruses isolated from Mithun and Cattle in Arunachal Pradesh. The IND 2001 viruses responsible for majority of outbreaks in Rajasthan, Uttar Pradesh, Assam, West Bengal, Andhra Pradesh, Kerala, Orissa, and Manipur were also antigenically closer to the

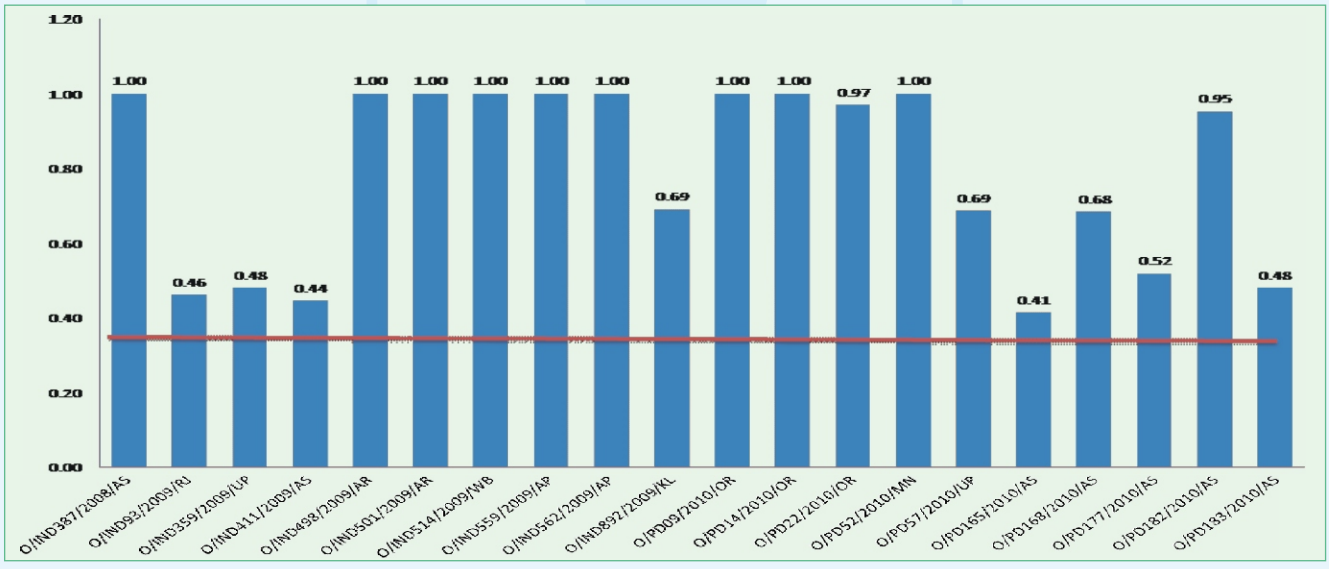


Figure 6. r-value (in 2D-MNT) of FMDV Serotype O Isolates recovered during 2008-2010 with in-use vaccine virus (INDR2/75)

currently used vaccine strain (O/IND/R2/1975) as indicated by their r-value of 0.44 to 1.0. To conclude the presently circulating field viruses of FMDV serotype O are protected/antigenically covered by in-use vaccine virus.

6.2.2 Type A FMD Virus

Molecular Epidemiology

Among all serotypes prevalent in India, type A virus population is found to be genetically and antigenically most heterogeneous in nature. VP1 coding (1D) region based molecular phylogeny has established circulation of four genotypes {showing more than 15% nucleotide (nt) divergence among them at 1D region} of type A so far in India. Since 2001, genotype VII has been exclusively responsible for all the field outbreaks and has outcompeted all other genotypes. Within the currently circulating genotype VII, a divergent and unique lineage emerged in late part of 2002, which showed an amino acid (aa) deletion at 59th position of VP3 (VIIb-VP3^{59} deletion group) and dominated the field outbreak scenario in 2002-03. Ever since then sporadic outbreaks due to this lineage has been documented. During 2007-08, there was once again an upsurge in incidence of outbreaks due to this lineage. This single aa deletion is at an antigenically critical position in the structural protein VP3, which is considered to be a major evolutionary jump probably due to immune selection in the scenario of sparse vaccination.

During the period under report, 7 field isolates of serotype A recovered from 6 different outbreaks in Punjab, Orissa, Andhra Pradesh and Uttarakhand were sequenced at complete 1D (VP1) region for molecular epidemiological analysis. As a matter of fact all the reported and documented field outbreak viruses detected to be type A could be sequenced and analysed. For the isolates IND 1/2010 from Uttarakhand, IND 332/09 and IND 333/09 from Andhra Pradesh and IND 744/09 and IND 747/09 from Orissa, complete capsid coding region (P1) sequence was also resolved. The determined sequences were aligned with other Indian sequences available in the data base of PD on FMD. During 2009-2010, all the isolates were found to cluster within genotype VII in the N-J tree (Fig. 7), but grouped both in the nondeletion and the VP3^{59} deletion lineages. Precisely, the viruses from Punjab (IND 749/09 and IND 898/09) and Uttarakhand (IND 1/2010) shared ancestry and clustered in the same VP3^{59} deletion lineage, VIIf, where as viruses from two different outbreaks from Orissa (IND 744/09 and IND 747/09) and those from Andhra Pradesh (IND 332/09 and IND 333/09) clustered together and revealed no deletion in the VP3 coding region. Hence, it is observed that both deletion and nondeletion mutants belonging to genotype VII are co-circulating in the field in recent times. The viruses from Andhra Pradesh and Orissa as well as those from Punjab and Uttarakhand appear to be epidemiologically related strains as they show less than 2% nucleotide divergence among themselves.

This finding corroborates with the collected history of animal movement between these states. 1D region based phylogeny has also revealed that this deletion group is genetically diverging with time giving rise to three lineages (VIIb, VIIf & VIIg) which show more than 5% nucleotide divergence among themselves. This is indicative that the deletion group is in the phase of continuous active evolution without intermittent stasis.

Complete genome sequencing and sequence analysis of the current type A vaccine strain virus (IND 40/00)

The complete molecular characterization of the in use vaccine strain (IND 40/00) including both antigenic and genetic is essential in understanding the genetic and evolutionary relationship with currently circulating field isolates which is of importance in terms of molecular epidemiology and FMD control programs. The sequence and phylogenetic analysis exhibited that the IND 40/00 belongs to genotype VII and

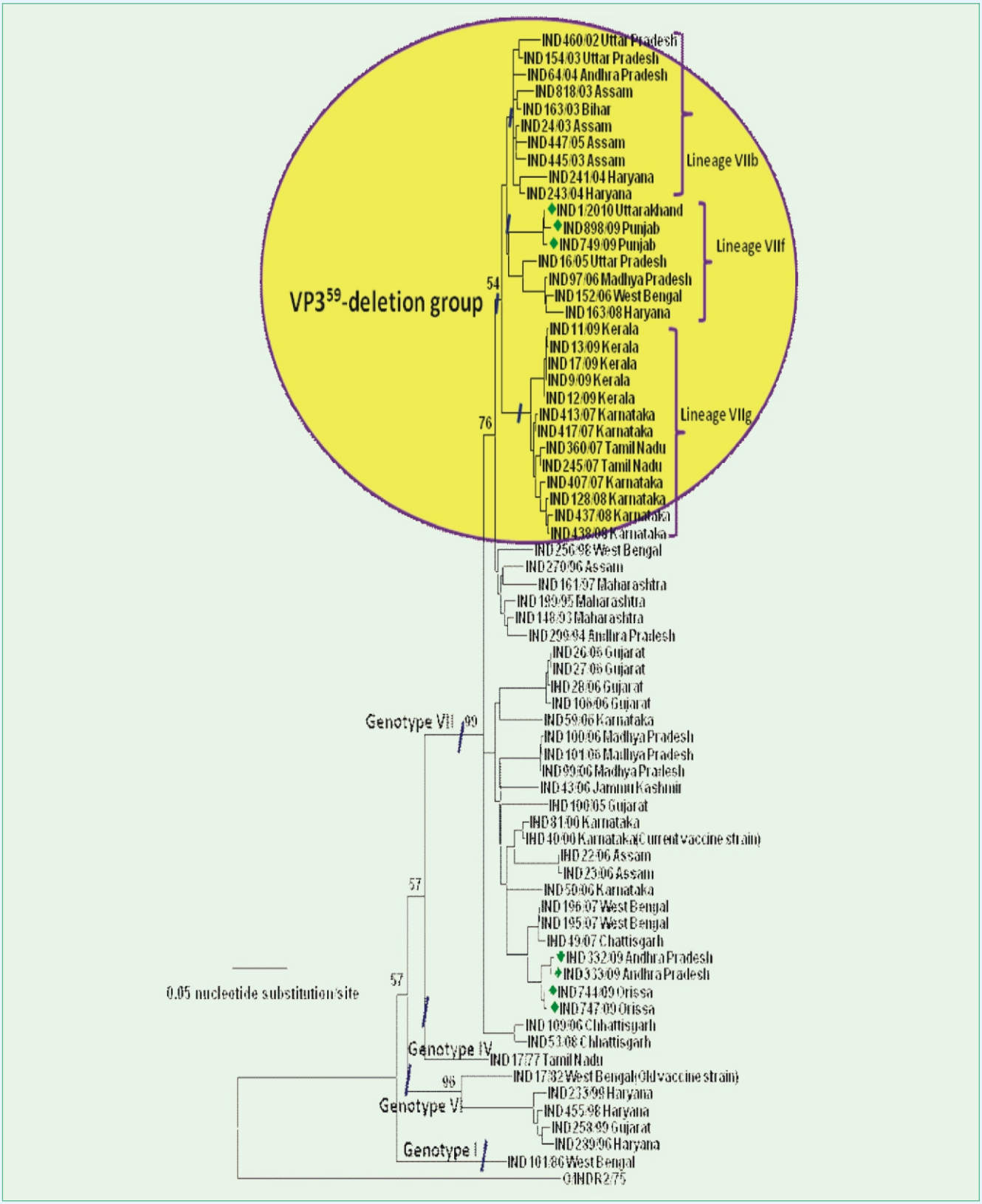


Fig 7. Phylogenetic relationship among type A FMD virus isolates at 1D region. Isolates sequenced in the reported period are marked with green rhombus and type O sequence is included as an outgroup

is similar to IND81/00. Moreover IND40/00 shares common ancestry with Middle East virus sequences and shows about 11% average nucleotide divergence with Middle East isolates (Fig 8). The complete genome nucleotide sequence of IND 40/00 is given below.

SFUTR

TTGAAAGGGGGCGCTAGGGTCTCACCCCTAGCATGCCACCGGCAGCTCCTGCGTTGCACCCCACACTTAC
GCTCGTGCACTCGCGGGAACCGATGGACTGTCGTTCACACCTACAGCTGGACTCACAGCACCGCGTGGCC
ATTTTAGCTGGATTGTGCGGACGAACAACGCTTGCGCATCTCGCGTGACCGTTTAGTACTCTTACCATTTTCCG
CCTACTTGGTCGTTAGCGCTGCCTTGGGCACTCCTTTGGGGGCCGTCCGACGCTTCACGGTCTCCCCCGTGTGACGGACTACGGTGATGGGGC
CGCCTCGTGCGAGTT GGTCGTTTGGTGTGCTTTGGCTGTC ACCCGGCGCCC GCCTTTCAT

LFUTR

CCCCCCTAAGTTTTACCGCGTTCCCGGCGTTAAAGGGATGTAACCACAAGATAAACCTTCACCCGAAGTAAAACGGC
AACCACACTCAGTTTTGCCCGTTTTCATGAGAAATGGGACGTCTGCGCACGAAACGCGCCGTCGCTTGAGGAT
GACTTGTACAAACACGATCTAAGCAGGTTTCCACAACTGACACACACCGTGCAATTTGAAACTCCGCCT
GGCCTTTCCAGGTCTAGAGGGGCGACACTTTGTACTGTGATTGACTCCACGCTCGGTCCACTGGCGGGTGC GAGTAACAGGACTGTTGCTTCGTAGCG
GAGCGTGTTGGCCGTGGGATTCCCTCCTTGGTAACAAGGACCCACGGGGCCGAAAGCCACGTCCTAACGGACCCAACATGT
GTGCAACCCCAGCACGGCAACTTTACTGTGAAAACCACTTTAAGGTGACACTGATACTGGTACTCAACCACTGGTGACAGGCT
AAGGATGCCCTTCAGGTACCCCGAGGTAACACGCGACACTCGGGATCTGAGAAGGGGACTGGGGCTTCTTTAAAGCGCCCAG
TTTAAAAAGCTTCTATGCCTGAATAGGCG ACCGGAGGCCGGC GCCTTTCTTTTACCATTACTAAACTG

L

ATG AAT ACG ACT GAC TGT TTT ATC GCT CTG TTA TAC GCT ATC AGA GAG ATC AAA ACA CTT TTT CGC TCA CGG ACA CAA GGA GAG ATG GAA TTC ACA
CTC TAC AAC GGT GAG AAA AAG ACT TTC TAT TCT AGA CCC AAC AAC CAC GAC AAC TGT TGG CTG AAC ACC ATC CTT CAG TTA TTC AGA TAT GTC GAT
GAA CCA TTC TTC GAC TGG GTT TAT GAT TCA CCT GAG AAC CTC ACT CTT GAG GCG ATC AAA CAA TTG GAG GAA CTC ACT GGT CTT GAG CTG CAC GAG
GGC GGA CCA CCT GCT CTC GTC ATT TGG AAC ATC AAG CAT CTG CTC CAC ACC GGA ATC GGC ACC GCC TCG CGA CCC AGC GAG GTG TGT ATG GTT
GAT GGT ACG GAC ATG TGC TTG GCT GAC TTT CAT GCT GGC ATC TTC CTG AAA GGG CAA GAG CAT GCT GTG TTC GCC TGT GTC ACC TCC GAT GGG
TGG TAC GCG ATT GAT GAC GAG GAC TTT TAC CCT TGG ACA CCG GAC CCG TCC GAT GTT CTG GTT TTT GTC CCG TAC GAT CAA GAA CCA CTT AAT GGA
GAA TGG AAG ATG AAG GTT CAG AAG CGA CTC AAG

VP4

GGA GCT GGG CAA TCC AGT CCG GCG ACC GGG TCG CAG AAC CAG TCA GGT AAC ACT GGA AGC ATC ATT AAC AAC TAC TAC ATG CAA CAA TAC CAG
AAT TCT ATG GAC ACA CAA CTT GGG GAC AAC GCC ATT AGC GGA GGT TCC AAC GAG GGT TCC ACA GAC ACC ACT TCC ACC CAC ACA ACC AAC ACC
CAA AAC AAC GAT TGG TTC TCA AAA TTG GCC AGC TCT GCC TTT AGC GGA CTT TTC GGC GCT CTT CTC GCC

VP2

GAC AAG AAG ACC GAG GAG ACT ACT CTC CTG GAG GAC CGC ATT CTT ACC ACC CGC AAC GGA CAC ACT ACC TCC ACA ACT CAA TCG AGT GTA GGA
GTC ACC TAC GGG TAC TCC ACT GGA GAG GAC CAC GTT TCC GGA CCC AAC ACA TCT GGT TTG GAA ACG CGG GTG GTG CAG GCA GAA AGA TTC TTT
AAG AAG CAC CTG TTT GAC TGG ACA GCG GAC AAG GCA TTT GGA CAC TTG GAA AAG CTG GAA CTC CCC ACT GAC CAC AAG GGC GTC TAC GGA CAC
CTG GTG GAC TCT TTT GCT TAC ATG AGG AAT GGC TGG GAC GTG GAG GTG TCT GCC GTT GGC AAC CAG TTC AAC GGC GGA TGT CTC CTC GTG GCC
ATG GTT CCT GAG TGG AAG GAG TTC ACC ACG CGT GAG AAG TAC CAG CTC ACC TTG TTC CCC CAC CAA TTC ATT AAC CCC AGA ACC AAC ACG ACC
GCT CAC ATC ACG GTT CCC TAC CTT GGT GTG AAC CGG TAC GAC CAG TAC AAG AAG CAC AAA CCA TGG ACG TTG GTT GTA ATG GTG GTT TCG CCG
CTT ACC AAT GCC GGC ATT GGT GCC ACT CAA ATC AAG GTC TAC GCC AAC ATC GCC CCG ACC TAC GTC CAC GTG GCC GGT GAG CTC CCG TCG AAA
GAG

VP3

GGG ATC GTA CCG GTT GCG TGT GCG GAC GGT TAT GGC GGT TTG GTG ACC ACG GAC CCT AAA ACA GCT GAC CCT GTT TAT GGC ATG GTG TAC AAC
CCC CCT CGG ACA AAT TTT CCT GGG CGG TTT ACA AAC CTG TTG GAC GTG GCG GAG GCC TGC CCC ACC TTC CTC TGT TTC GAC AAC GGG AAA CCG
TAC GTT GAG ACA AGA ACG GAT GAG CAA CGT CTT CTG GCA AAA TTC GAC GTT TCA TTG GCT GCA AAA CAC ATG TCA AAC ACC TAT CTT TCA GGG
ATA GCA CAG TAC TAC GCA CAG TAC TCT GGC ACC ATC AAC CTC CAC TTC ATG TTT ACT GGC TCC ACT GAC TCA AAG GCC CGC TAC ATG GTG GCG
TAC GTC CCG CCT GGT GTG GAA ACA CCG CCG GAC ACG CCT GAG AAA GCT GCA CAC TGC ATC CAC GCT GAG TGG GAC ACA GGG TTG AAC TCC AAA
TTC ACC TTT TCT ATC CCG TAC GTG TCT GCT GCA GAC TAC GCG TAC ACC GCG TCT GAC ACG GCA GAA ACA ACA AAC GTA CAG GGA TGG GTC TGC
ATT TAC CAG ATC ACC CAC GGG AAG GCT GAA AAT GAC ACT CTG GTC GTG TCG GCC AGC GCC GGC AAA GAC TTT GAG TTG CGT CTC CCG ATT GAC
CCC CGC GCA CAA

VP1

ACC ACC ACG GCC GGG GAA TCT GCA GAC CCA GTC ACC ACC ACT GTT GAG AAC TAC GGC GGT GAG ACA CAA GTC CAG CGG CGT CAC CAC ACT GAC
GTC AGC TTC ATA ATG GAC AGA TTT GTG AAG ATT GGA ACC ACT AAC CCC ACA CAT GTC ATT GAC CTC ATG CAA ACC CAC CAA CAC GGG CTG GTG
GGT GCC CTG TTG CGT GCT GCC ACG TAC TAC TTC TCC GAC TTG GAG ATC GTG GTT CGT CAC AGC GGC AAC CTG ACA TGG GTA CCC AAT GGA GCA
CCT GAG GCA GCC CTG TCC AAC ACA GGA AAC CCT ACC GCC TAC AAC AAA GCG CCG TTC ACG AGA CTT GCG CTC CCC TAC ACT GCG CCA CAC CGC
GTG TTG GCA ACA GTG TAC AAC GGG ACG AAC AAG TAC TCC GCG GCC AGT GGG CGC ACA CGG GGT GAC CTG GGG CAG CTC GCA GCG CGA ATT
GCC GCC CAA CTT CCT GCC TCT TTC AAT TTT GGT GCA ATC AAG GCT GAC GCC ATC CAC GAG CTT CTC GTG CGC ATG AAG CGT GCC GAA CTC TAC
TGC CCC AGA CCA CTG TTG GCG GTG GAG GTC TCG TCT CAA GAC AGA CAC AAA CAG AAG ATC ATT GCA CCA GCA AAA CAG CTC TTG

2A

AAC TTT GAC CTG CTC AAG TTG GCG GGA GAC GTT GAG TCC AAC CCC GGG

2B

CCC TTC TTT TTC TCC GAC GTT AGG TCG AAC TTC TCC AAA CTG GTG GAG ACC ATC AAC CAG ATG CAG GAA GAC ATG TCA ACA AAG CAC GGA CCT GAC TTT AAC CGG TTG GTG TCC GCA TTT GAG GAA CTG GCC ACT GGA GTG AAG GCT ATC AGG ACC GGT CTC GAC GAG GCC AAA CCC TGG TAC AAA CTT ATC AAA CTC CTG AGC CGT CTG TCG TGC ATG GCC GCT GTA GCA GCA CGG TCA AAG GAC CCA GTC CTT GTG GCC ATC ATG CTA GCT GAC ACC GGT CTC GAG ATT CTG GAC AGC ACC TTT GTC GTG AAG AAA ATC TCC GAC TCG CTC TCC AGT CTC TTT CAC GTG CCG GCC CCC GTC TTC AGT TTT GGA GCT CCA ATC CTG TTG GCC GGG TTG GTC AAG GTC GCC TCG AGT TTC TTC CGG TCC ACA CCC GAA GAC CTT GAG AGA GCA GAA AAA CAG

2C

CTC AAA GCA CGT GAC ATC AAC GAC ATA TTC GCC ATT CTC AAG AAC GGC GAG TGG CTG GTC AAG CTG ATT CTT GCC ATC CGC GAC TGG ATT AAG GCA TGG ATC GCC TCA GAA GAA AAG TTT GTC ACC ATG ACA GAC TTG GTG CCT GGC ATC CTT GAA AAG CAG CGG GAC CTC AAC GAC CCG AGC AAG TAC AAG GAG GCC AAG GAG TGG CTC GAC AAC GCG CGC CAA GCG TGC TTG AAG AGC GGG AAC GTC CAC ATT GCC AAC CTT TGC AAA GTG GTC GCC CCG GCA CCC AGC AAG TCG AGA CCC GAA CCT GTG GTC GTT TGC CTC CGT GGC AAG TCC GGC CAG GGT AAG AGT TTC CTT GCG AAC GTG CTC GCG CAA GCA ATC TCC ACC CAC TTC ACT GGC AGA ACC GAT TCA GTT TGG TAC TGC CCG CCT GAC CCC GAC CAC TTC GAC GGT TAC AAC CAG CAG ACC GTT GTT GTG ATG GAT GAT TTG GGC CAA AAC CCT GAT GGC AAG GAC TTT AAG TAC TTT GCC CAG ATG GTT TCC ACC ACA GGG TTC ATC CCG CCC ATG GCC TCA CTC GAA GAC AAA GGC AAA CCT TTC AAC AGC AAG GTT ATC ATA GCC ACC ACC AAC CTG TAC TCG GGG TTC ACC CCG AGA ACC ATG GTG TGC CCT GAT GCA CTG AAC CGC AGG TTT CAC TTT GAC ATT GAC GTG AGC GCT AAG GAC GGG TAC AAA ATT AAC AAC AAA TTG GAC ATA ATC AAA GCT CTT GAG GAC ACC CAC ACC AAT CCC GTG GCA ATG TTT CAA TAT GAC TGT GCC CTT CTC AAC GGC ATG GCC GTT GAA ATG AAG AGA ATG CAA CAA GAC ATG TTT AAG CCT CAG CCA CCC CTC CAG AAC GTG TAC CAG CTC GTT CAG GAG GTA ATT GAA CGG GTC GAG CTC CAC GAG AAA GTG TCG AGC CAC CCG ATC TTC AAG CAA

3A

ATC TCA ATT CCT TCC CAA AAG TCC GTG TTG TAC TTT CTC ATT GAG AAG GGG CAA CAC GAA GCA GCA ATT GAA TTC TTT GAG GGG ATG GTC AGT GAC TCC GTC AAG GAG GAG CTC CGA CCC CTC ATC CAA CAG ACC TCA TTT GTG AAA CGT GCG TTC AAA CGC TTG AAG GAA AAC TTT GAG ATT GTT GCC CTA TGT TTG ACC CTT TTG GCA AAC ATA GTG ATC ATG ATC CGC GAG ACT CGT AAG AGG CAA CAA ATG GTG GAT GAT GCA GTG AAT GAG TAC ATT GAG AAA GCA AAC ATC ACC ACA GAT GAC AAG ACT CTT GAT GAG GCG GAA AAG AAC CCT CAG GAG ATT AGC AGT GCC AGC ACT GTT GGC TTC AGA GAG AGG ACT CTT CCA GGA CAG AAG GTG GGT GAT GAC GTG AAA TCC GAG CCC ACC GAA CCT GCG AGA GAG CAA CCA CAA GCT GAA

3B1

GGA CCC TAC GCC GGG CCA CTC GAG CGT CAG AAA CCT CTG AAG GTG AGA GCC AAG CTA CCA CAA CAG GAG

3B2

GGA CCC TAC GCT GGC CCG ATG GAG AGA CAG AAA CCA CTA AAA GTG AAA GCA AAA GCC CCG GTC GTG AAG GAA

3B3

GGA CCT TAC GAG GGA CCG GTG AAG AAG CCT GTC GCT TTG AAA GTG AAA GCT AAG AAC TTG ATT GTC ACT GAG

3C

AGT GGA GCC CCA CCG ACT GAC CTG CAA AAG ATG GTC ATG GGC AAC ACG AAG CCT GTT GAG CTC ATC CTC GAC GGG AAG ACA GTA GCT ATC TGC TGT GCT ACT GGA GTG TTC GGC ACT GCC TAC CTC GTG CCT CGT CAT CTT TTC GCA GAG AAG TAT GAC AAG ATC ATG TTG GAC GGT AGA GCC ATG ACA GAC AGT GAC TAC AGA GTG TTT GAG TTT GAG ATA AAA GTA AAA GGA CAA GAC ATG CTC TCA GAC GCC GCG CTC ATG GTG CTT CAC CGT GGG AAT CGC GTG CGG GAC ATC ACG AAG CAC TTC CGT GAT GTG GCA AAA ATG AAG AAA GGC ACC CCC GTC GTT GGC GTG ATT AAC AAC GCC GAT GTC GGG AGA CTG ATT TTC TCT GGT GAG GCC CTT ACC TAC AAG GAC ATT GTA GTG TGC ATG GAC GGA GAC ACC ATG CCT GGC CTC TTT GCC TAC AAA GCC GCC ACC AAG GCT GGC TAC TGT GGA GGA GCC GTT CTT GCC AAG GAC GGT GCC GAG ACA TTC ATC GTT GGC ACT CAC TCC GCA GGT GGC AAT GGA GTT GGA TAC TGC TCC TGT GTT TCC AGG TCC ATG CTC ATG AAA ATG AAG GCA CAC ATC GAC CCT GAA CCA CAC CAC GAG

3D

GGT TTG ATT GTT GAC ACC AGA GAT GTG GAA GAG CGC GTG CAC GTC ATG CGC AAA ACC AAG CTT GCA CCC ACC GTC GCG CAC GGT GTG TTC AAC CCT GAA TTC GGG CCT GCC GCC TTG TCC AGC AAG GAT CCG CGC CTG AAC GAC GGG GTC GTC CTC GAC GAA GTC ATC TTC TCC AAA CAC AAG GGA GAC ACT AAG ATG TCT GAG GAG GAC AAA GCG CTG TTC CGC CGC TGT GCT GCT GAC TAC GCG TCA CGC CTG CAT AGT GTG TTG GGT ACA GCA AAT GCC CCA CTG AGC ATT TAC GAG GCA ATC AAA GGC GTT GAC GGG CTC GAC GCC ATG GAG CCA GAC ACT GCA CCT GGC CTT CCC TGG GCC CTC CAG GGA AAG CGC CGT GGC GCA CTC ATC GAC TTC GAG AAC GGC ACG GTC GGA CCC GAG GTT GAA GCT GCC TTG AAG CTC ATG GAG AAA AGG GAA TAC AAA TTT GCT TGT CAG ACC TTC CTG AAG GAC GAA ATT CGC CCA ATG GAG AAA GTG CGT GCC GGC AAG ACT CGC ATT GTC GAC GTC CTG CCC GTT GAA CAC ATT CTT TAC ACC AGA ATG ATG ATT GGC AGA TTT TGT GCT CAA ATG CAC TCG AAC AAC GGA CCG CAA ATT GGC TCT GCG GTC GGC TGT AAT CCT GAT GTT GAT TGG CAA AGA TTC GGA ACC CAT TTT GCT CAG TAC AGA AAT GTG TGG GAT GTG GAC TAT TCG GCC TTT GAT GCT AAC CAC TGC AGT GAC GCG ATG AAC ATC ATG TTT GAG GAA GTG TTT CGC ACG GAG TTC GGT TTC CAC CCA AAC GCT GAG TGG ATT CTG AAG ACT CTA GTG AAC ACG GAG CAC GCC TAT GAG AAC AAA CGC GTC ACT GTT GAG GGC GGG ATG CCG TCT GGC TGT TCC GCG ACA AGC ATT ATC AAC ACA ATT TTG AAC AAC ATT TAC GTG CTC TAC GCG CTG CGT AGA CAC TAT GAG GGA GTT GAG CTG GAC ACT TAC ACC ATG ATC TCC TAC GGA GAC GAC ATC GTG GTT GCA AGT GAT CAC GAT TTG GAC TTT GAG GCC CTC AAG CCT CAC TTT AAA TCT CTT GGT CAA ACC ATC ACT CCA GCT GAC AAA AGC GAC AAA GGT TTT GTT CTT GGT CAC TCC ATC ACT GAT GTC ACT TTC CTC AAA AGA CAC TTC CAC ATG GAT TAT GGA ACT GGG TTT TAC AAA CCT GTG ATG GCC TCA AAG ACC CTT GAG GCT ATC CTC TCC TTT GCA CGC CGT GGG ACC ATA CAG GAG AAG TTG ATC TCC GTG GCA GGA CTC GCT GTC CAC TCT GGA CCT GAC GAG TAC CGG CGT CTC TTT GAG CCC TTC CAG GGC CTC TTT GAG ATT CCA AGC TAC AGA TCA CTT TAC CTG CGT TGG GTG AAC GCC GTG TGC GGT GAC GCA

3'UTR

TAATCCCTCAGATGTCACAATTGGCAGAAAGACTCTGAGGCGAGCGACGCCGTAGGAGTGAAAGGTCGAAAGGCCTTTCCCGCTTCCTAA

CTCCAAAAAAAAAAAAAA

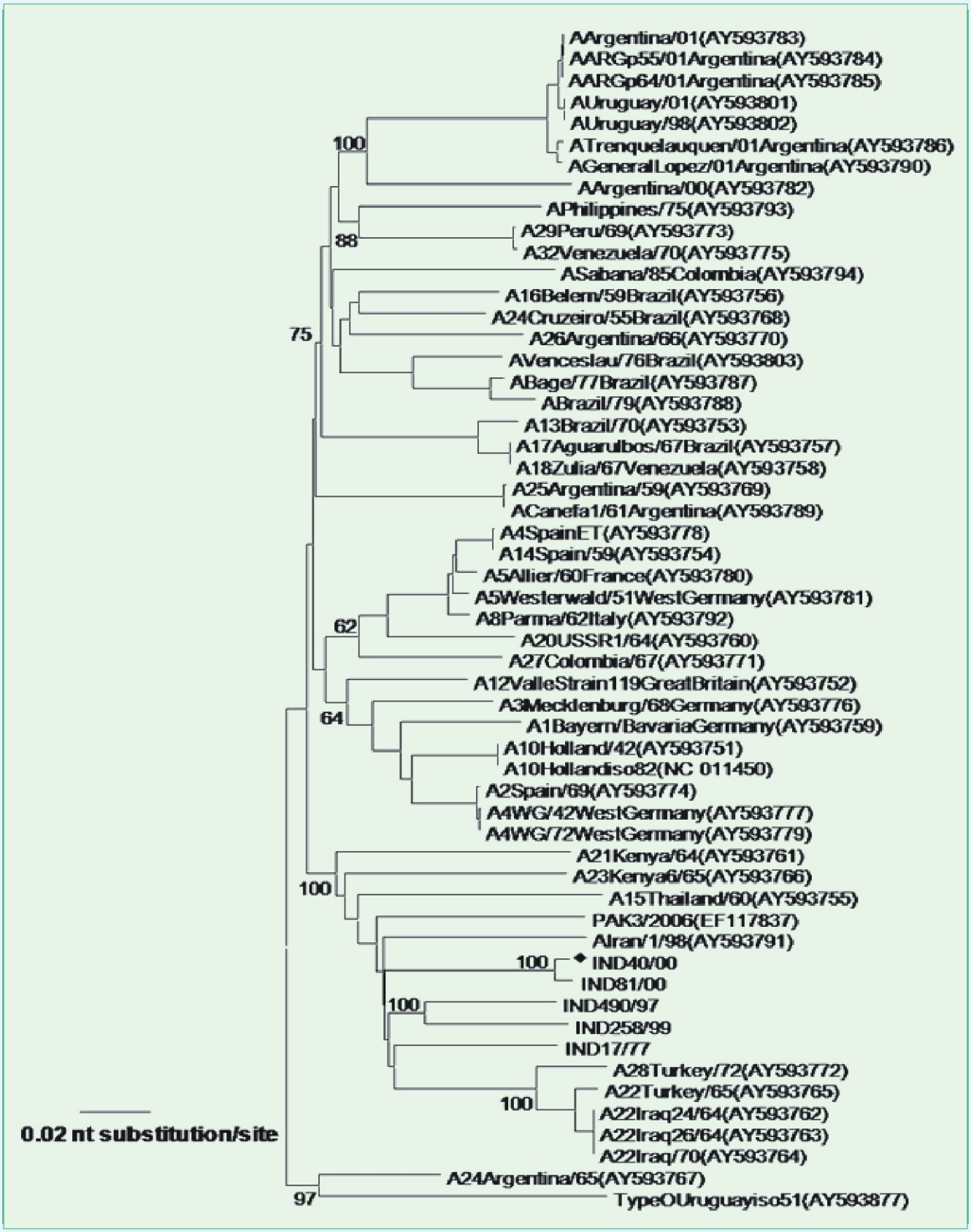


Fig 8. Phylogenetic relationship of current type A vaccine strain virus (IND 40/00) at complete coding region

Antigenic Characterization

A total number of 6 field isolates covering five different outbreaks were subjected to 2D-MNT using bovine vaccinate and convalescent serum against the current vaccine strain, IND 40/00 and one way antigenic relationship was investigated. The 'r-value' figures which directly correlate with antigenic relationship of isolates with the vaccine strain varied from 0.47 to 1.0 for individual strains (Fig. 9). As an 'r-value' range of 0.3 to 1.0 indicates close antigenic relatedness, it can be deduced that all the recent outbreak strains show a close antigenic match with the vaccine strain. Hence the current vaccine strain, IND 40/00 offers optimum antigenic coverage to all the circulating viruses of recent origin. Even though some of these outbreak strains belong to the VP3[59]-deletion group, which is known to be antigenically quite heterogeneous in nature, they are expected to be antigenically covered by the vaccine strain in the field conditions.

6.2.3 Type Asia1 FMD Virus

Molecular Epidemiology

Molecular phylogeny based on VP1 encoding region has established circulation of three prominent lineages (lineage B, C and D) in India. The Indian Asia1 field isolates form a single genotype (Gurumurthy et al., 2002) with two different genetic lineages (Mohapatra et al., 2002). The lineage B which includes the vaccine strain IND 63/72 has 210 amino acids in VP1 and this lineage never appeared after the year 2000. The lineage C which was prominently circulating during the period 1993 to 2001 has an extra amino acid at position 44 of VP1(Gurumurthy et al., 2002). A novel divergent genetic lineage (lineage D) with-in lineage C appeared in 2001 and it outnumbered the parent lineage in terms of field outbreaks (Sanyal et al., 2004). The isolates of lineage D was 8-13% divergent at nucleotide level from the isolates of lineage C (Sanyal et al., 2004; Mohapatra et al., 2004). Lineage C has been responsible for all Asia1 outbreaks in the country since 2005. During the period outbreak due to Asia1 serotype was recorded in Gujarat, Madhya Pradesh, Maharashtra, Uttar Pradesh and West Bengal. The FMDV Asia1 isolates from Gujarat and UttarPradesh could be seuqnced for molecular epidemiological studies. The Asia1 field isolates were grouped with lineage C (Fig. 10) reiterating the supremacy of this lineage in the field since 2005.

Antigenic Characterization

FMDV Asia1 isolates were subjected to

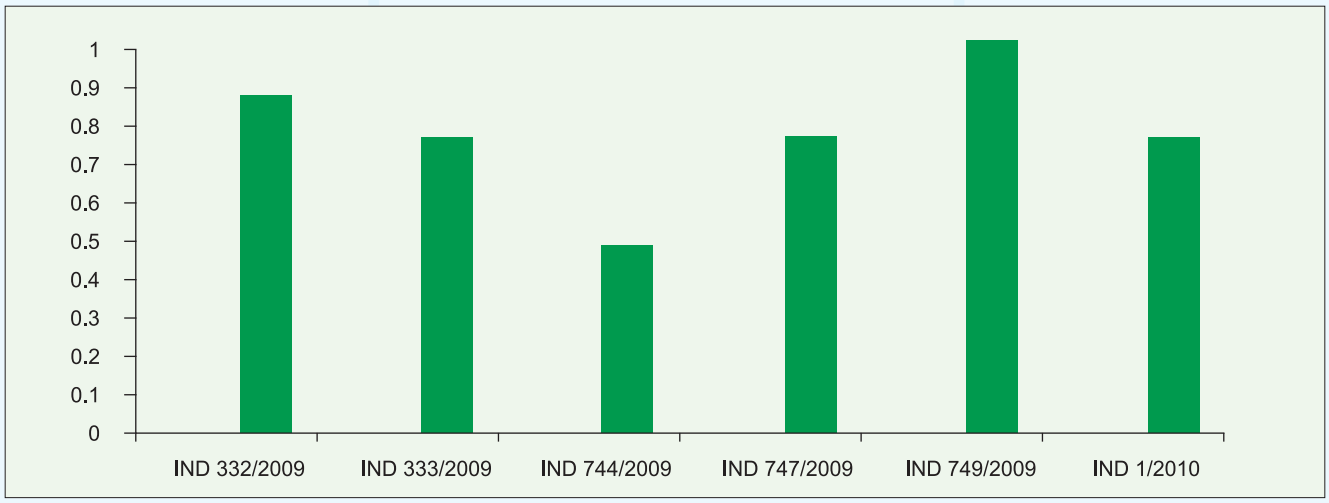


Fig 9. Antigenic relationship of type A isolates with the vaccine strain IND 40/00

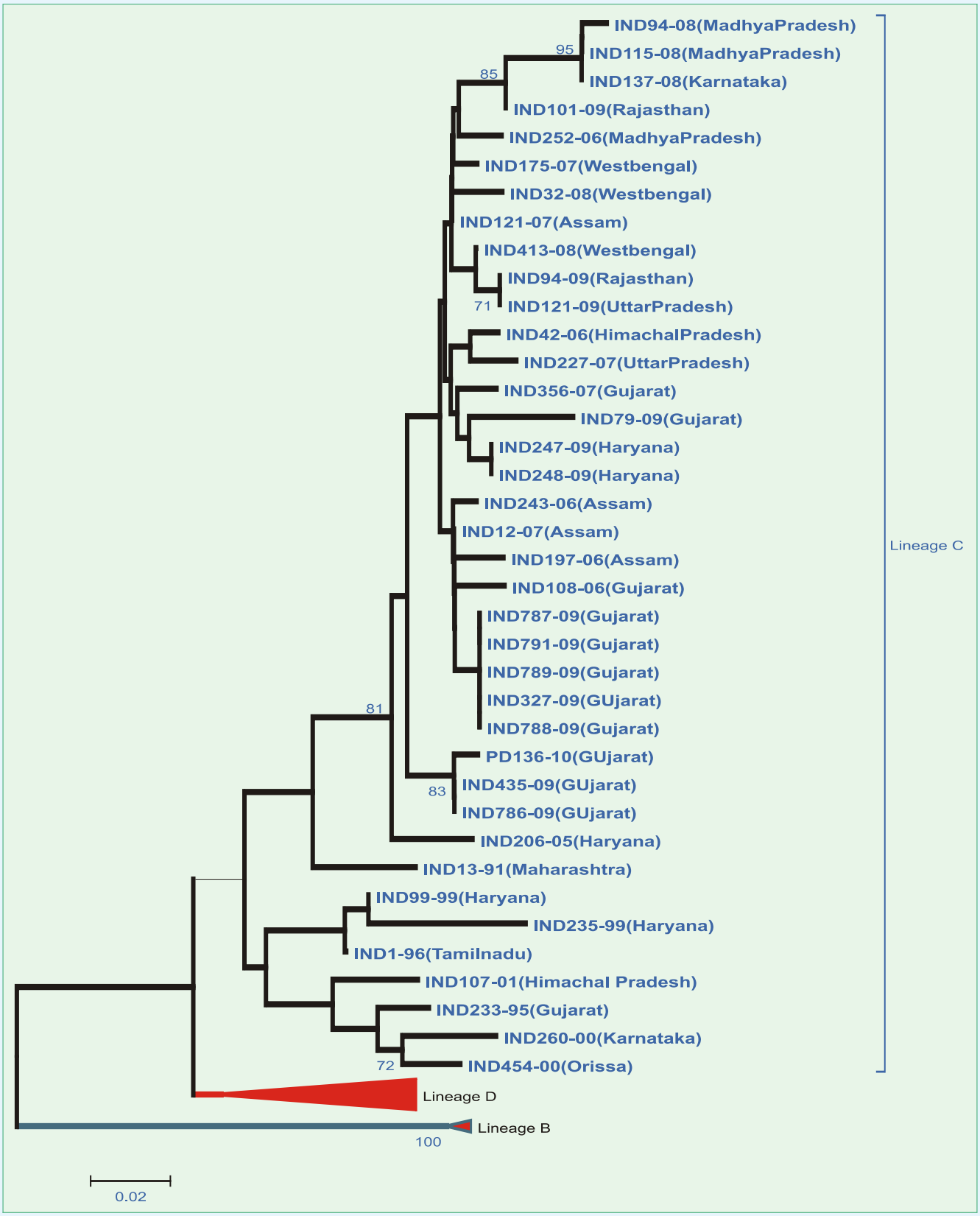


Fig 10. Neighbour-joining tree depicting phylogenetic relationship among type Asia1 FMD virus isolates at 1D region

antigenic analysis using anti-146S bovine serum against the vaccine strain (IND 63/72). All of them showed an r-value of more than 0.3 with in-use vaccine strain indicating its appropriate antigenic coverage (Fig 11).

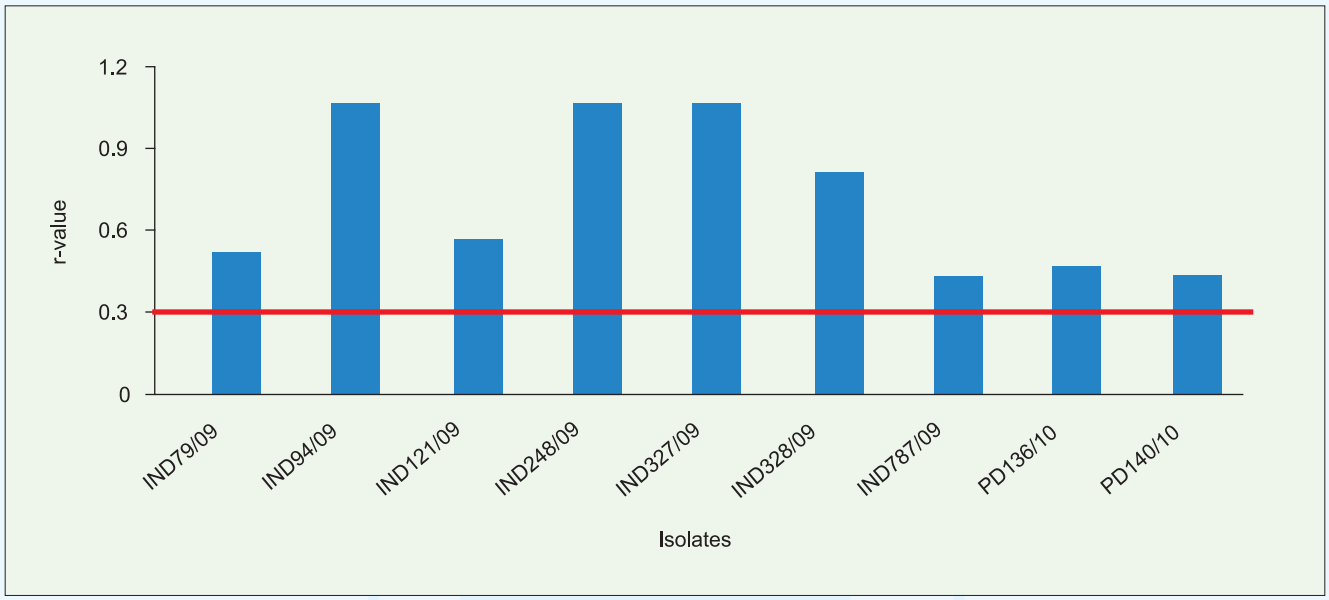


Fig 11. Antigenic relationship (r-value) of type Asia1 isolates recovered during 2009-10 with in-use vaccine strain (IND 63/72)

7.0 National FMD Virus Repository

The Central FMD laboratory of the Project Directorate maintains the National FMD Virus Repository that is upgraded annually with latest/ new virus isolates. The virus repository has served the cause of the project by providing isolates for molecular epidemiological studies, evaluation of antigenic relatedness between the field and vaccine strains and selection of new candidate vaccine strains whenever required. This year, a total of 133 virus isolates including 97 type O, 12 type Asia1 and 24 type A field isolates were added to the repository. At present the National repository contains a total of 1687 (1096-O, 259-A, 317-Asia1, 15-C) well characterized field isolates (Fig 12, 13, 14 and 15).

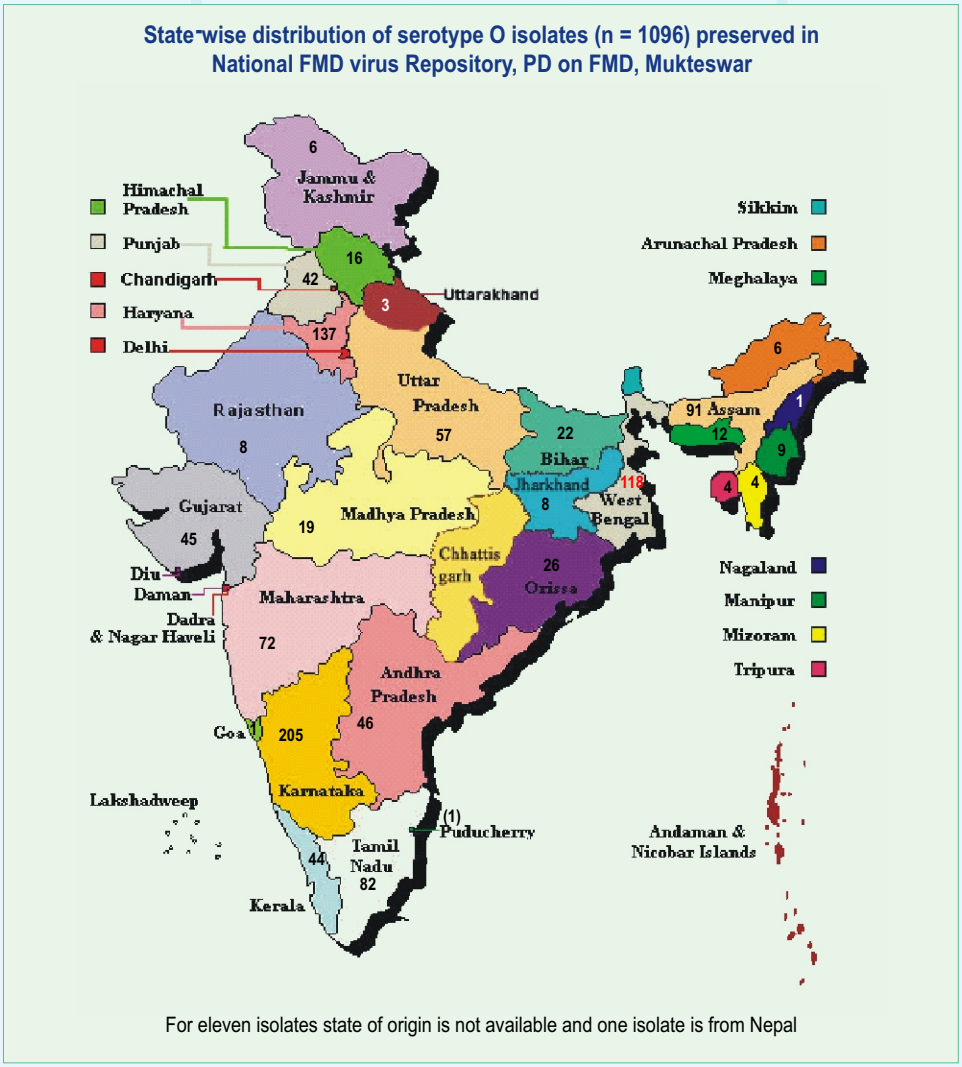


Fig 12. State-wise distribution of serotype O isolates (n = 1096) preserved in National FMD virus Repository, PD on FMD, Mukteswar

Fig 13. State-wise distribution of serotype A isolates (n = 259) preserved in National FMD virus Repository, PD on FMD, Mukteswar

Fig 14. State-wise distribution of serotype Asia 1 isolates (n = 317) preserved in National FMD virus Repository, PD on FMD, Mukteswar

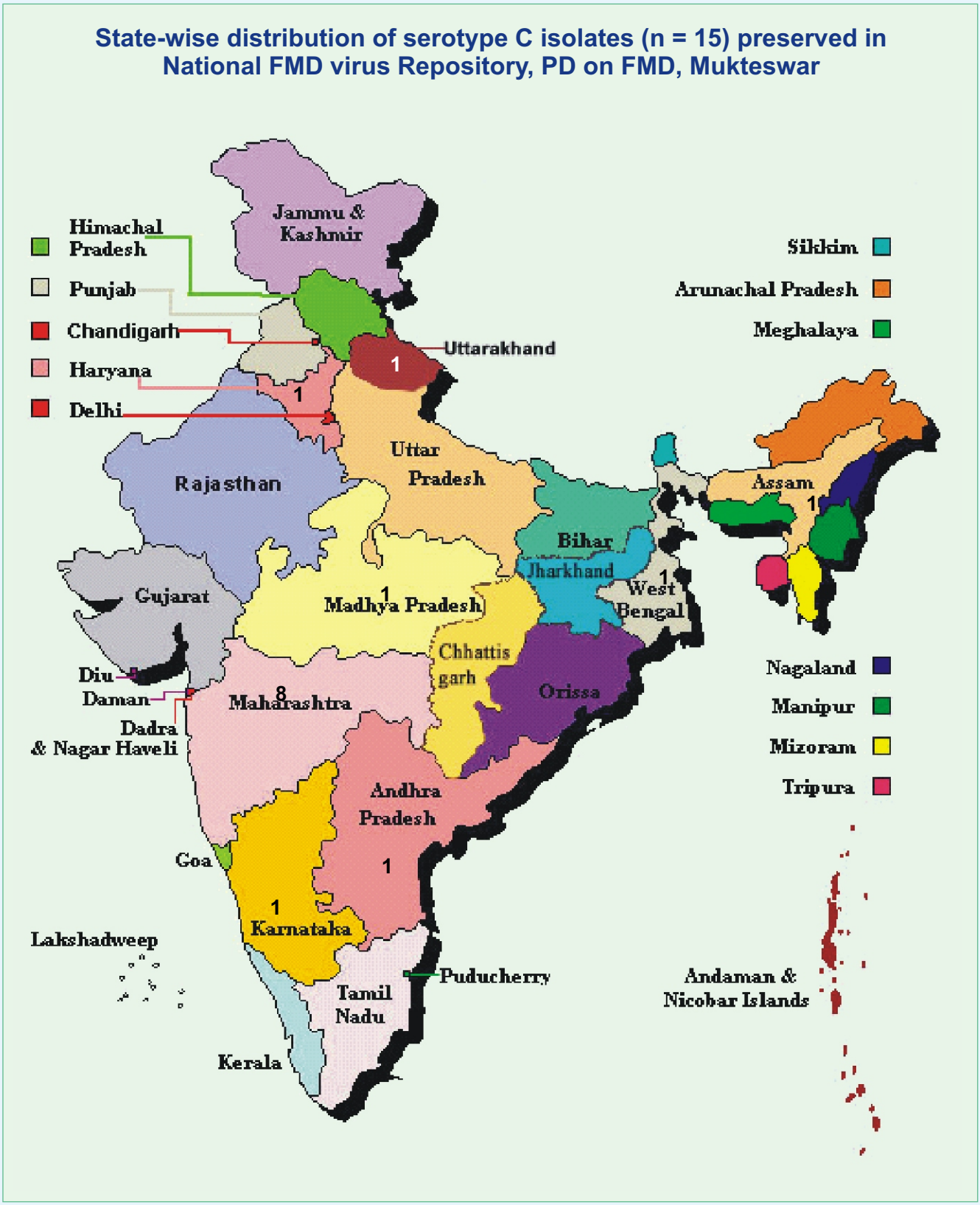


Fig 15. State-wise distribution of serotype C isolates (n = 15) preserved in National FMD virus Repository, PD on FMD, Mukteswar

8.0 New Research Initiative

8.1 Detection of FMDV in semen samples (SOP developed)

Due to the importance of shedding of FMD virus in semen of convalenscent bulls and its detection by a suitable test to certify negativity/ positivity, SOP was devised and validated for detection and identification of FMD virus in bull semen by mPCR. It is to mention that semen being a body fluid; it has several enzyme/PCR inhibitors. During the period 22 semen samples were received from IVRI Izatnagar following FMD in bulls for FMD virus testing. Ten semen samples were also collected from Project Directorate on Cattle, Meerut following FMD in the animals. Semen sample was collected in RNAase and DNAase free vials and transported on ice to the Central Laboratory. Samples were stored at 4ºC till further use. Total RNA was extracted with below mwntioned method to increase the total quantity of RNA and reducing non specific inhibitors of PCR. Neat semen was diluted and mixed thoroughly in RLT buffer (lysis buffer provided in RNAeasy mini Kit, Qiagen) in equal quantity and passed through QIAashredder columns. Flow through was used for RNA extraction using QIAamp viral RNA extraction kit. Extracted RNA was quantified in Nanodrop spectrophotometer and cDNA was synthesized using FMDV specific NK61 reverse primer and *MMLV* Reverse Transcriptase (Promega). Multiplex PCR was performed as per the standard protocol. PCR controls included a known negative semen sample spiked with different log dilutions of cell culture FMD virus and RNA extraction was performed simultaneously. Multiple negative controls were also included to check specificity and contaminations. Test was considered valid only if minimum 100 $TCID_{50}$ spiked sample was positive and known negative sample was negative in PCR (Fig 16). Out of 22 semen samples

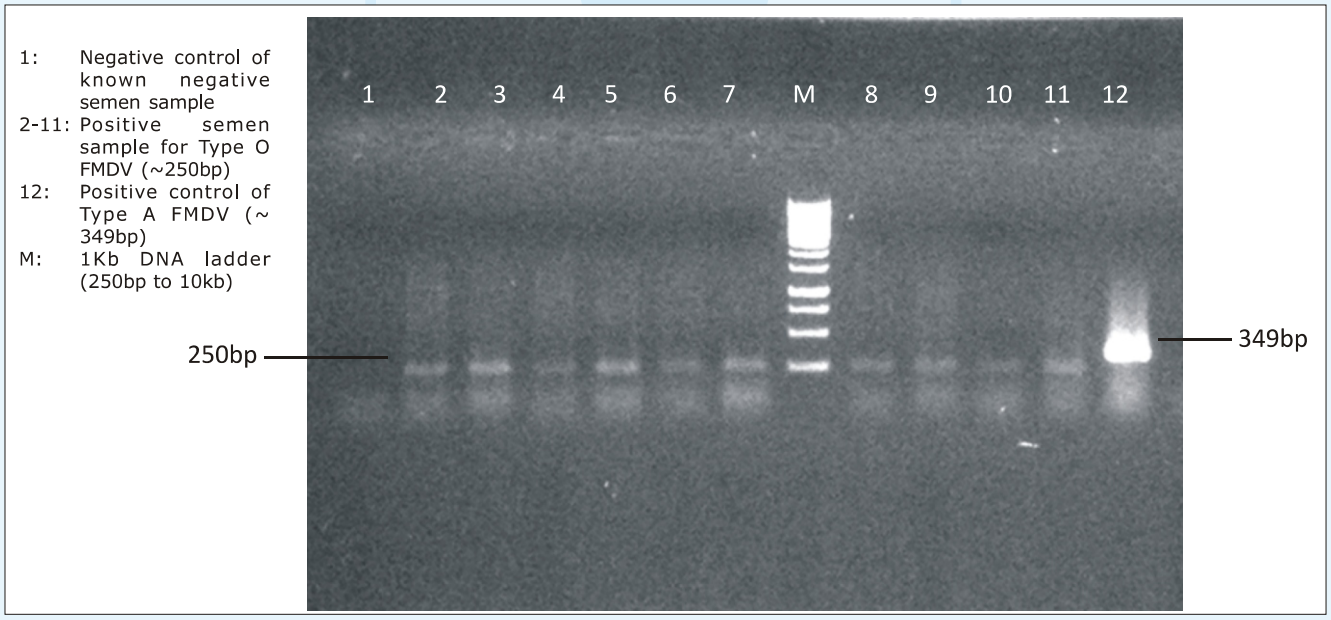


Fig 16: Multiplex PCR result of semen samples collected from cattle

received from IVRI, 7 were positive for Type O FMDV while remaining 15 samples were found negative. Out of 10 semen samples from PDC, Meerut, 8 samples were positive for Type O FMDV and remaining 2 were negative. It is to mention that there was FMD in the animals in recent past.

8.2 Development of LAMP assay for diagnosis of FMD virus

Loop mediated isothermal amplification (LAMP) amplify DNA under isothermal conditions with high specificity, efficiency and rapidity using a DNA polymerase and a set of four specially designed primers that recognize a total of six distinct sequences on the target DNA. The final products are a mixture of stem-loop DNAs with various stem lengths and cauliflower-like structures with multiple loops, which gives ladder like pattern on the agarose gel. The one-step, reverse transcription loop-mediated amplification (RT-LAMP) assay enables FMD virus (FMDV) to be detected in under an hour in a single tube without thermal cycling. A fragment of the 3D RNA polymerase gene of the virus is amplified at 65æ%C in the presence of a primer mixture and both reverse transcriptase and *Bst* DNA polymerase. The development of one-step RT-LAMP for FMDV is under process in our lab. The below mentioned figure 17 indicate optimization of LAMP assay for betaine concentration. It indicate 1M betaine concentration is optimum for the assay.

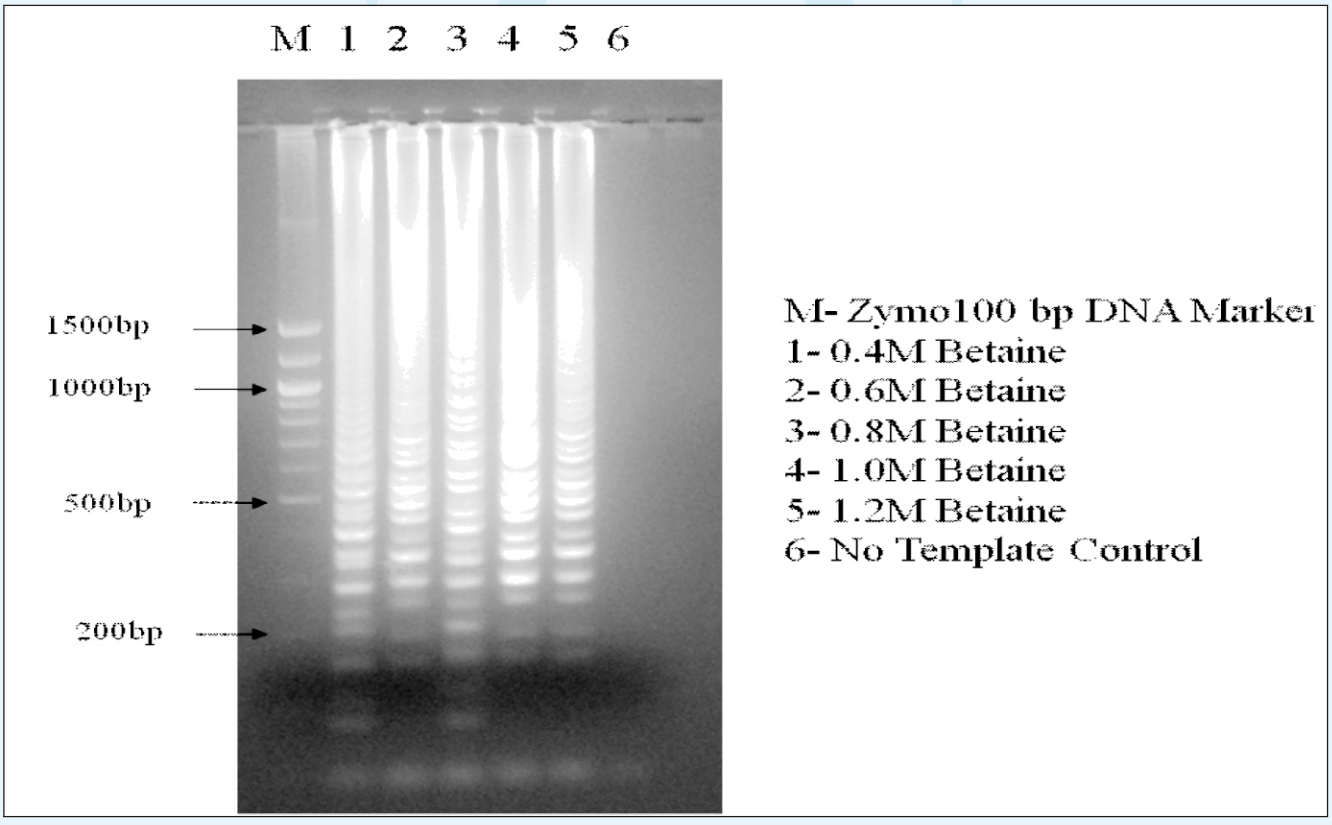


Fig 17: Optimization of Betain concentration in LAMP assay

9.0 Estimation of FMD prevalence under National FMD Serosurveillence

A total number of 29,763 random bovine serum samples collected at the rate of 100 per district from 335 districts covering 21 different states of the country were tested in r3AB3 DIVA ELISA in an exercise to estimate FMD prevalence in the country from time to time. This revealed 27.9 % of the bovine population in the country to be FMD virus infected (3AB3 reactors) upon testing of serum samples collected during 2008-2010 at a confidence level of 95%. State-wise FMD prevalence in bovines varied from 5.1% in Himachal Pradesh to 56% in Rajasthan (Table 6 and 7; Fig. 18 and19). Level of virus circulation in the states differed perhaps either due to geographical factors acting as natural impediments for migration of animals/virus or due to positive effect of regular vaccination. A prevalence of less than 20% in states like Mizoram, Tripura, H.P., Manipur and Punjab stands in testament to the above hypothesis and commensurates the outbreak scenario in the mentioned states. Interestingly, in many states like Kerala and Punjab, a clear and direct correlation could be made in the prevalence figure and the natural outbreak history for the districts. For instance, in case of Kerala, the prevalence figure for the southern districts was the highest, which have experienced very recent outbreaks

Table 6. Summary of FMD prevalence in various states as estimated in DIVA ELISA during 2008-2009 December

S.No	States	No. of districts	Total No. of samples tested	Total no. positive	% FMD infected
1.	Tripura	04	391	44	11.2
2.	Gujarat	21	1995	761	38.2
3.	Mizoram	08	799	56	7
4.	Himachal Pradesh	11	957	49	5.1
5.	Nagaland	08	582	134	23
6.	Bihar	38	3820	1020	26.7
7.	Madhya Pradesh	46	3992	1184	29.7
8.	West Bengal	08	657	203	30.8
9.	Manipur	09	1001	160	16
10.	Maharashtra	33	2776	826	29.8
11.	Punjab	20	2055	256	12.4
12.	Kerala	14	1409	214	15.1
13.	AndhraPradesh	17	1700	363	21.3
14.	Arunachal Pradesh	13	718	215	30
15.	Orissa	30	2780	1099	39.5
16.	Haryana	7	676	178	26.4
17.	Jammu & Kashmir	03	74	23	31
18.	Rajasthan	11	462	260	56
19.	Karnataka	25	2109	1021	48.4
20.	Tamil Nadu	04	360	88	24.4
21.	Assam	05	450	149	33.2
	Grand total for India	**335**	**29763**	**8303**	**27.9**

and continuing to do so. In case of Punjab, the disease prevalence was estimated to be in a graded fashion decreasing gradually from Pakistan and Rajasthan border to Himachal Pradesh border. In case of Haryana, after testing of only 7 districts serum samples from FMD CP area, a surprisingly high prevalence figure was obtained for districts bordering Rajasthan like Bhiwani. Though it does not reflect disease outbreak picture in those districts, we guess, as these districts share borders and animal migration routes with Rajasthan there could be concurrent viral activity in the population without conspicuous symptoms owing to high vaccine coverage. A higher proportion of positive samples (~30%) were found in a higher T/N ratio range (>8.0) in case of states like karnataka, Bihar and Madhya Pradesh, which have been continuously experiencing outbreaks, suggesting ongoing circulation of virus in the population (Fig. 20). In contrast, a lower proportion of positive samples (~10%) were found in the same T/N ratio range for states like Haryana, Himachal Pradesh and Punjab, where a few sporadic cases have been reported since last 3 years suggesting either gradual waning of anti-3AB3 response from past infections and minimal current virus circulation or presence of subclinical carriers. The % infected bovines in Punjab is almost half of that of the country's average indicating the effectiveness of regular FMD vaccination in disease containment and suitability of this DIVA kit in assessing the impact of regular vaccination and detecting evidence of infection in multiply vaccinated animals.

Table 7. District-wise FMD prevalence as estimated in DIVA ELISA during 2008-2009 December

States	Districts	Total No. of samples tested	Total No. positive	% FMD infected
Tripura	1. North Tripura Dist.	101	23	23
	2. South Tripura Dist.	99	9	9
	3. Dhalai Tripura Dist.	99	7	7
	4. West Tripura Dist.	92	5	5
	Total for Tripura	**391**	**44**	**11.2**
Gujarat	1. Rajkot	100	38.5	38.5
	2. Dang	100	15	15
	3. Junagadh	107	45.5	42.5
	4. Valsad	100	21	21
	5. Surat	105	61	58
	6. Navasari	100	32	32
	7. Amreli	40	6	15
	8. Anand	100	38	38
	9. Banaskantha	100	44	44
	10. Bhavnagar	60	11	18.3
	11. Dahod	100	31.5	31.5
	12. Gandhinagar	100	7	7
	13. Jamnagar	100	53.5	53.5
	14. Kutch	103	53.5	52

States	Districts	Total No. of samples tested	Total No. positive	% FMD infected
Gujarat	15. Kheda	100	19	19
	16. Narmada	100	42	42
	17. Panchmahal	100	53	53
	18. Porbander	90	50	55.5
	19. Sabarkantha	90	24	26.6
	20. Surendranagar	100	60.5	60.5
	21. Vadodara	100	55	55
	Grand Total for Gujarat	**1995**	**761**	**38.2**
Mizoram	1. Aizawl	100	16	16
	2. Kolasib	100	27	27
	3. Serchhip	100	11	11
	4. Champhai	100	1	1
	5. Mamit	100	1	1
	6. Lunglei	100	0	0
	7. Saiha	100	4	4
	8. Lawngtlai	99	10	10
	Grand Total for Mizoram	**799**	**56**	**7**
Himachal Pradesh	1. Shimla	33	13	39
	2. Solan	100	7	7
	3. Sirmour	100	4	4
	4. Kinnaur	92	4	4
	5. Mandi.	100	4	4
	6. Kangla	100	6	6
	7. Chamba	57	0	0
	8. Bilaspur	100	0	0
	9. Hamirpur	100	3	3
	10. Una	100	0	0
	11. Lahaul spiti	76	8	11
	Grand total for Himachal Pradesh	**957**	**49**	**5.1**
Nagaland	1. Kohima	78	36	46.2
	2. Tuensang	84	29	34.5
	3. Phek	37	5	14
	4. Longleng	153	42	27.5
	5. Kipheri	85	9	10.6
	6. Zunheboto	77	12	15.6
	7. Peren	50	0.5	1
	8. Wokha	18	0.5	2.8
	Grand total for Nagaland	**582**	**134**	**23**

States	Districts	Total No. of samples tested	Total No. positive	% FMD infected
Bihar	1. Patna	120	25.0	20.83
	2. Nalanda	100	37.0	37.0
	3. Gaya	100	14.5	14.5
	4. Bhojpur	100	18.5	18.5
	5. Muzaffarpur	100	30.0	30.0
	6. Banka	100	20.0	20.0
	7. East Champaran	100	34.0	34.0
	8. Gopalganj	100	12.5	12.5
	9. Buxar	100	28.0	28.0
	10. Madhepura	100	56.5	56.5
	11. Purnia	100	34.5	34.5
	12. Katihar	100	28.5	28.5
	13. West Champaran	100	13.5	13.5
	14. Jumui	100	19.5	19.5
	15. Samastipur	100	16.0	16.0
	16. Saran (Chapra)	100	16.0	16.0
	17. Araria	100	35.0	35.0
	18. Darbhanga	100	31.5	31.5
	19. Munger	100	43.5	43.5
	20. Saharsa	100	39.5	39.5
	21. Aurangabed	100	36.0	36.0
	22. Bhagalpur	100	15.0	15.0
	23. Rohtas	100	29.5	29.5
	24. Nawada	100	26.0	26.0
	25. Lackhisarai	100	40.5	40.5
	26. Sheikhpura	100	28.0	28.0
	27. Kaimur	100	34.5	34.5
	28. Jehanabad	100	20.0	20.0
	29. Arwal	100	39.0	39.0
	30. Begusarai	100	27.0	27.0
	31. Supaul	100	21.5	21.5
	32. Khagaria	100	16.5	16.5
	33. Kisanganj	100	42.0	42.0
	34. Siwan	100	20.0	20.0
	35. Sitamarhi	100	21.5	21.5
	36. Sheohar	100	25.0	25.0
	37. Vaishali	100	17.5	17.5
	38.Madhubani	100	24.5	24.5
	Grand total for Bihar	**3820**	**1020.5**	**26.7**

States	Districts	Total No. of samples tested	Total No. positive	% FMD infected
Madhya Pradesh	1. Betul	100	23	23
	2. Bhopal	100	24	24
	3. Burhanpur	100	19	19
	4. Chhaterpur	58	16	28
	5. Chhindwara	90	50.5	56
	6. Gwalior	100	58	58
	7. Indore	100	37	37
	8. Jabalpur	100	24	24
	9. Jhabua	100	39	39
	10. Khandwa	100	37	37
	11. Shajapur	100	48	48
	12. Khargone	95	61	65
	13. Mandla	100	26	26
	14. Mandsor	100	22	22
	15. Morena	70	21.5	30.7
	16. Raisen	82	31	38
	17. Rewa	99	11	11
	18. Sagar	100	57	57
	19. Shahdol	100	10	10
	20. Sehore	100	9	9
	21. Seoni	100	37	37
	22. Shivpuri	100	33	33
	23. Sidhi	100	01	1
	24. Tikamgarh	100	25	25
	25. Ujjain	100	35	35
	26. Umaria	55	21	38
	27. Vidisha	100	11	11
	28. Badwani	100	57	57
	29. Damoh	100	27	27
	30. Satna	100	30	30
	31. Neemach	80	26	33
	32. Katni	100	42	42
	33. Harda	70	14	20
	34. Balaghat	100	35	35
	35. Dewas	100	28	28
	36. Panna	60	22	36.6
	37. Guna	92	19	19
	38. Rajgarh	100	21	21

States	Districts	Total No. of samples tested	Total No. positive	% FMD infected
Madhya Pradesh	39. Ratlam	63	9	14
	40. Ashoknagar	90	17	18.8
	41. Dindori	58	15	26
	42. Sheopur	70	11	15.7
	43. Bhind	50	9	18
	44. Dhar	30	5	16.6
	45. Hoshangabad	50	7	14
	46. Datia	30	3	10
	Grand total for Madhya Pradesh	**3992**	**1184**	**29.7**
West Bengal	1. Burdwan	100	26	26
	2. Nadia	71	15	21
	3. South 24 Parganas	50	6	12
	4. Hoogly	100	33	33
	5. Murshidabad	100	24	24
	6. Bankura	100	55	55
	7. Birbhum	74	21	28
	8. North 24 Parganas	62	5	8
	Grand total for West Bengal	**657**	**184**	**28**
Manipur	1. Imphal-West	201	51.5	25.6
	2. Imphal-East	100	20	20
	3. Bishnupur	100	12.5	12.5
	4. Thoubal	100	3.5	3.5
	5. Chandel	100	23	23
	6. Churachandpur	100	5.5	5.5
	7. Senapati	100	11	11
	8. Ukhrul	100	24.5	24.5
	9. Tamenglong	100	8.5	8.5
	Grand total for Manipur	**1001**	**160**	**16**
Maharashtra	1. Mumbai	60	34	56.6
	2. Raigad	97	16	16.4
	3. Sindhudurg	100	9	9
	4. Thane	80	49	61.2
	5. Ratanagiri	93	26	27.9
	6. Pune	104	11	10.5
	7. Kolhapur	74	10	13.5
	8. Solapur	100	30	30
	9. Satara	80	15	18.7
	10. Nashik	90	26	28.8
	11. Dhule	100	63	63
	12. Jalgaon	100	20	20

States	Districts	Total No. of samples tested	Total No. positive	% FMD infected
Maharashtra	13. Nandurabar	100	25	25
	14. Ahamadnagar	101	4	3.9
	15. Nagpur	80	38	47.5
	16. Gondia	115	55	47.8
	17. Gadchiroli	90	12	13.3
	18. Wardha	61	35	57.3
	19. Chandrapur	80	8	10
	20. Bhandara	84	19	22.6
	21. Aurangabad	80	5	6.2
	22. Jalana	90	58	64.4
	23. Parbhani	94	32	34
	24. Beed	100	39	39
	25. Hingoli	58	36	62
	26. Nanded	100	42	42
	27. Latur	72	14	19.4
	28. Osmanabad	85	14	16.4
	29. Buldhana	83	27	32.5
	30. Akola	100	51	51
	31. Amravati	30	0	0
	32. Washim	20	0	0
	33. Yavatamal	75	3	4
	Grand total for Maharashtra	**2776**	**826**	**29.8**
Punjab	1. Amritsar	144	10.5	7.2
	2. Bathinda	111	7	6.3
	3. Barnala	138	10.5	7.6
	4. Fatehgarh Sahib	130	13.5	10.3
	5. Faridkot	219	28.5	13
	6. Ferozpur	152	28.5	18.7
	7. Gurdaspur	100	10.5	10.5
	8. Hoshiarpur	154	9.5	6.1
	9. Jalandhar	83	14.5	17.4
	10. Kapurthala	110	23.5	21.3
	11. Ludhiana	08	0	0
	12. Mansa	99	30	30.3
	13. Mohali	20	2	10
	14. Moga	109	16.5	15.1
	15. Muktsar	25	6	24
	16. Nawashahar	49	1	2
	17. Patiala	160	18	11.2

States	Districts	Total No. of samples tested	Total No. positive	% FMD infected
Punjab	18. Ropar	102	5.5	5.3
	19. Sangrur	79	10.5	13.2
	20. Tarantaran	63	10	15.8
	Grand total for Punjab	**205**	**256**	**12.4**
Kerala	1.Alappuzha	100	43.5	43.5
	2.Ernakulam	100	10	10
	3.Kannur	100	29.5	29.5
	4.Kasargode	100	2	2
	5.Kollam	100	17.5	17.5
	6.Kozhikode	100	23	23
	7.Malappuram	104	6	5.7
	8.Thiruvananthapuram	100	23	23
	9.Thrissur	100	17	17
	10. Pathanamthitta	100	20	20
	11. Kottayam	100	4	4
	12. Idukki	101	1	0.9
	13. Palakkad	103	11.5	11.1
	14. Wayanad	101	6	5.9
	Grand total for Kerala	**1409**	**214**	**15.1**
Andhra Pradesh	1.Srikakulam	100	19	19
	2.Vizianagar	100	00	00
	3Visakapatnam	100	11	11
	4.West Godavari	100	35	35
	5.East Godavari	100	16	16
	6.Krishna	100	7	7
	7.Guntur	100	25	25
	8.Prakasam	100	17	17
	9.Nellore	100	22	22
	10.Adilabad	100	21	21
	11.Nizamabad	100	34	34
	12.Karimnagar	100	57	57
	13.Mahabubnagar	100	19	19
	14.Nalgonda	100	17	17
	15.Khammam	100	17	17
	16.Kadapa	100	18	18
	17.Kurnool	100	28	28
	Grand total for AndhraPradesh	**1700**	**363**	**21.3**

States	Districts	Total No. of samples tested	Total No. positive	% FMD infected
Arunachal Pradesh	1. L.Subansiri	30	18	60
	2. U.Siang	60	26	43
	3. Lohit	52	14	27
	4. Tirap	45	6	13
	5. P.Pare	17	12	71
	6. U.Subansiri	64	13	20.3
	7. Changlang	92	34	37
	8. Anjaw	64	8	12
	9. W.Kameng	61	6	9
	10. Twang	8	0	00
	11. L.D.Valley	75	13	17
	12. West Siang	50	36	72
	13. Dibang valley	100	29.5	29.5
	Grand total for Arunachal Pradesh	**718**	**215.5**	**30**
Orissa	1. Bolangir	100	47	47
	2. Ganjam	100	83	83
	3. Jharsuguda	96	38	40
	4. Khurda	99	60	60
	5. Boudh	99	20	20
	6. Rayagada	93	64	69
	7. Sonepur	97	27	28
	8. Gjapati	100	38	38
	9. Kalahandi	100	44	44
	10. Mayurbhanj	95	48	50
	11. Kandhmal	95	35	37
	12. Puri	100	20	20
	13. Kendrapada	100	21	21
	14. Dhenkanal	96	27	28
	15. Nayagarh	100	31	31
	16. Bargarh	96	61	64
	17. Anugul	96	51	53
	18. Bhadrak	37	6	16
	19. Balesore	52	4	8
	20-21. Sundargarh,Sambalpur	200	30	60
	22-27. Korapat,Malkangir, Nabrangpur,Jagatsinghpur, Keonjhar, Deogarh	697	251	36

States	Districts	Total No. of samples tested	Total No. positive	% FMD infected
Orissa	28. BBSR	53	42	79
	29. Orissa Network Unit 16/12/08- tested	39	31	80
	30. Orissa Network Unit 2/09/08	40	20	50
	Grand total for Orissa	**2780**	**1099**	**39.5**
Haryana	1. Bhiwani	100	46	46
	2. Fatehabad	100	21	21
	3. Hisar	100	23	23
	4. Jind	100	20	20
	5. Rohtak	98	29	30.1
	6. Sirsa	78	21	26.9
	7. Sonipat	100	18	18
	Grand total for Haryana	**676**	**178**	**26.4**
Jammu & Kashmir	1. Kargil	50	21	42
	2. Udhampur	16	2	13
	3. Leh	8	0	0
	Grand total for Jammu & Kashmir	**74**	**23**	**31**
Rajasthan	1. Udaipur	57	15	26
	2. Ajmer	109	84	77
	3. Kota	12	3	25
	4. Hanumangarh	46	28	61
	5. Sikar	26	3	11
	6. Alwar	33	0	0
	7. Bikaner	19	17	89
	8. Nagaur	20	13	65
	9. Ganganagar	21	10	48
	10. Bhilwara	86	62	72.7
	11. Tonk	33	25	75.7
	Grand total for Rajasthan	**462**	**260**	**56**
Karnataka	1. Bangalore urban (BU)	98	45.5	45.4
	2. Bangalore rural (BR)	90	65.5	72
	3. Belgaum	52	11.5	22
	4. Bellary	30	7.5	25
	5. Bidar	58	25	43
	6. Bijapur	95	44.5	46
	7. Chikkaballapur (CB.PUR)	111	65	58.5
	8. Chikkamaglore	90	46.5	51
	9. Chitradurga	100	70	70

States	Districts	Total No. of samples tested	Total No. positive	% FMD infected
Karnataka	10. Dakshina Kannada	90	15	16.6
	11. Davangere	44	29.5	67
	12. Dharwad	79	24	30
	13. Gadag	100	43	47
	14. Gulbarga	147	79	53
	15. Hassan	61	24.5	40.1
	16. Haveri	85	37	47.05
	17. Kolar	107	58.5	54.6
	18. Koppal	106	67.5	63.6
	19. Mandya	90	50	55.5
	20. Mysore	41	19.5	47
	21. Raichur	57	26	45.6
	22. Shimoga	108	41	37.9
	23. Tumkur	87	35.5	40.8
	24. Udupi	90	17	18
	25. Uttara Kannada	93	73.5	79
	Grand total for Karnataka	**2109**	**1021.5**	**48.4**
Tamil Nadu	1. Karur	100	7	7
	2. Coimbatore	100	42	42
	3. Tirupur	60	10	17
	4. Erode	100	29	29
	Grand total for Tamil Nadu	**360**	**88**	**24.4**
Assam	1. Kamrup	90	23	25.5
	2. Darrang	100	26	26
	3. Nalbari	80	44	55
	4. Kokrajhar	90	25	27.8
	5. Bongaigaon	90	31.5	35
	Grand total for Assam	**450**	**149.5**	**33.2**

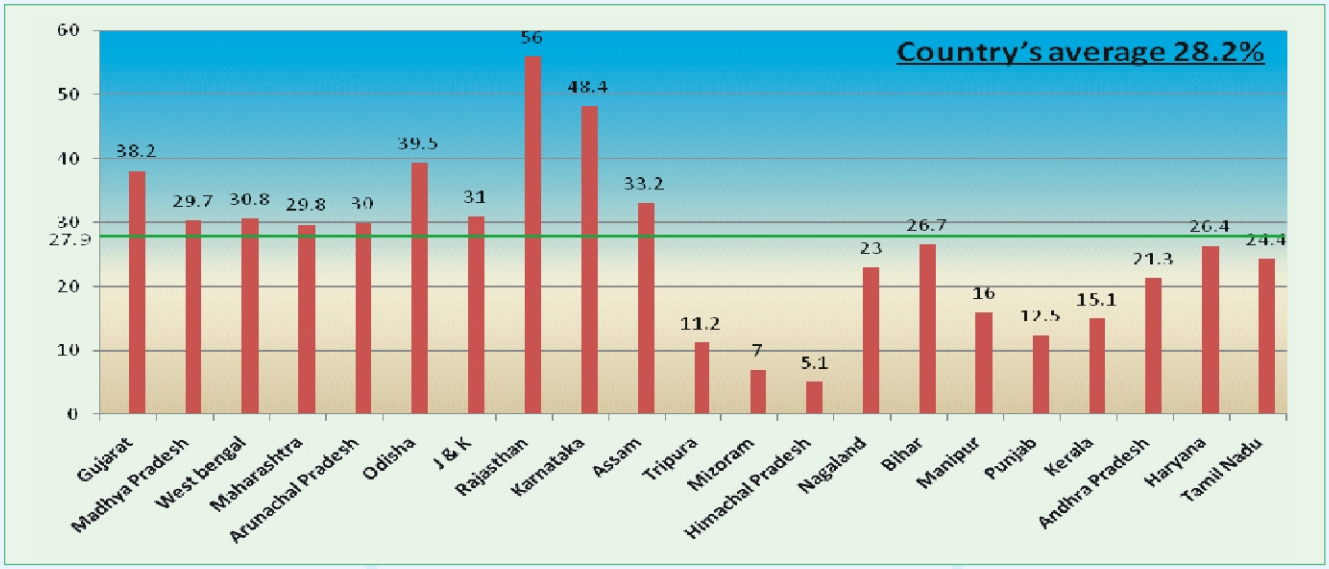


Fig. 18. The FMD seroprevalence in terms of % bovine infected (3AB3 reactors) in various states till December 2009

Fig 19. % FMD virus infected bovines during 2008-2010 depicted on individual state maps as estimated applying DIVA ELISA on random serum samples

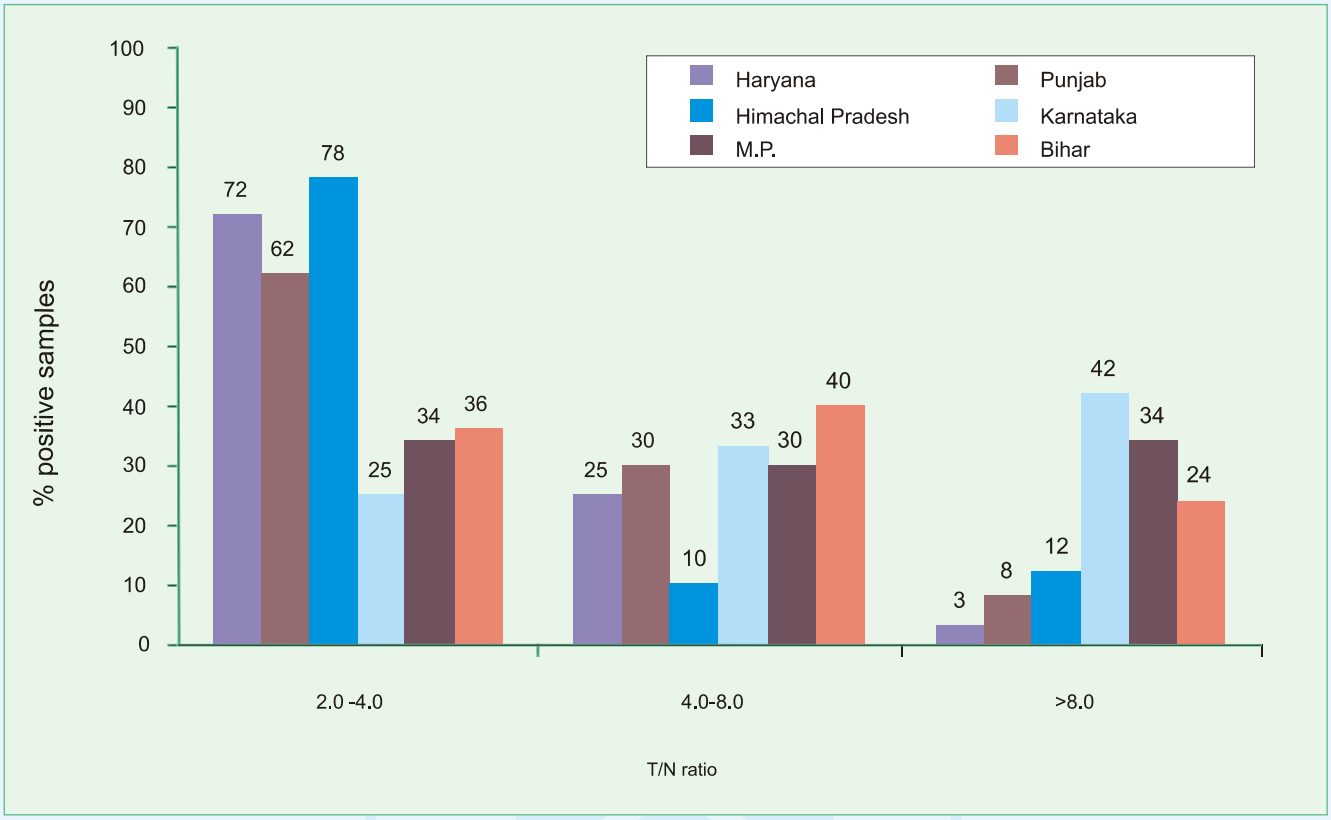


Fig 20. Distribution of positive samples in three categories of T/N ratio

10.0 Post Vaccinal Seroconversion Studies

10.1 Sero-monitoring under FMD Control Programme of Government of India

A major programme has been initiated by the Central government since August 2003 through the FMD Control Programme (FMD-CP) covering at present 54 specified districts in the country (Fig 21) to control the disease. This involves 6 monthly vaccinations of susceptible livestock against FMD. Serum samples before vaccination and 21 to 30 days post vaccination were collected and screened for level of type specific neutralizing antibodies by Liquid Phase Blocking ELISA (LPBE) developed at central FMD laboratory. The Regional Centers and Network Units of the Project Directorate participated in the post vaccinal sero-conversion study. All

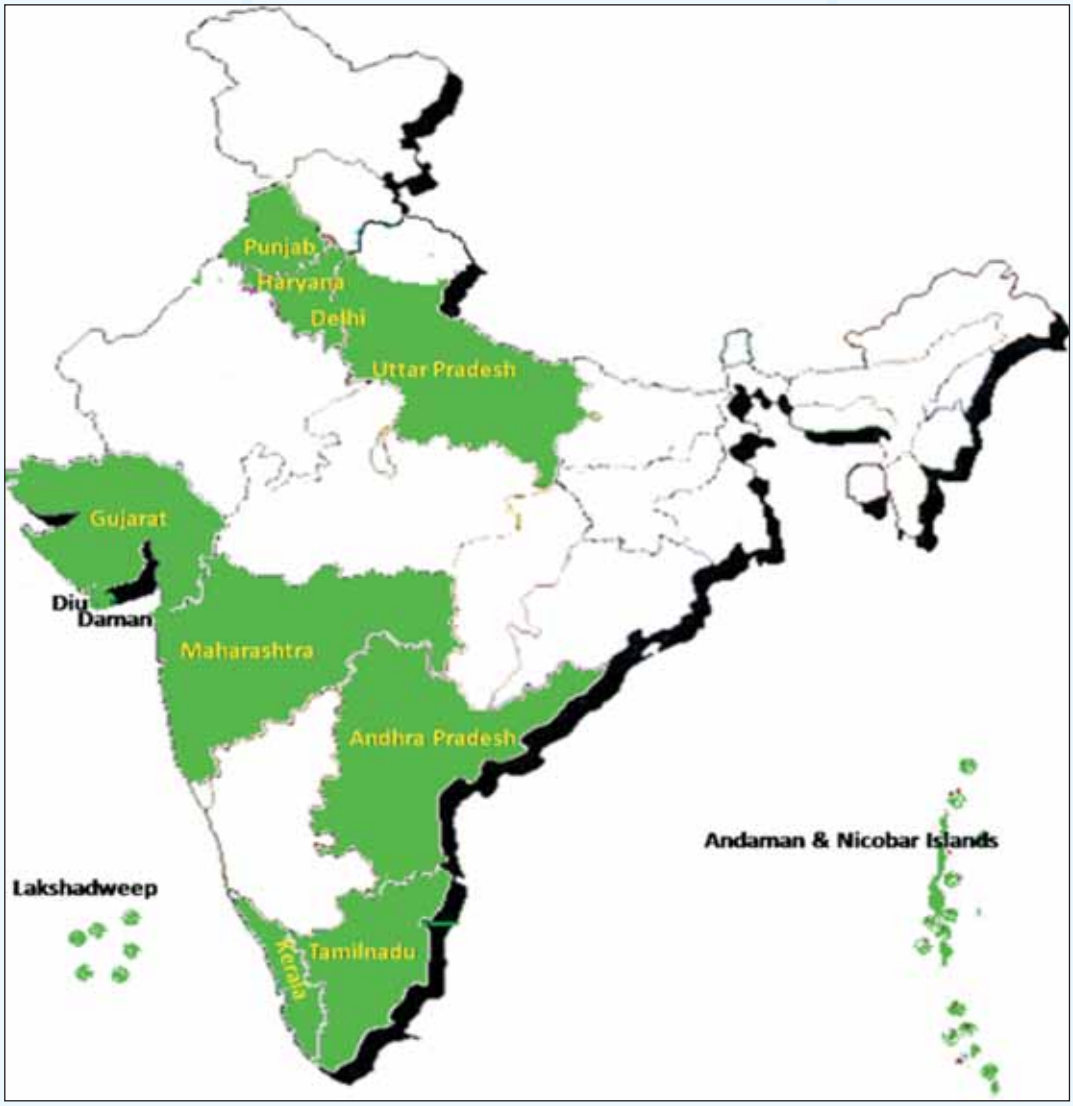


Fig 21: States covered under FMD Control Programme (Gov. of India)

reagent and training to conduct LPB ELISA were provided by the central FMD laboratory, Mukteswar. The test was compared with SNT and LPB titer (in serum) of e" $\log_{10}$ 1.8 was indicative of protection against FMD.

Currently the serum samples f 7th and 8th phase are being tested and serum samples for 9th phase are being collected. Up till now more than 95,000 pre and post vaccinal samples have been tested under FMDCP. Over the years as more rounds of vaccination are completed, there has been marked improvement in the antibody status of the animal population in FMDCP areas which is evident from the results of serum testing. After 8th phase the post vaccinal antibody titre of e"1.8 and above was found in 79.5% samples against type O, 71.6% against type A and 66.2% against type Asia1 as compared to 53.5% samples against type O, 49.5% against type A and 57.6% against type Asia1 after 1st phase. The detailed report of sero-surveillance under FMDCP is presented herewith.

10.1.1Sero-surveillance under FMDCP for Andaman & Nicobar

Eight villages from Andaman & Nicobar are covered under FMDCP namely, Junglighat, Rangachang, Portmout, Garacharama, Wimberligunj, Monglutan, Elephant Point and Dollygunj. Serum samples were tested by Kolkata Regional Center.

- No serum samples were received for phases I and II.
- In phase III, 154 pre and 162 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 25.9 for type 'O', 2.8 for type 'A' and 34.0 for type 'Asia-1'. The same for post-vac samples was 60.0 for type 'O', 20.3 for type 'A' and 73.6 for type 'Asia-1'.
- In phase IV 149 pre and 146 post-vac serum samples were tested. Percent serum samples having protective antibody titer of 1.8 and above for pre-vac samples was 33.5 for type 'O', 33.5 for type 'A' and 23.4 for type 'Asia-1'. The same for post-vac samples were 64.6 for type 'O', 65.9 for type 'A' and 67.6 for type 'Asia-1'.
- In phase V 126 pre and 122 post-vac serum samples were tested. Percent serum samples having protective antibody titer of 1.8 and above for pre-vac samples were 57.2 for type 'O', 50.8 for type 'A' and 44.3 for type 'Asia-1'. The same for post-vac samples were 55.8 for type 'O', 52.5 for type 'A' and 50.8 for type 'Asia-1'.
- In phase VI 270 pre and 270 post-vac serum samples were tested. Percent serum samples having protective antibody titer of 1.8 and above for pre-vac samples were 18.5 for type 'O', 24.4 for type 'A' and 10.2 for type 'Asia-1'. The same for post-vac samples were 29.6 for type 'O', 38.4 for type 'A' and 13.2 for type 'Asia-1'.
- In phase VII 265 pre and 265 post-vac serum samples were tested. Percent serum samples having protective antibody titer of 1.8 and above for pre-vac samples were 42.3 for type 'O', 30.9 for type 'A' and 21.1 for type 'Asia-1'. The same for post-vac samples were 65.7

Table 8. Result of seroconversion in Andaman & Nicobar Islands

Phase	Species	Number & % animals showing titres e"1.8 log_{10} against FMDV					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
I	Cattle+Buffalo	Serum samples not available					
II	Cattle+Buffalo	Serum samples not available					
III	Cattle+Buffalo	40(25.9)	97(60.0)	5(2.8)	37(20.3)	52(34.0)	118(73.6)
IV	Cattle+Buffalo	50(33.5)	94(64.6)	50(33.5)	96(65.9)	35(23.4)	101(67.6)
V	Cattle+Buffalo	72(57.2)	68(55.8)	62(50.8)	64(52.5)	54(44.3)	62(50.8)
VI	Cattle+Buffalo	50 (18.5)	80 (29.6)	66 (24.4)	104 (38.4)	28 (10.2)	36 (13.2)
VII	Cattle+Buffalo	112 (42.3)	174 (65.7)	82 (30.9)	110 (41.5)	56 (21.1)	66 (24.9)

*Serum samples were not available for Phases I & II.

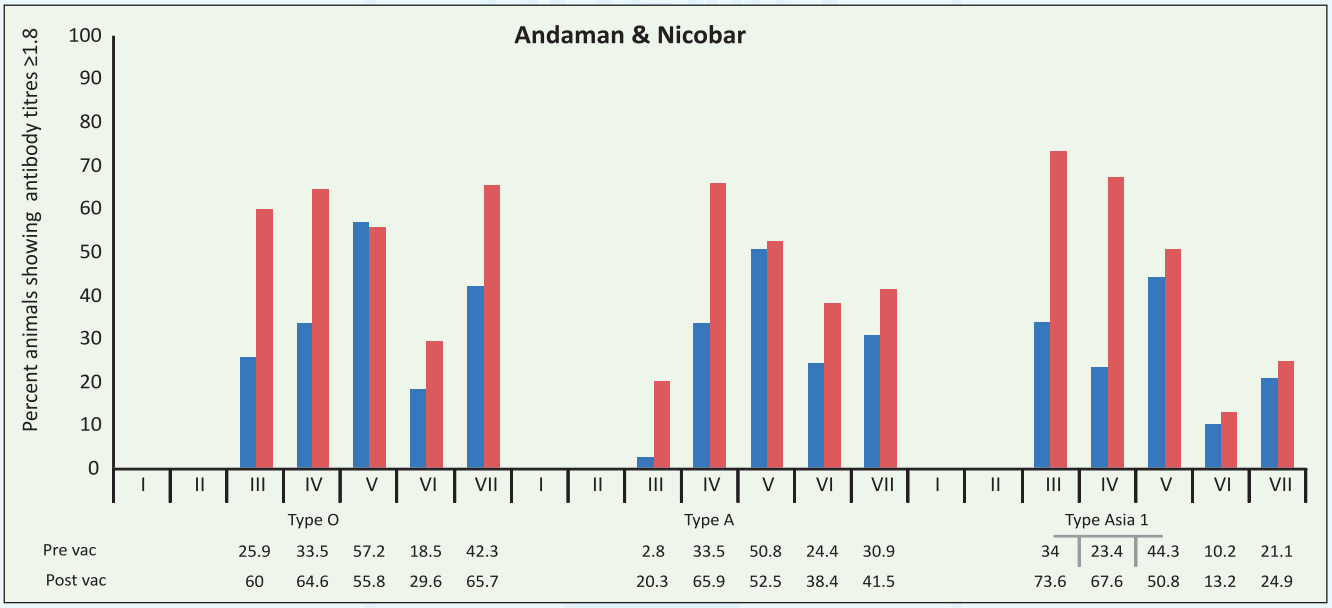


Fig 22: Seroconversion in Andaman & Nicobar

for type 'O', 41.5 for type 'A' and 24.9 for type 'Asia-1'.

- Response is poor in A & N islands.

10.1.2 Sero-surveillance under FMDCP for Andhra Pradesh

From the state of Andhra Pradesh a total of four districts are covered under FMDCP namely, Ananthapur, Chitoor, Medak and Rangareddy. The serum samples were tested by Hyderabad Regional Center.

- In phase I, 800 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of

Fig 23: Districts covered under FMD Control Programme in Andhra Pradesh

1.8 and above for pre-vac samples was 10.3 for type 'O', 5.3 for type 'A' and 11.5 for type 'Asia-1'. The same for post-vac samples was 42.5 for type 'O', 30.5 for type 'A' and 42.5 for type 'Asia-1'.

- In phase II, 800 post-vac serum samples were tested. Pre-vac serum samples were not available. Percent serum sample having protective antibody titer of 1.8 and above was 54.2 for type 'O', 62.3 for type 'A' and 54.7 for type 'Asia-1'.
- In phase III, 800 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 26.2 for type 'O', 49.3 for type 'A' and 38.2 for type 'Asia-1'. The same for post-vac samples was 35.7 for type 'O', 66.5 for type 'A' and 52.7 for type 'Asia-1'.
- In phase IV, 800 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 35.1 for type 'O', 58.1 for type 'A' and 41.1 for type 'Asia-1'. The same for post-vac samples was 46.8 for type 'O', 77.1 for type 'A' and 64.8 for type 'Asia-1'.
- In phase V, 800 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 30.8 for type 'O', 58.2 for type 'A' and 42.8 for type 'Asia-1'. The same for post-vac samples was 55.0 for type 'O', 71.8 for type 'A' and 56.3 for type 'Asia-1'.
- In phase VI, 800 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 34.3 for type 'O', 69.2 for type 'A' and 55.7 for type 'Asia-1'. The same for post-vac samples was 61.3 for type 'O', 86.3 for type 'A' and 79.3 for type 'Asia-1'.
- In phase VII, 800 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 34.0 for type 'O', 44.0 for type 'A' and 48.8 for type 'Asia-1'. The same for post-vac samples was 60.3 for type 'O', 67.5 for type 'A' and 64.7 for type 'Asia-1'.
- In phase VIII, 800 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 44.5 for type 'O', 51.8 for type 'A' and 41.6 for type 'Asia-1'. The same for post-vac samples was 74.0 for type 'O', 78.0 for type 'A' and 65.8 for type 'Asia-1'.

Table 9. Result of seroconversion in Andhra Pradesh

Phase	Species	Number & % animals showing titres e"1.8 log_{10} against FMDV					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
I	Cattle+Buffalo	83 (10.3)	340 (42.5)	43 (5.3)	244 (30.5)	92 (11.5)	340 (42.5)
II	Cattle+Buffalo	N.A.	434 (54.2)	N.A.	498 (62.3)	N.A.	438 (54.7)
III	Cattle+Buffalo	210 (26.2)	286 (35.7)	395 (49.3)	532 (66.5)	306 (38.2)	422 (52.7)
IV	Cattle+Buffalo	281 (35.1)	374 (46.8)	465 (58.1)	617 (77.1)	329 (41.1)	518 (64.8)
V	Cattle+Buffalo	247 (30.8)	440 (55)	466 (58.2)	574 (71.8)	343 (42.8)	450 (56.3)
VI	Cattle+Buffalo	275 (34.3)	490 (61.3)	554 (69.2)	690 (86.3)	446 (55.7)	634 (79.3)
VII	Cattle+Buffalo	274 (34.0)	483 (60.3)	349 (44.0)	540 (67.5)	391 (48.8)	518 (64.7)
VIII	Cattle+Buffalo	356 (44.5)	594 (74.0)	415 (51.8)	624 (78.0)	333 (41.6)	527 (65.8)
IX	Cattle+Buffalo	422 (52.8)	673 (84.1)	329 (41.1)	534 (66.8)	287 (35.9)	534 (66.8)

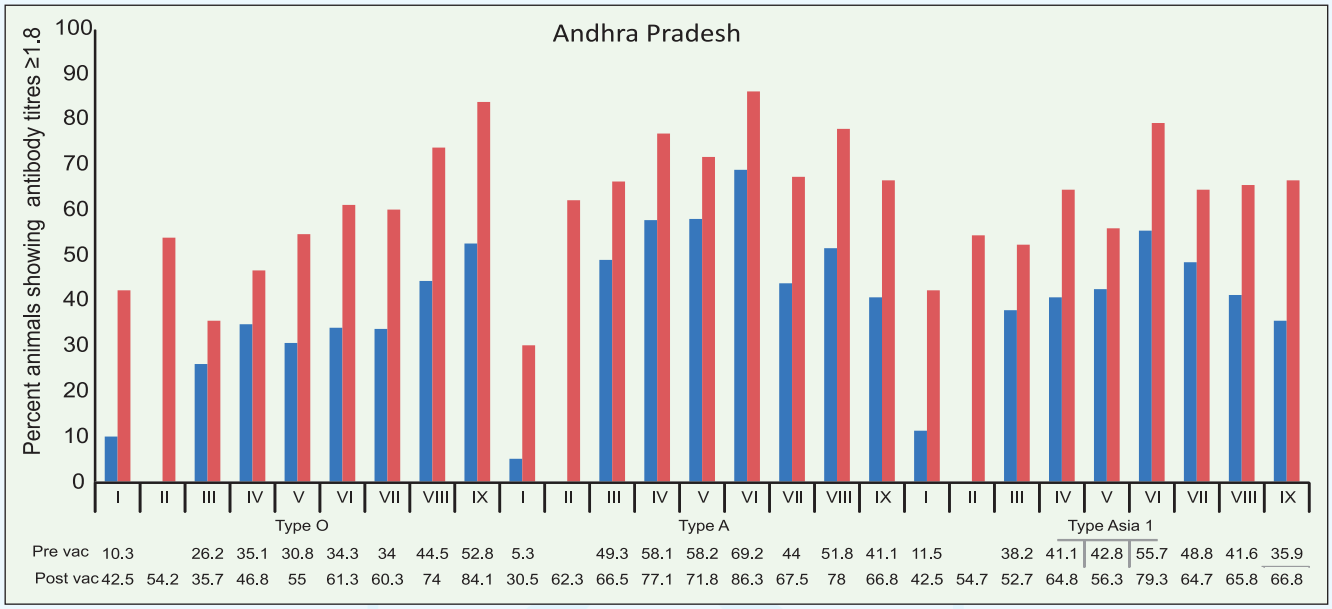


Fig 24: Seroconversion in Andhra Pradesh

- In phase IX, 800 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 52.8 for type 'O', 41.1 for type 'A' and 35.9 for type 'Asia-1'. The same for post-vac samples was 84.1 for type 'O', 66.8 for type 'A' and 66.8 for type 'Asia-1'.

10.1.3 Sero-surveillance under FMDCP for Delhi

Serum samples collected under FMDCP from the state of Delhi were tested by Hissar Regional Center.

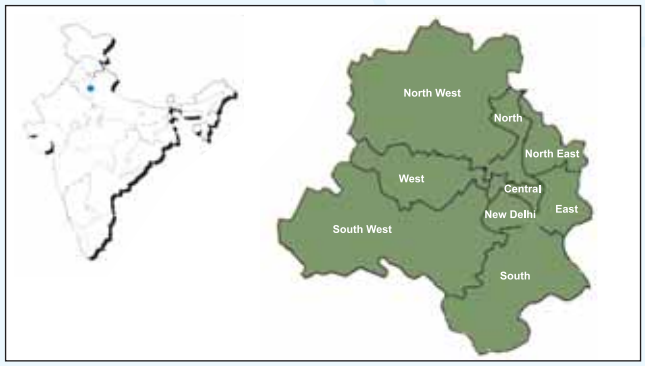


Fig 25: Districts covered under FMD Control Programme in Delhi

- In phase I, 50 each of pre and post-vac serum samples from buffaloes were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 53 for type 'O', 26 for type 'A' and 34 for type 'Asia-1'. The same for post-vac samples was 100 for type 'O', 94 for type 'A' and 96 for type 'Asia-1'.
- In phase II, 24 each of pre-vac and post-vac serum samples from buffaloes were tested. Pre-vac serum samples were not available. Percent serum sample having protective antibody titer of 1.8 and above was 91 for type 'O', 40 for type 'A' and 95 for type 'Asia-1'. The same for post-vac samples was 96 for type 'O', 62 for type 'A' and 86 for type 'Asia-1'.
- In phase III, 50 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 94 for type 'O', 60 for type 'A' and 86 for type 'Asia-1'. The same for post-vac samples was 98 for type 'O', 80 for type 'A' and 92 for type 'Asia-1'.

- In phase IV, 50 pre and 46 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 76 for type 'O', 28 for type 'A' and 54 for type 'Asia-1'. The same for post-vac samples was 82.6 for type 'O', 86.9 for type 'A' and 89.1 for type 'Asia-1'.
- In phase V 44 pre and 53 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 59 for type 'O', 52.2 for type 'A' and 72.7 for type 'Asia-1'. The same for post-vac samples was 88.6 for type 'O', 69.8 for type 'A' and 77.3 for type 'Asia-1'.
- In phase VI, 98 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 77.5 for type 'O', 61.2 for type 'A' and 72.4 for type 'Asia-1'. The same for post-vac samples was 98.9 for type 'O', 94.9 for type 'A' and 98.9 for type 'Asia-1'.
- In phase VII, 50 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 78

Table 10. Result of seroconversion in Delhi

Phase	Species	Number & % animals showing titres e"1.8 log_{10} against FMDV					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
I	Buffalo	26 (53)	50 (100)	13 (26)	47 (94)	17 (34)	48 (96)
II	Buffalo	22 (91)	23 (96)	12 (40)	15 (62)	23 (95)	22 (86)
III	Cattle+Buffalo	47 (94)	49 (98)	30 (60)	40 (80)	43 (86)	46 (92)
IV	Cattle+Buffalo	38 (76)	38 (82.6)	14 (28)	40 (86.9)	27 (54)	41 (89.1)
V	Cattle+Buffalo	26 (59)	47 (88.6)	23 (52.2)	37 (69.8)	32 (72.7)	41 (77.3)
VI	Cattle+Buffalo	76 (77.5)	97 (98.9)	60 (61.2)	93 (94.9)	71 (72.4)	97 (98.9)
VII	Cattle+Buffalo	39(78)	44(88)	33(66)	43(86)	25(50)	41(82)
VIII	Cattle+Buffalo	92 (92)	100 (100)	66 (66)	86 (86)	83 (83)	98 (98)

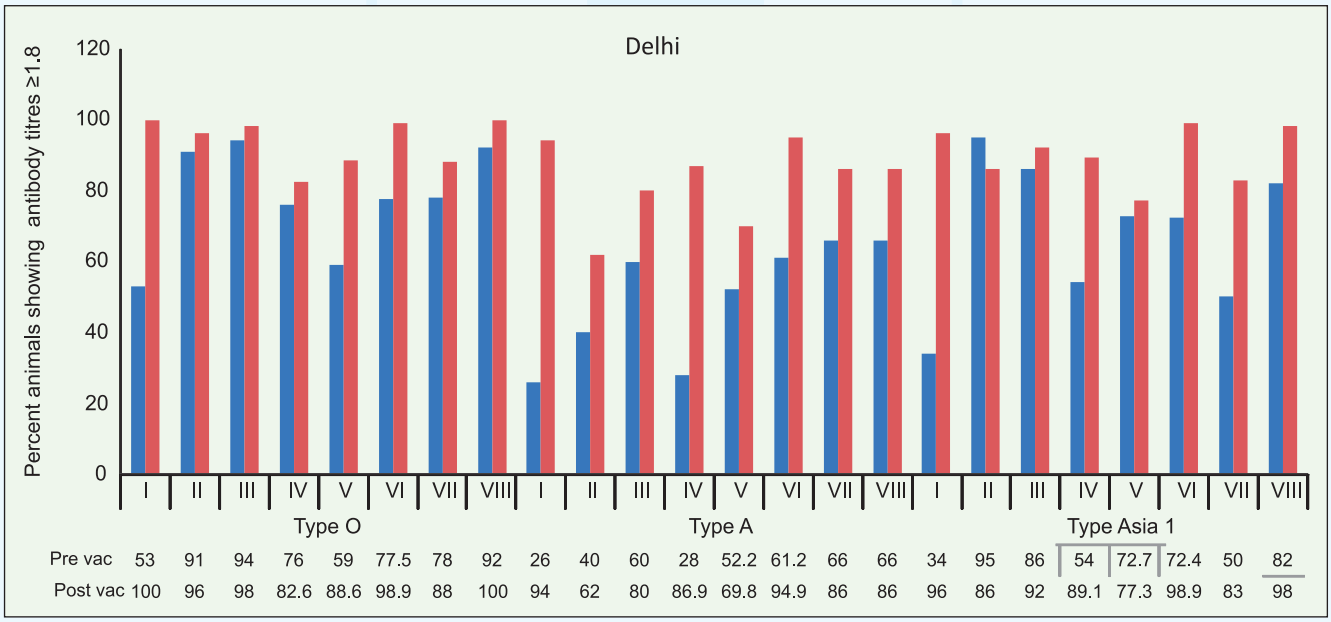


Fig 26: Seroconversion in Delhi

for type 'O', 66 for type 'A' and 50 for type 'Asia-1'. The same for post-vac samples was 88 for type 'O', 86 for type 'A' and 82 for type 'Asia-1'.

- In phase VIII, 100 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 92 for type 'O', 66 for type 'A' and 83 for type 'Asia-1'. The same for post-vac samples was 100 for type 'O', 86 for type 'A' and 98 for type 'Asia-1'.
- Vaccination programme in Delhi region achieved and crossed 80% herd immunity level after eight phases of vaccination.

10.1.4Sero-surveillance under FMDCP for Gujarat

Under FMDCP four districts are covered namely, Banaskantha, Sabarkantha, Mehsana and Patan.

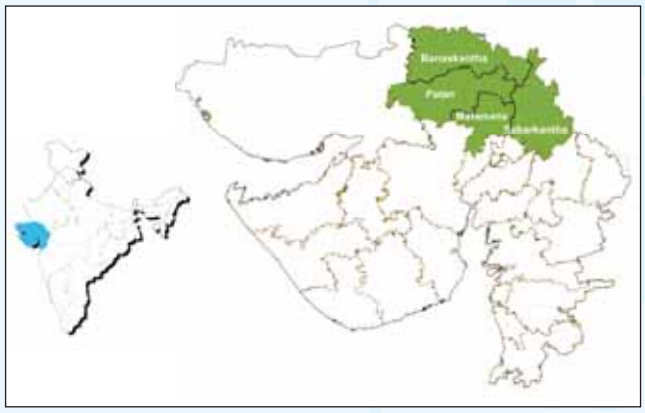


Fig27: Districts covered under FMD Control Programme in Gujarat

- Till fifth phase serum samples were tested at Pune Regional Center and sixth phase onwards the samples were tested at Ahmadabad Network Unit.
- In phase I, 382 pre and 259 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 19.1 for type 'O', 24.5 for type 'A' and 16.1 for type 'Asia-1'. The same for post-vac samples was 44.7 for type 'O', 48.7 for type 'A' and 43.5 for type 'Asia-1'.
- Serum samples were not available for Phase II.
- In phase III, 442 pre and 357 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 27.8 for type 'O', 39.2 for type 'A' and 12.4 for type 'Asia-1'. The same for post-vac samples was 47.9 for type 'O', 58.3 for type 'A' and 35.4 for type 'Asia-1'.
- In phase IV, 497 and 456 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 22.7 for type 'O', 40.7 for type 'A' and 14.6 for type 'Asia-1'. The same for post-vac samples was 60.7 for type 'O', 81.2 for type 'A' and 46.8 for type 'Asia-1'.
- In phase V, 195 pre and 202 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 23.6 for type 'O', 66.1 for type 'A' and 26.5 for type 'Asia-1'. The same for post-vac samples was 49 for type 'O', 91.6 for type 'A' and 51.3 for type 'Asia-1'.
- In phase VI, 395 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 30.1 for type 'O', 63.0 for type 'A' and 49.3 for type 'Asia-1'. The same for post-vac samples was 56.4 for type 'O', 80.2 for type 'A' and 60.7 for type 'Asia-1'.
- In phase VII, 800 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 30.1 for type 'O', 63.0 for type 'A' and 49.3 for

Table 11. Result of seroconversion in Gujarat

Phase	Species	Number & % animals showing titres e"1.8 log_{10} against FMDV					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
I	Cattle+Buffalo	50 (19.1)	116 (44.7)	59 (24.5)	128 (48.7)	42 (16.1)	114 (43.5)
II	Cattle+Buffalo	Serum samples not available					
III	Cattle+Buffalo	123 (27.8)	171 (47.9)	171 (39.2)	268 (58.3)	51 (12.4)	149 (35.4)
IV	Cattle+Buffalo	113 (22.7)	277 (60.7)	184 (40.7)	355 (81.2)	73 (14.6)	218 (46.8)
V	Cattle+Buffalo	46 (23.6)	99 (49.0)	126 (66.1)	179 (91.6)	44 (26.5)	92 (51.3)
VI	Cattle+Buffalo	119 (30.1)	223 (56.4)	249 (63.0)	317(80.2)	195 (49.3)	240 (60.7)
VII	Cattle+Buffalo	434 (54.3)	630 (78.8)	385 (48.1)	559 (69.9)	344 (43.0)	556 (69.5)
VIII	Cattle+Buffalo	191 (23.9)	394 (49.3)	197 (24.6)	357 (44.6)	264 (33.0)	403 (50.4)

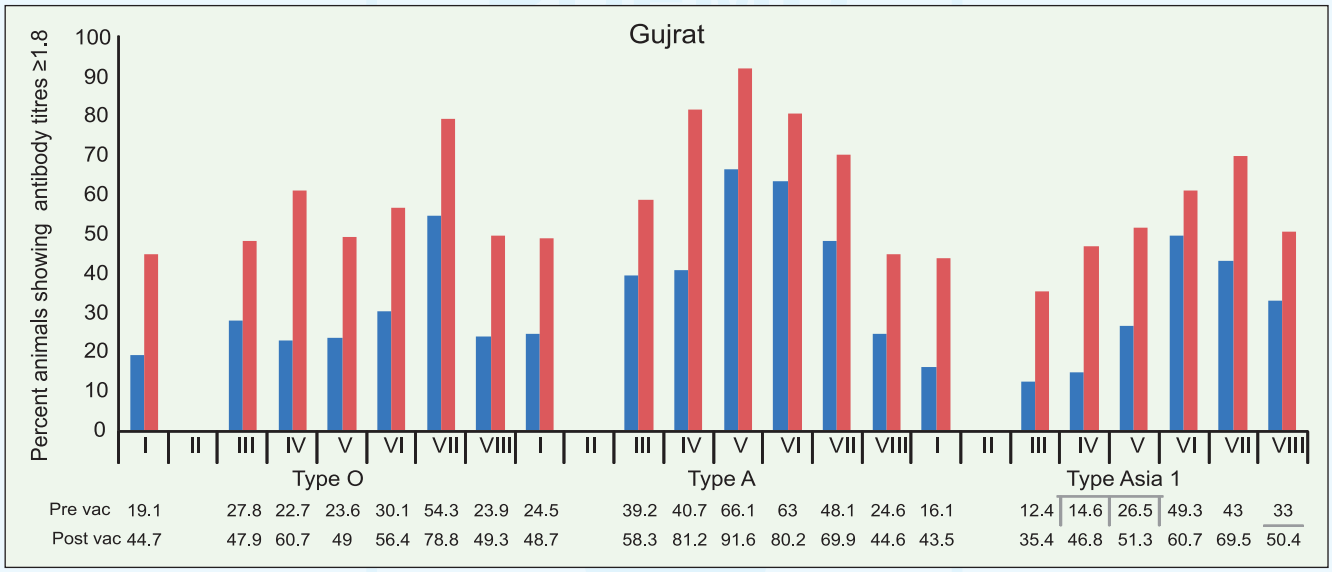


Fig 28: Seroconversion in Gujarat

type 'Asia-1'. The same for post-vac samples was 56.4 for type 'O', 80.2 for type 'A' and 60.7 for type 'Asia-1'.

- In phase VIII, 800 each of pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 30.1 for type 'O', 63.0 for type 'A' and 49.3 for type 'Asia-1'. The same for post-vac samples was 56.4 for type 'O', 80.2 for type 'A' and 60.7 for type 'Asia-1'.

10.1.4 Sero-surveillance under FMDCP for Haryana

Under FMDCP eight districts are covered

Fig 29: Districts covered under FMD Control Programme in Haryana

namely, Bhiwani, Fatehabad, Hisar, Jhajjar, Jind, Rohtak, Sirsa and Sonipat.

- Serum samples were tested by Hissar Regional Center.
- Serum samples were not available for Phase I.
- In phase II, 1558 post-vac serum samples were tested. Pre-vac serum samples were not available. Percent serum sample having protective antibody titer of 1.8 and above was 68.3 for type 'O', 55.1 for type 'A' and 53.3 for type 'Asia-1'.
- In phase III, 1585 post-vac serum samples were tested. Pre-vac serum samples were not available. Percent serum sample having protective antibody titer of 1.8 and above was 72.3 for type 'O', 63.6 for type 'A' and 63.4 for type 'Asia-1'.
- In phase IV, 1589 pre and 1552 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 60.1 for type 'O', 42.1 for type 'A' and 53.2 for type 'Asia-1'. The same for post-vac samples was 78.7 for type 'O', 57.1 for type 'A' and 75.3 for type 'Asia-1'.
- In phase V, 1600 pre and 1599 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 59.7 for type 'O', 50.8 for type 'A' and 58.8 for type 'Asia-1'. The same for post-vac samples was 84.5 for type 'O', 79.6 for type 'A' and 84.5 for type 'Asia-1'.
- In phase VI, 1496 pre and 1499 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 66.5 for type 'O', 59.8 for type 'A' and 56.4 for type 'Asia-1'. The same for post-vac samples was 87.1 for type 'O', 82 for type 'A' and 74.6 for type 'Asia-1'.
- In phase VII, 1562 pre and 1574 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 54.8 for type 'O', 65.3 for type 'A' and 56.8 for type 'Asia-1'. The same for post-vac samples was 82.3 for type 'O', 87.6 for type 'A' and 83.6 for type 'Asia-1'.
- In phase VIII, 1547 pre and 1540 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 61.3 for type 'O', 56.6 for type 'A' and 49.4 for type 'Asia-1'. The same for post-vac samples was 83.7 for type 'O', 64.4 for type 'A' and 71.4 for type 'Asia-1'.

Table 12. Result of seroconversion in Haryana

Phase	Species	Number & % animals showing titres e"1.8 log_{10} against FMDV					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
I	Cattle+Buffalo	Serum samples not available					
II	Cattle+Buffalo	NA	1065(68.3)	NA	859 (55.1)	NA	831 (53.3)
III	Cattle+Buffalo	NA	1146(72.3)	NA	1007(63.6)	NA	1005(63.4)
IV	Cattle+Buffalo	953 (60.1)	1222(78.7)	668 (42.1)	887 (57.1)	844(53.2)	1170(75.3)
V	Cattle+Buffalo	955 (59.7)	1352(84.5)	813 (50.8)	1274(79.6)	941(58.8)	1353(84.5)
VI	Cattle+Buffalo	995 (66.5)	1306(87.1)	895 (59.8)	1229(82.0)	844(56.4)	1118(74.6)
VII	Cattle+Buffalo	856(54.8)	1296 (82.3)	1021 (65.3)	1380 (87.6)	888 (56.8)	1317 (83.6)
VIII	Cattle+Buffalo	949(61.3)	1289 (83.7)	877 (56.6)	992 (64.4)	765 (49.4)	1101 (71.4)

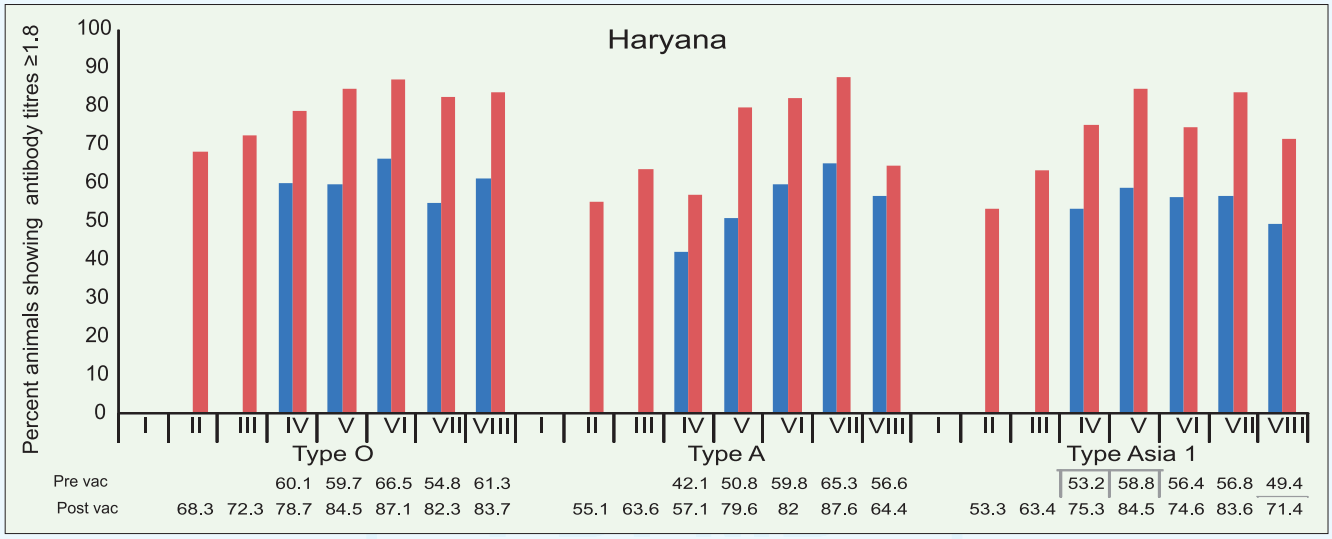


Fig 30: Seroconversion in Haryana

10.1.6 Sero-surveillance under FMDCP for Kerala

Three districts namely, Trivandrum, Kollam and Pathanamthitta are covered under FMDCP.

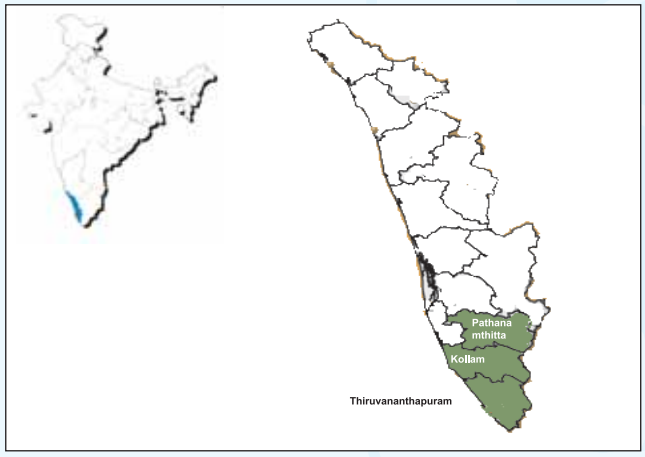


Fig 31: Districts covered under FMD Control Programme in Kerala

- Serum samples were tested by Ranipet Network Unit.
- In phase I, II & IV 483 pre and 496 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 32.7 for type 'O', 29 for type 'A' and 34.2 for type 'Asia-1'. The same for post-vac samples was 51.4 for type 'O', 47.5 for type 'A' and 56.4 for type 'Asia-1'.
- For phase III, serum samples were not available.
- In phase V, each of 290 pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 23.1 for type 'O', 17.9 for type 'A' and 21 for type 'Asia-1'. The same for post-vac samples was 67.9 for type 'O', 58.9 for type 'A' and 72.7 for type 'Asia-1'.
- In phase VI, each of 70 pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 20.4 for type 'O', 17.1 for type 'A' and 15.8 for type 'Asia-1'. The same for post-vac samples was 77.1 for type 'O', 70.4 for type 'A' and 71.3 for type 'Asia-1'.
- In phase VII, each of 300 pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 16.0 for type 'O', 14.3 for type 'A' and 17.3 for type 'Asia-1'. The same for post-vac samples was 69.3 for type 'O', 71.0 for type 'A' and 70.0 for type 'Asia-1'.

Table 13. Result of seroconversion in Kerala

Phase	Species	Number & % animals showing titres e"1.8 log_{10} against FMDV					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
I	Cattle+Buffalo	158(32.7)	255(51.4)	140(29.0)	236(47.5)	165(34.2)	280(56.4)
II	Cattle+Buffalo	158(32.7)	255(51.4)	140(29.0)	236(47.5)	165(34.2)	280(56.4)
III	Cattle+Buffalo	Serum samples not available					
IV	Cattle+Buffalo	158(32.7)	255(51.4)	140(29.0)	236(47.5)	165(34.2)	280(56.4)
V	Cattle+Buffalo	67(23.1)	197(67.9)	52(17.9)	171(58.9)	61(21.0)	211(72.7)
VI	Cattle+Buffalo	49 (20.4)	185(77.1)	41(17.1)	169(70.4)	38(15.8)	171(71.3)
VII	Cattle+Buffalo	48 (16.0)	208 (69.3)	43 (14.3)	213 (71.0)	52 (17.3)	210 (70.0)

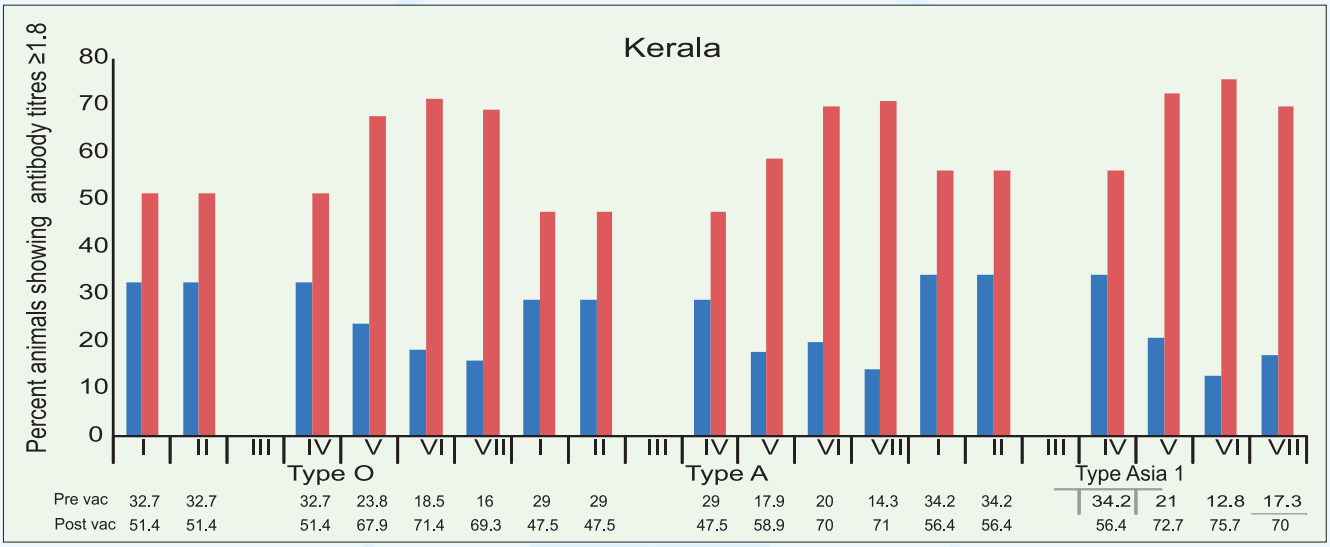


Fig 32: Seroconversion in Kerala

10.1.7 Sero-surveillance under FMDCP for Maharashtra

Under FMDCP six districts are covered namely, Ahmadnagar, Aurangabad, Pune, Satara, Mumbai and Thane.

Fig 33: Districts covered under FMD Control Programme in Maharashtra

- Serum samples were submitted to Pune FMD center for testing.
- In phase I, 844 pre and 761 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 20.5 for type 'O', 17.9 for type 'A' and 22.8 for type 'Asia-1'. The same for post-vac samples was 59.9 for type 'O', 57.4 for type 'A' and 61.2 for type 'Asia-1'.
- In phase II, 834 post-vac serum samples were tested. Pre-vac serum samples were not available. Percent serum sample having protective antibody titer of 1.8 and above was 60.9 for type 'O', 58.6 for type 'A' and 66.2 for type 'Asia-1'.
- In phase III, 753 pre and 799 post-vac serum

samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 34.4 for type 'O', 46.8 for type 'A' and 34.7 for type 'Asia-1'. The same for post-vac samples was 54.8 for type 'O', 72.7 for type 'A' and 66.9 for type 'Asia-1'.

- In phase IV, 789 and 797 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 24.2 for type 'O', 65.6 for type 'A' and 35.2 for type 'Asia-1'. The same for post-vac samples was 52.3 for type 'O', 85.3 for type 'A' and 63.9 for type 'Asia-1'.
- In phase V, 802 pre and 772 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 17.7 for type 'O', 44.2 for type 'A' and 15 for type 'Asia-1'. The same for post-vac samples was 35.1 for type 'O', 62.3 for type 'A' and 31.8 for type 'Asia-1'.
- In phase VI, 901 pre and 928 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 44.9 for type 'O', 69 for type 'A' and 27.2 for type 'Asia-1'. The same for post-vac samples was 71.4 for type 'O', 91.9 for type 'A' and 48.1 for type

Table 14. Result of seroconversion in Maharashtra

Phase	Species	Number & % animals showing titres e"1.8 log_{10} against FMDV					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
I	Cattle+Buffalo	173 (20.5)	456 (59.9)	151(17.9)	437 (57.4)	192 (22.8)	466 (61.2)
II	Cattle+Buffalo	N.A.	508 (60.9)	N.A.	490 (58.6)	N.A.	553 (66.2)
III	Cattle+Buffalo	184 (24.4)	438 (54.8)	351 (46.8)	580 (72.7)	262 (34.7)	534 (66.9)
IV	Cattle+Buffalo	191 (24.2)	417 (52.3)	517 (65.6)	679 (85.3)	278 (35.2)	509 (63.9)
V	Cattle+Buffalo	142 (17.7)	271 (35.1)	353 (44.2)	477 (62.3)	121 (15.0)	245 (31.8)
VI	Cattle+Buffalo	404 (44.9)	663 (71.4)	622 (69)	853 (91.9)	245 (27.2)	446 (48.1)
VII	Cattle+Buffalo	446 (44.6)	692 (69.2)	701 (70.1)	893 (89.3)	431 (43.1)	667 (66.7)
VIII	Cattle+Buffalo	646 (64.6)	904 (90.4)	574 (57.4)	848 (84.8)	198 (19.8)	452 (45.2)

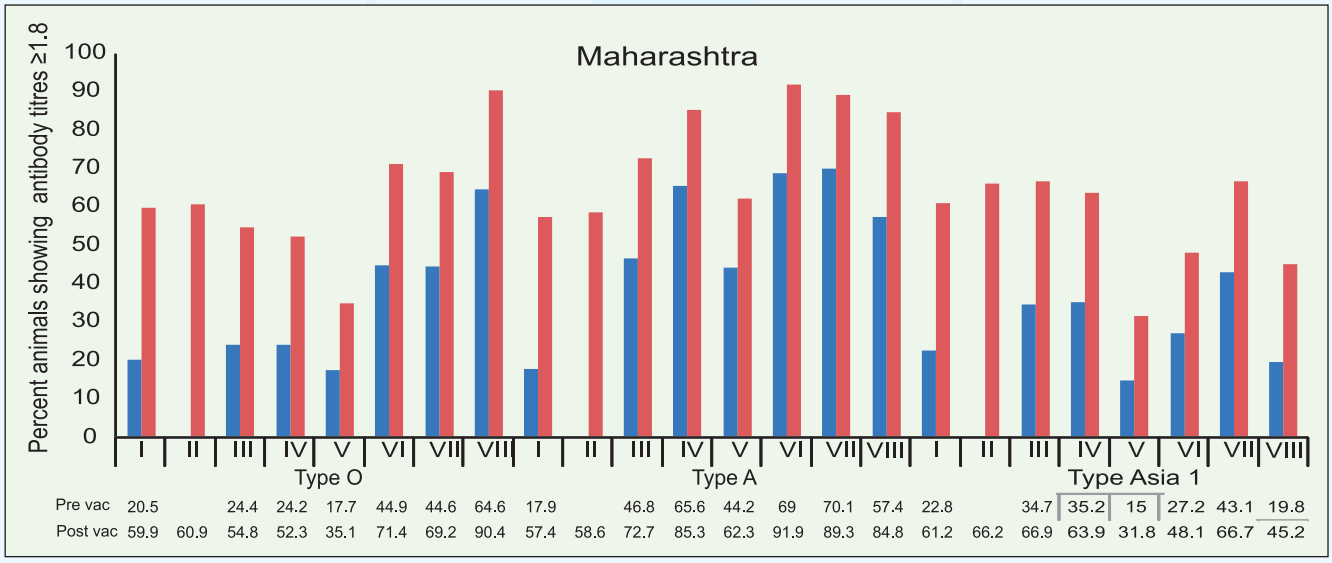


Fig 34: Seroconversion in Maharashtra

'Asia-1'.

- In phase VII, 1000 pre and 1000 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 44.6 for type 'O', 70.1 for type 'A' and 43.1 for type 'Asia-1'. The same for post-vac samples was 69.2 for type 'O', 89.3 for type 'A' and 66.7 for type 'Asia-1'.
- In phase VIII, 1000 pre and 1000 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 64.6 for type 'O', 57.4 for type 'A' and 19.8 for type 'Asia-1'. The same for post-vac samples was 90.4 for type 'O', 84.8 for type 'A' and 45.2 for type 'Asia-1'.

10.1.8 Sero-surveillance under FMDCP for Punjab

Under FMDCP eight districts are covered namely, Amritsar, Bhatinda , Fatehgarh Sahib, Ferozpur , Mansa , Sangrur, Patiala and Gurdaspur.

Fig 35: Districts covered under FMD Control Programme in Punjab

- In phase I, 742 post-vac serum samples were tested. Pre-vac serum samples were not available. Percent serum sample having protective antibody titer of 1.8 and above was 25.2 for type 'O', 11.5 for type 'A' and 49.5 for type 'Asia-1'.
- In phase II, 500 post-vac serum samples were tested. Pre-vac serum samples were not available. Percent serum sample having protective antibody titer of 1.8 and above was 43.8 for type 'O', 20.9 for type 'A' and 58.1 for type 'Asia-1'.
- In Phase III, 1084 pre and 1365 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 84.4 for type 'O', 75.3 for type 'A' and 40.2 for type 'Asia-1'. The same for post-vac samples was 86.1 for type 'O', 73.8 for type 'A' and 42.0 for type 'Asia-1'.
- In phase IV, 1291 pre and 978 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 76.5 for type 'O', 61.5 for type 'A' and 53.8 for type 'Asia-1'. The same for post-vac samples was 81.0 for type 'O', 64.1 for type 'A' and 36.4 for type 'Asia-1'.
- In phase V, 1370 pre and 1139 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 34.8 for type 'O', 32.8 for type 'A' and 38.5 for type 'Asia-1'. The same for post-vac samples was 54.5 for type 'O', 53.7 for type 'A' and 60.1 for type 'Asia-1'.
- In phase VI, 1509 pre and 1568 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 43.3 for type 'O', 43.3 for type 'A' and 32.9 for type 'Asia-1'. The same for post-vac samples was 60.2 for type 'O', 58.7 for type 'A' and 47.4 for type 'Asia-1'.
- In phase VII, 1139 pre and 1124 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 36.3 for type 'O', 22.8 for type 'A' and 33.0 for

Table 15. Result of seroconversion in Punjab

Phase	Species	Number & % animals showing titres e"1.8 log_{10} against FMDV					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
I	Cattle+Buffalo	N.A.	187(25.2)	N.A.	90(11.5)	N.A.	273(49.5)
II	Cattle+Buffalo	N.A.	219(43.8)	N.A.	113(20.9)	N.A.	279(58.1)
III	Cattle+Buffalo	915(84.4)	1175(86.1)	816(75.3)	1007(73.8)	437(40.2)	573(42.0)
IV	Cattle+Buffalo	988(76.5)	792 (81.0)	794(61.5)	627 (64.1)	694 (53.8)	356(36.4)
V	Cattle+Buffalo	477(34.8)	621(54.5)	445(32.8)	630(53.7)	513(38.5)	690(60.1)
VI	Cattle+Buffalo	653 (43.3)	944 (60.2)	654 (43.3)	921 (58.7)	496 (32.9)	743 (47.4)
VII	Cattle+Buffalo	413 (36.3)	650 (57.8)	260 (22.8)	472 (42.0)	376 (33.0)	521 (46.4)

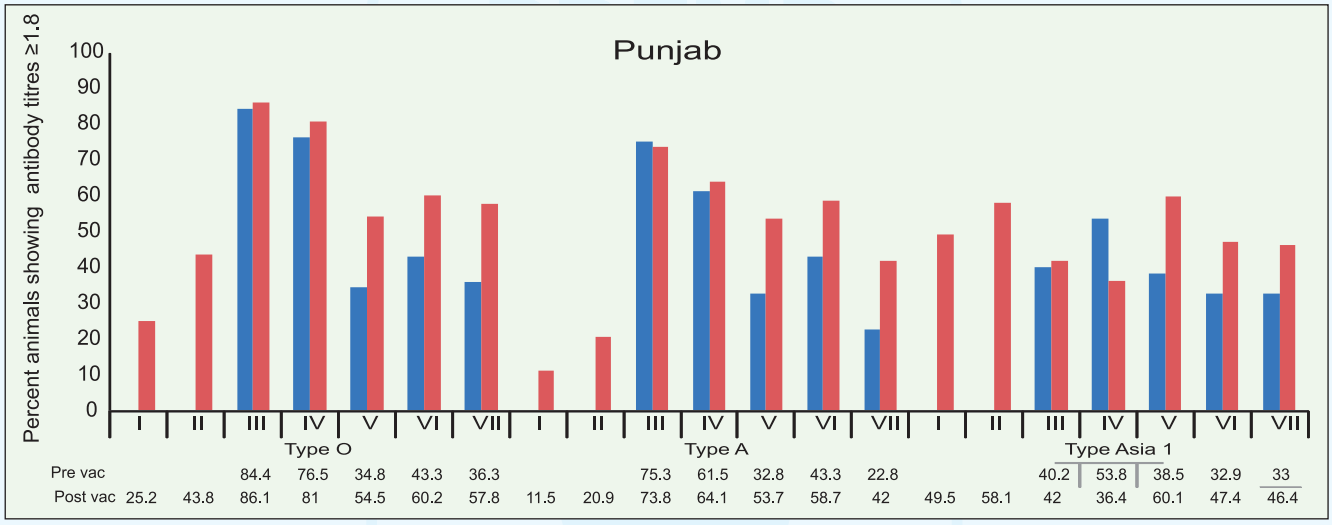


Fig 36: Seroconversion in Punjab

type 'Asia-1'. The same for post-vac samples was 57.8 for type 'O', 42.0 for type 'A' and 46.4 for type 'Asia-1'.

10.1.9 Sero-surveillance under FMDCP for Tamil Nadu

Kanyakumari district is covered under FMDCP.

- Serum samples were tested by Ranipet centre.
- In phase I, each of 100 pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 28 for type 'O', 29 for type 'A' and 24 for type 'Asia-1'. The same for post-vac samples was 51 for type 'O', 57 for type 'A' and 54 for type 'Asia-1'.

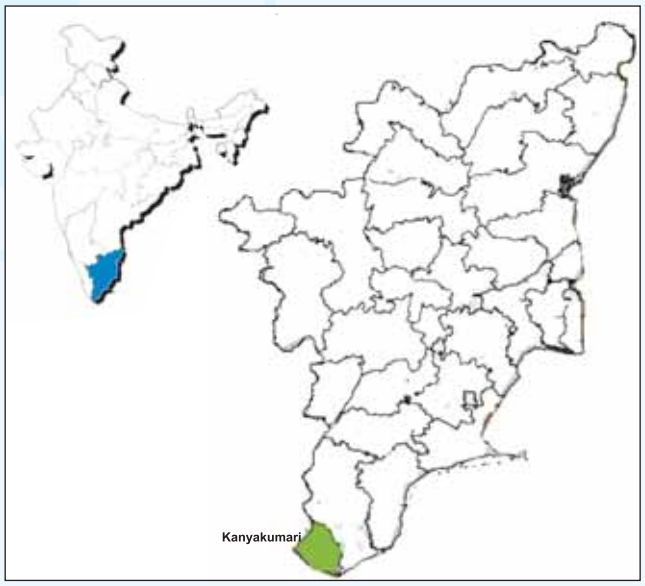


Fig 37: Districts covered under FMD Control Programme in Tamil Nadu

- In phase II, each of 100 pre and post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 23 for type 'O', 24 for type 'A' and 18 for type 'Asia-1'. The same for post-vac samples was 63 for type 'O', 40 for type 'A' and 61 for type 'Asia-1'.
- In phase III & IV, 180 pre and 330 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 32.7 for type 'O', 33.8 for type 'A' and 25 for type 'Asia-1'. The same for post-vac samples was 74.5 for type 'O', 60.9 for type 'A' and 65.4 for type 'Asia-1'.
- For phase V, serum samples were not available.
- In phase VI, 160 pre and 130 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 18.7 for type 'O', 23.8 for type 'A' and 21.5 for type 'Asia-1'. The same for post-vac samples was 76.1 for type 'O', 83.8 for type 'A' and 79.2 for type 'Asia-1'.

Table 16. Result of seroconversion in Tamil Nadu

Phase	Species	Number & % animals showing titres e"1.8 log_{10} against FMDV					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
I	Cattle+Buffalo	28(28)	51(51)	29(29)	57(57)	24(24)	54(54)
II	Cattle+Buffalo	23(23.0)	63(63.0)	24(24.0)	40(40.0)	18(18.0)	61(61.0)
III	Cattle+Buffalo	59(32.7)	246(74.5)	61(33.8)	201(60.9)	45(25.0)	216(65.4)
IV	Cattle+Buffalo	59(32.7)	246(74.5)	61(33.8)	201(60.9)	45(25.0)	216(65.4)
V	Cattle+Buffalo	Serum samples not available					
VI	Cattle+Buffalo	30(18.7)	99(76.1)	31(23.8)	109(83.8)	28(21.5)	103(79.2)
VII	Cattle+Buffalo	35(11.7)	210(70)	34(11.3)	231(77)	36(12)	226(75.3)
VIII	Cattle+Buffalo	Serum testing is in progress					
IX	Cattle+Buffalo	72(36)	42(21)	47(23.5)	153(76.5)	55(27.5)	65(32.5)

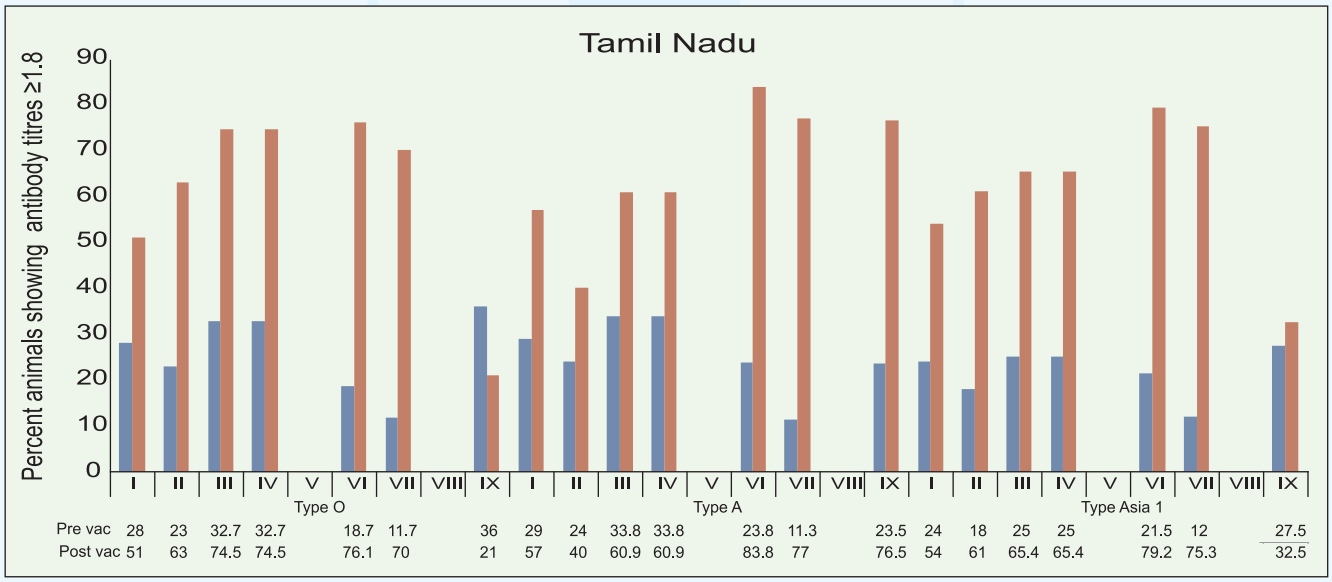


Fig 38: Seroconversion in Tamil Nadu

- In phase VII, 300 pre and 300 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 11.7 for type 'O', 11.3 for type 'A' and 12.0 for type 'Asia-1'. The same for post-vac samples was 70.0 for type 'O', 77.0 for type 'A' and 75.3 for type 'Asia-1'.
- For phase VIII, serum testing is in progress.
- In phase IX, 200 pre and 200 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 36 for type 'O', 23.5 for type 'A' and 27.5 for type 'Asia-1'. The same for post-vac samples was 21 for type 'O', 76.5 for type 'A' and 32.5 for type 'Asia-1'.

10.1.10 Sero-surveillance under FMDCP for Uttar Pradesh

Total of 16 districts in UP (Agra, Aligarh, Budaun, Bulandsahar, Etah, Ferozabad, Gautam Bhuddha Nagar, Gaziabad, Hatras, J.P.Nagar, Mathura, Meerut, Baghpat, Saharanpur, Muzaffarnagar and Muradabad) are covered under FMDCP.

Fig 39: Districts covered under FMD Control Programme in Uttar Pradesh

- Mathura center received and tested serum samples collected in 12 districts of UP (Agra, Aligarh, Budaun Bulandsahar, Etah, Ferozabad, Gautam Bhuddha Nagar, Gaziabad, Hatras, J.P.Nagar Mathura and Muradabad) during phases II to VI.
- Bangalore center received and tested serum samples collected from 4 districts of UP (Meerut, Baghpat, Saharanpur and Muzaffarnagar) during phases III to VI.
- No serum samples were received for phase I.
- In phase II, 139 and 407 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 0 for type 'O', 'A' and 'Asia-1'. The same for post-vac samples was 44.2 for type 'O', 38.1 for type 'A' and 72.0 for type 'Asia-1'.
- In phase III, 1155 and 1584 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 34.5 for type 'O', 42.7 for type 'A' and 42.4 for type 'Asia-1'. The same for post-vac samples was 49.2 for type 'O', 57.4 for type 'A' and 71.8 for type 'Asia-1'.
- In phase IV, 1910 and 1770 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 18 for type 'O', 31.9 for type 'A' and 27.2 for type 'Asia-1'. The same for post-vac samples was 30.3 for type 'O', 48.9 for type 'A' and 45.6 for type 'Asia-1'.
- In phase V, 1440 pre and 1591 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 35.8 for type 'O', 43.4 for type 'A' and 47.5 for type 'Asia-1'. The same for post-vac samples was 44.9 for type 'O', 50.4 for type 'A' and 49.4 for type 'Asia-1'.
- In phase VI at present 1488 pre and 1579 post vac serum samples out of total 2182 pre and 1986 post vac serum samples collected were tested. Percent serum sample

Table 17. Result of seroconversion in Uttar Pradesh

Phase	Species	Number & % animals showing titres e"1.8 log_{10} against FMDV					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
I	Cattle+Buffalo	Serum samples not available					
II	Cattle+Buffalo	0(0)	180(44.2)	0(0)	155(38.1)	0(0)	293(72.0)
III	Cattle+Buffalo	399(34.5)	780(49.2)	494(42.7)	910(57.4)	490(42.4)	1138(71.8)
IV	Cattle+Buffalo	344(18.0)	537(30.3)	610(31.9)	866(48.9)	519(27.2)	808(45.6)
V	Cattle+Buffalo	516(35.8)	715(44.9)	625(43.4)	802(50.4)	684(47.5)	786(49.4)
VI	Cattle+Buffalo	514 (34.5)	968 (61.3)	520 (34.9)	826 (52.3)	400 (26.9)	838 (53.1)
VII	Cattle+Buffalo	706 (24.9)	911 (43.9)	597 (21.1)	808 (38.9)	740 (26.1)	930 (44.8)

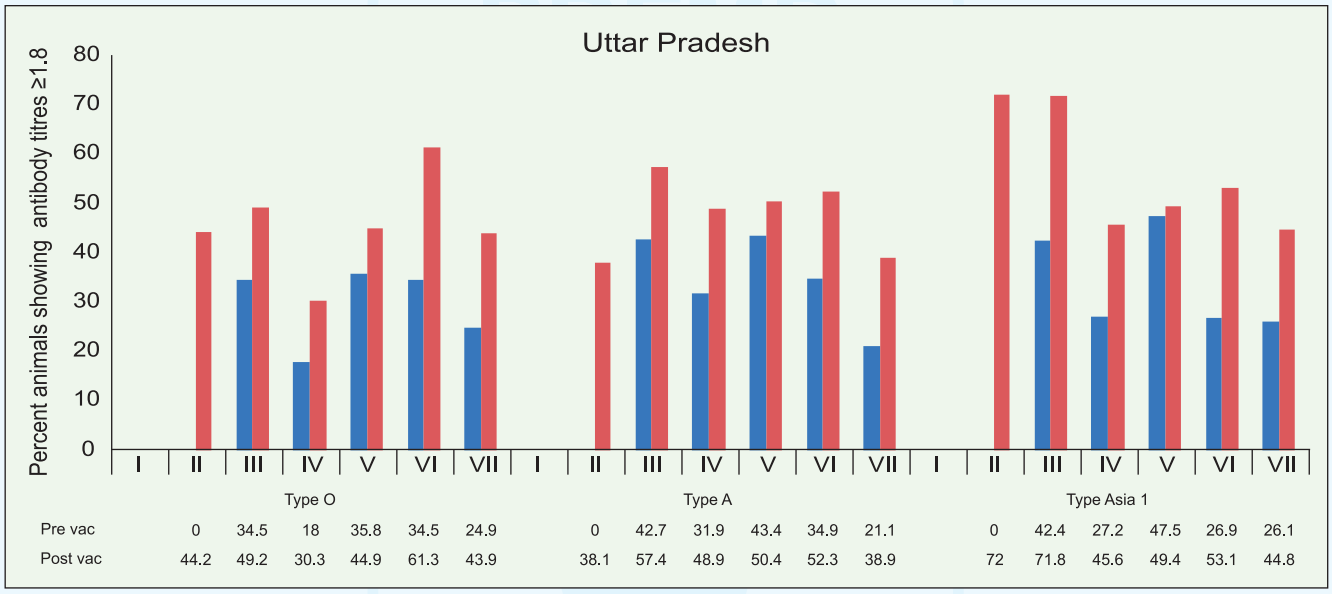


Fig 40: Seroconversion in Uttar Pradesh

having protective antibody titer of 1.8 and above for pre-vac samples was 34.5 for type 'O', 34.9 for type 'A' and 26.9 for type 'Asia-1'. The same for post-vac samples was 61.3 for type 'O', 52.3 for type 'A' and 53.1 for type 'Asia-1'.

- In phase VII at present 2833 pre and 2075 post vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for a pre-vac sample was 23.4 for type 'O', 18.6 for type 'A' and 19.3 for type 'Asia-1'. The same for post-vac samples was 43.9 for type 'O', 38.9 for type 'A' and 44.8 for type 'Asia1'.

10.2 PHASE WISE NUMBER AND PERCENT OF ANIMALS SHOWING ANTIBODY TITER e"1.8 LOG10 AGAINST FMD VIRUS FROM PHASE I TO VIII

Phase I

State	Species	Number & % animals showing titres e"1.8 log_{10} against FMD Virus					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
Andaman Nicobar	Serum samples not available						&
Andhra Pradesh	Cattle+Buff	83 (10.3)	340 (42.5)	43 (5.3)	244 (30.5)	92 (11.5)	340 (42.5)
Delhi	Buffalo	26 (53)	50 (100)	13 (26)	47 (94)	17 (34)	48 (96)
Gujarat	Cattle+Buff	50 (19.1)	116 (44.7)	59 (24.5)	128 (48.7)	42 (16.1)	114 (43.5)
Haryana	Serum samples not available						
Kerala*	Cattle+Buff	158(32.7)	255(51.4)	140(29.0)	236(47.5)	165(34.2)	280(56.4)
Maharashtra	Cattle+Buff	173 (20.5)	456 (59.9)	151(17.9)	437 (57.4)	192 (22.8)	466 (61.2)
Punjab	Cattle+Buff	N.A.	187(25.2)	N.A.	90(11.5)	N.A.	273(49.5)
Tamil Nadu	Cattle+Buff	28(28)	51(51)	29(29)	57(57)	24(24)	54(54)
Uttar Pradesh	Serum samples not available						

* Kerala Phase I, II & IV data is combined.

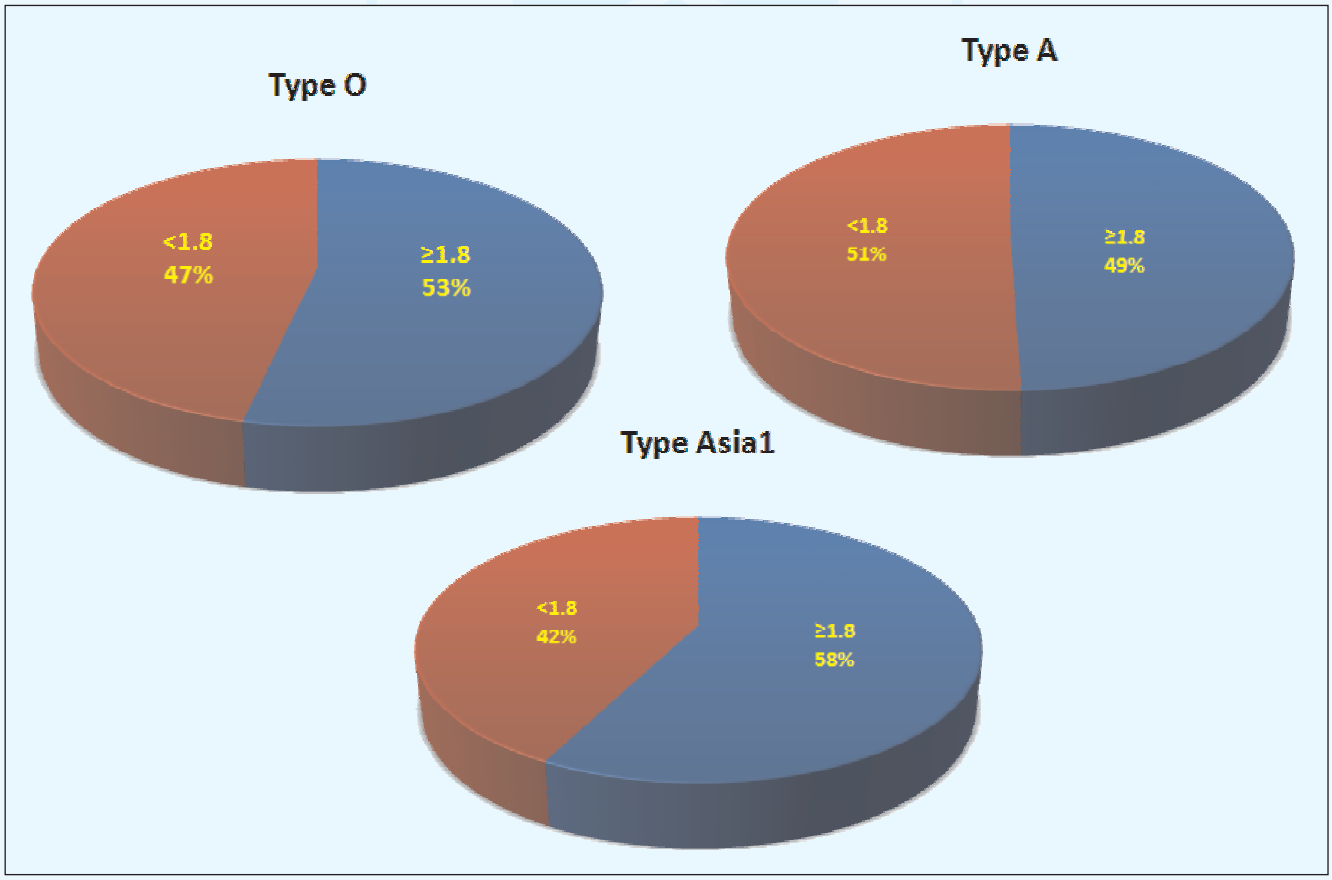


Fig 41: Average post vaccinal seroconversion in Phase I

Phase II

State	Species	Number & % animals showing titres e"1.8 log_{10} against FMD Virus					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
Andaman & Nicobar	Serum samples not available						
Andhra Pradesh	Cattle+Buff	N.A.	434 (54.2)	N.A.	498 (62.3)	N.A.	438 (54.7)
Delhi	Buffalo	22 (91)	23 (96)	12 (40)	15 (62)	23 (95)	22 (86)
Gujarat	Serum samples not available						
Haryana	Cattle+Buff	NA	1065(68.3)	NA	859 (55.1)	NA	831 (53.3)
Kerala*	Cattle+Buff	158(32.7)	255(51.4)	140(29.0)	236(47.5)	165(34.2)	280(56.4)
Maharashtra	Cattle+Buff	N.A.	508 (60.9)	N.A.	490 (58.6)	N.A.	553 (66.2)
Punjab	Cattle+Buff	N.A.	219(43.8)	N.A.	113(20.9)	N.A.	279(58.1)
Tamil Nadu	Cattle+Buff	23(23.0)	63(63.0)	24(24.0)	40(40.0)	18(18.0)	61(61.0)
Uttar Pradesh	Cattle+Buff	0(0)	180(44.2)	0(0)	155(38.1)	0(0)	293(72.0)

* Kerala Phase I, II & IV data is combined.

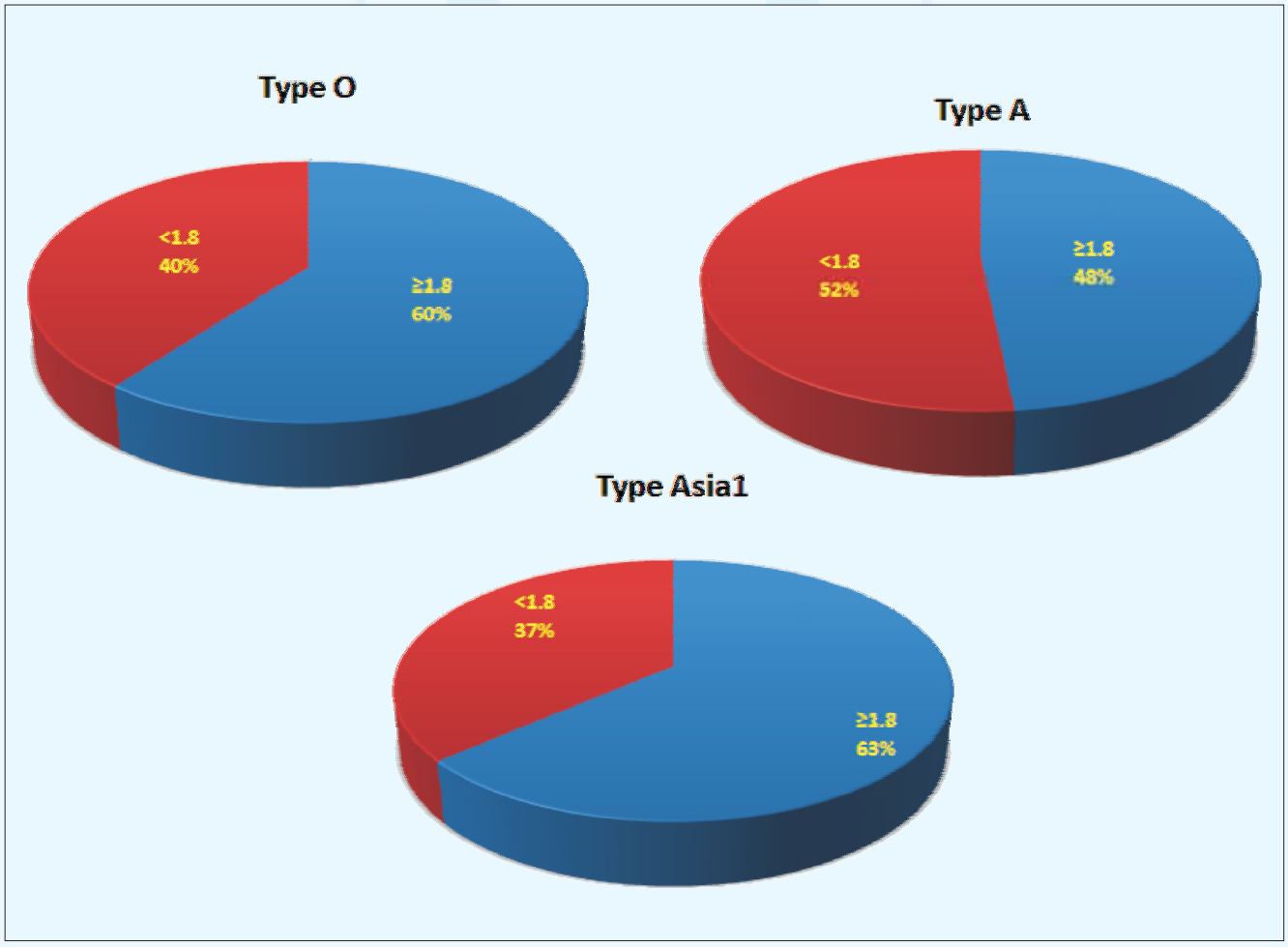


Fig 42: Average post vaccinal seroconversion in Phase II

Phase III

State	Species	Number & % animals showing titres e"1.8 log_{10} against FMD Virus					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
Andaman & Nicobar	Cattle+Buff	40(25.9)	97(60.0)	5(2.8)	37(20.3)	52(34.0)	118(73.6)
Andhra Pradesh	Cattle+Buff	210 (26.2)	286 (35.7)	395 (49.3)	532 (66.5)	306 (38.2)	422 (52.7)
Delhi	Cattle+Buff	47 (94)	49 (98)	30 (60)	40 (80)	43 (86)	46 (92)
Gujarat	Cattle+Buff	123 (27.8)	171 (47.9)	171 (39.2)	268 (58.3)	51 (12.4)	149 (35.4)
Haryana	Cattle+Buff	NA	1146(72.3)	NA	1007(63.6)	NA	1005(63.4)
Kerala	Serum samples not available						
Maharashtra	Cattle+Buff	184 (24.4)	438 (54.8)	351 (46.8)	580 (72.7)	262 (34.7)	534 (66.9)
Punjab	Cattle+Buff	915(84.4)	1175(86.1)	816(75.3)	1007(73.8)	437(40.2)	573(42.0)
Tamil Nadu**	Cattle+Buff	59(32.7)	246(74.5)	61(33.8)	201(60.9)	45(25.0)	216(65.4)
Uttar Pradesh	Cattle+Buff	399(34.5)	780(49.2)	494(42.7)	910(57.4)	490(42.4)	1138(71.8)

** Tamil Nadu Phase III & IV data is combined.

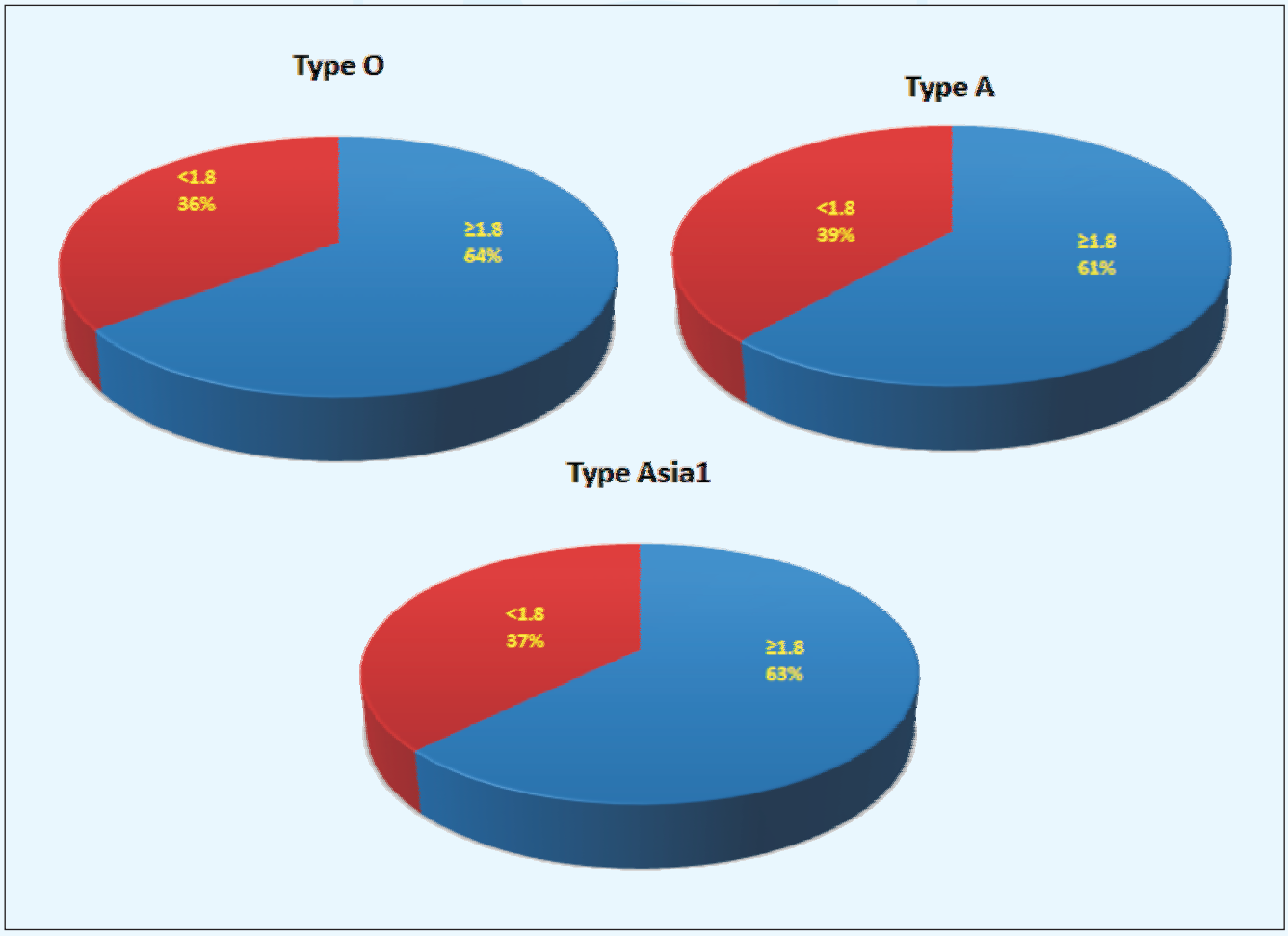


Fig 43: Average post vaccinal seroconversion in Phase III

Phase IV

State	Species	Number & % animals showing titres e"1.8 log_{10} against FMD Virus					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
Andaman &Nicobar	Cattle+Buff	50(33.5)	94(64.6)	50(33.5)	96(65.9)	35(23.4)	101(67.6)
Andhra Pradesh	Cattle+Buff	281 (35.1)	374 (46.8)	465 (58.1)	617 (77.1)	329 (41.1)	518 (64.8)
Delhi	Cattle+Buff	38 (76)	38 (82.6)	14 (28)	40 (86.9)	27 (54)	41 (89.1)
Gujarat	Cattle+Buff	113 (22.7)	277 (60.7)	184 (40.7)	355 (81.2)	73 (14.6)	218 (46.8)
Haryana	Cattle+Buff.	953 (60.1)	1222(78.7)	668 (42.1)	887 (57.1)	844 (53.2)	1170(75.3)
Kerala*	Cattle+Buff	158(32.7)	255(51.4)	140(29.0)	236(47.5)	165(34.2)	280(56.4)
Maharashtra	Cattle+Buff	191 (24.2)	417 (52.3)	517 (65.6)	679 (85.3)	278 (35.2)	509 (63.9)
Punjab	Cattle+Buff	988(76.5)	792 (81.0)	794(61.5)	627 (64.1)	694 (53.8)	356(36.4)
Tamil Nadu**	Cattle+Buff	59(32.7)	246(74.5)	61(33.8)	201(60.9)	45(25.0)	216(65.4)
Uttar Pradesh	Cattle+ Buff	344(18.0)	537(30.3)	610(31.9)	866(48.9)	519(27.2)	808(45.6)

*Kerala Phase I, II & IV data is combined;

**Tamil Nadu Phase III & IV data is combined.

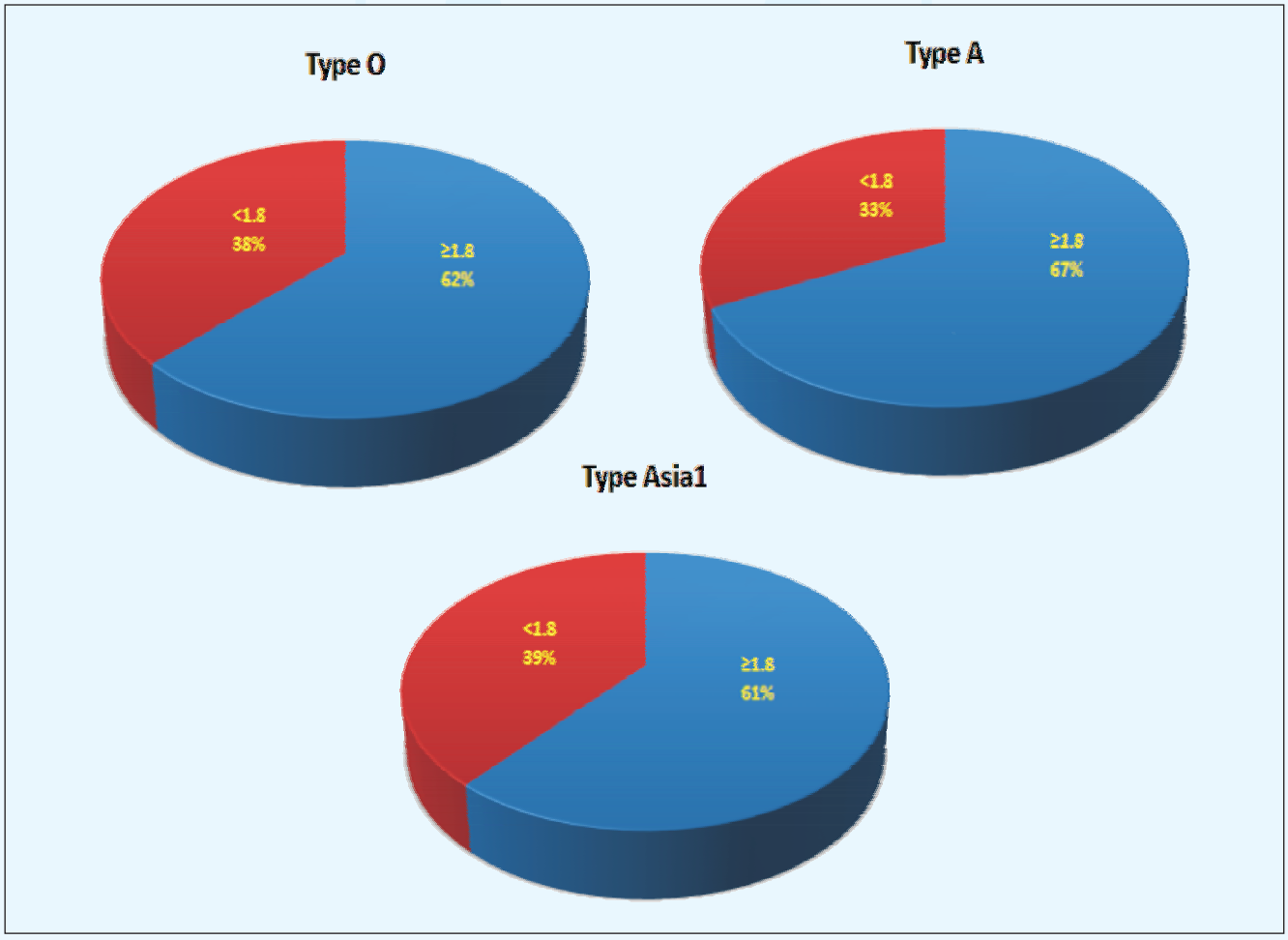


Fig 44: Average post vaccinal seroconversion in Phase IV

Phase V

State	Species	Number & % animals showing titres e"1.8 log_{10} against FMD Virus					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
Andaman & Nicobar	Cattle+Buff	72(57.2)	68(55.8)	62(50.8)	64(52.5)	54(44.3)	62(50.8)
Andhra Pradesh	Cattle+Buff.	247 (30.8)	440 (55)	466 (58.2)	574 (71.8)	343 (42.8)	450 (56.3)
Delhi	Cattle+Buff	26 (59)	47 (88.6)	23 (52.2)	37 (69.8)	32 (72.7)	41 (77.3)
Gujarat	Cattle+ Buff	46 (23.6)	99 (49.0)	126 (66.1)	179 (91.6)	44 (26.5)	92 (51.3)
Haryana	Cattle+Buff.	955 (59.7)	1352(84.5)	813 (50.8)	1274(79.6)	941 (58.8)	1353(84.5)
Kerala	Cattle+Buff	67(23.1)	197(67.9)	52(17.9)	171(58.9)	61(21.0)	211(72.7)
Maharashtra	Cattle+Buff	142 (17.7)	271 (35.1)	353 (44.2)	477 (62.3)	121 (15.0)	245 (31.8)
Punjab	Cattle	477(34.8)	621(54.5)	445(32.8)	630(53.7)	513(38.5)	690(60.1)
Tamil Nadu	Serum samples not available						
Uttar Pradesh	Cattle+ Buff	516(35.8)	715(44.9)	625(43.4)	802(50.4)	684(47.5)	786(49.4)

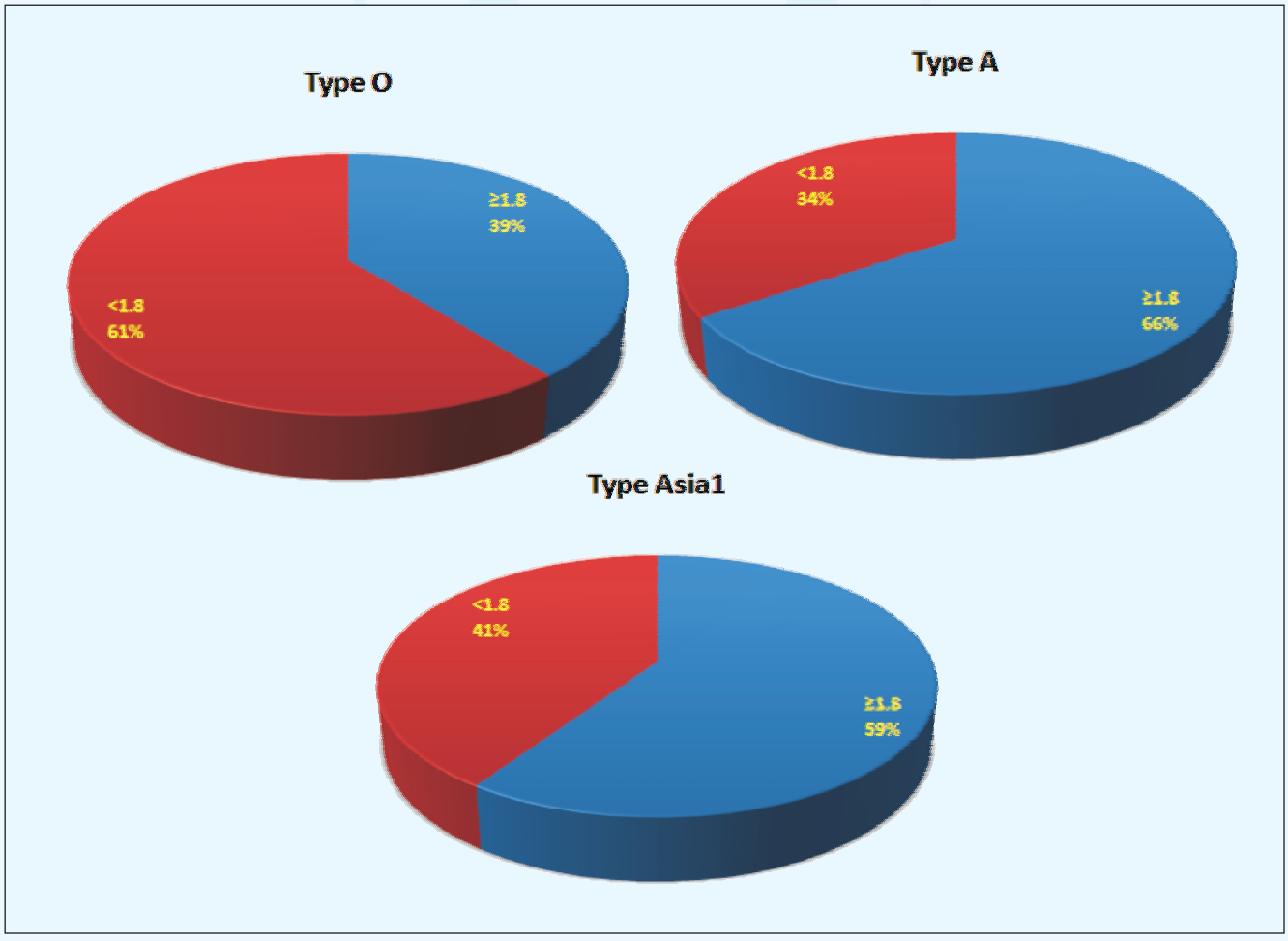


Fig 45: Average post vaccinal seroconversion in Phase V

Phase VI

State	Species	Number & % animals showing titres e"1.8 log_{10} against FMD Virus					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
Andaman & Nicobar		50 (18.5)	80 (29.6)	66 (24.4)	104 (38.4)	28 (10.2)	36 (13.2)
Andhra Pradesh	Cattle+Buff.	275 (34.3)	490 (61.3)	554 (69.2)	690 (86.3)	446 (55.7)	634 (79.3)
Delhi	Cattle+Buff	76 (77.5)	97 (98.9)	60 (61.2)	93 (94.9)	71 (72.4)	97 (98.9)
Gujarat	Cattle+Buff	119 (30.1)	223 (56.4)	249 (63.0)	317(80.2)	195 (49.3)	240 (60.7)
Haryana	Cattle+Buff.	995 (66.5)	1306(87.1)	895 (59.8)	1229(82.0)	844 (56.4)	1118(74.6)
Kerala	Cattle+Buff	49 (20.4)	185(77.1)	41(17.1)	169(70.4)	38(15.8)	171(71.3)
Maharashtra	Cattle+Buff	404 (44.9)	663 (71.4)	622 (69)	853 (91.9)	245 (27.2)	446 (48.1)
Punjab	Cattle+ Buff	653 (43.3)	944 (60.2)	654 (43.3)	921 (58.7)	496 (32.9)	743 (47.4)
Tamil Nadu	Cattle+Buff.	30(18.7)	99(76.1)	31(23.8)	109(83.8)	28(21.5)	103(79.2)
Uttar Pradesh	Cattle+Buff.	514 (34.5)	968 (61.3)	520 (34.9)	826 (52.3)	400 (26.9)	838 (53.1)

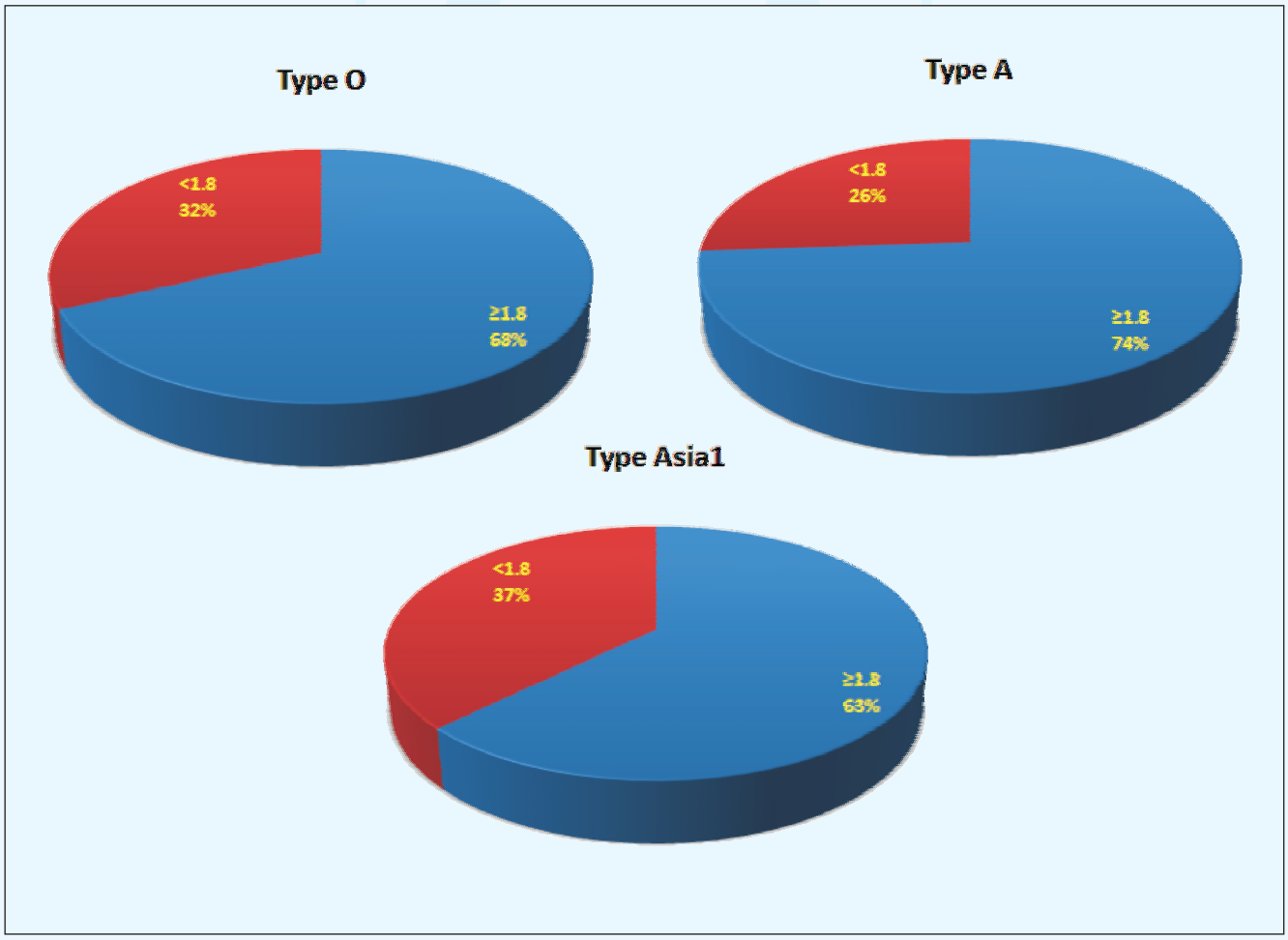


Fig 46: Average post vaccinal seroconversion in Phase VI

Phase VII

State	Species	Number & % animals showing titres e"1.8 log_{10} against FMD Virus					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
Andaman & Nicobar	Cattle+Buff.	112 (42.3)	174 (65.7)	82 (30.9)	110 (41.5)	56 (21.1)	66 (24.9)
Andhra Pradesh	Cattle+Buff.	274 (34.0)	483 (60.3)	349 (44.0)	540 (67.5)	391 (48.8)	518 (64.7)
Delhi	Cattle+Buff.	39(78)	44(88)	33(66)	43(86)	25(50)	41(82)
Gujarat	Cattle+Buff.	434 (54.3)	630 (78.8)	385 (48.1)	559 (69.9)	344 (43.0)	556 (69.5)
Haryana (83.6)	Cattle+Buff.	856(54.8)	1296 (82.3)	1021 (65.3)	1380 (87.6)	888 (56.8)	1 3 1 7
Kerala	Cattle+Buff.	48 (16.0)	208 (69.3)	43 (14.3)	213 (71.0)	52 (17.3)	210 (70.0)
Maharashtra	Cattle+Buff.	446 (44.6)	692 (69.2)	701 (70.1)	893 (89.3)	431 (43.1)	667 (66.7)
Punjab	Cattle+Buff.	413 (36.3)	650 (57.8)	260 (22.8)	472 (42.0)	376 (33.0)	521 (46.4)
Tamil Nadu	Cattle+Buff.	35(11.7)	210(70)	34(11.3)	231(77)	36(12)	226(75.3)
Uttar Pradesh	Cattle+Buff.	706 (24.9)	911 (43.9)	597 (21.1)	808 (38.9)	740 (26.1)	930 (44.8)

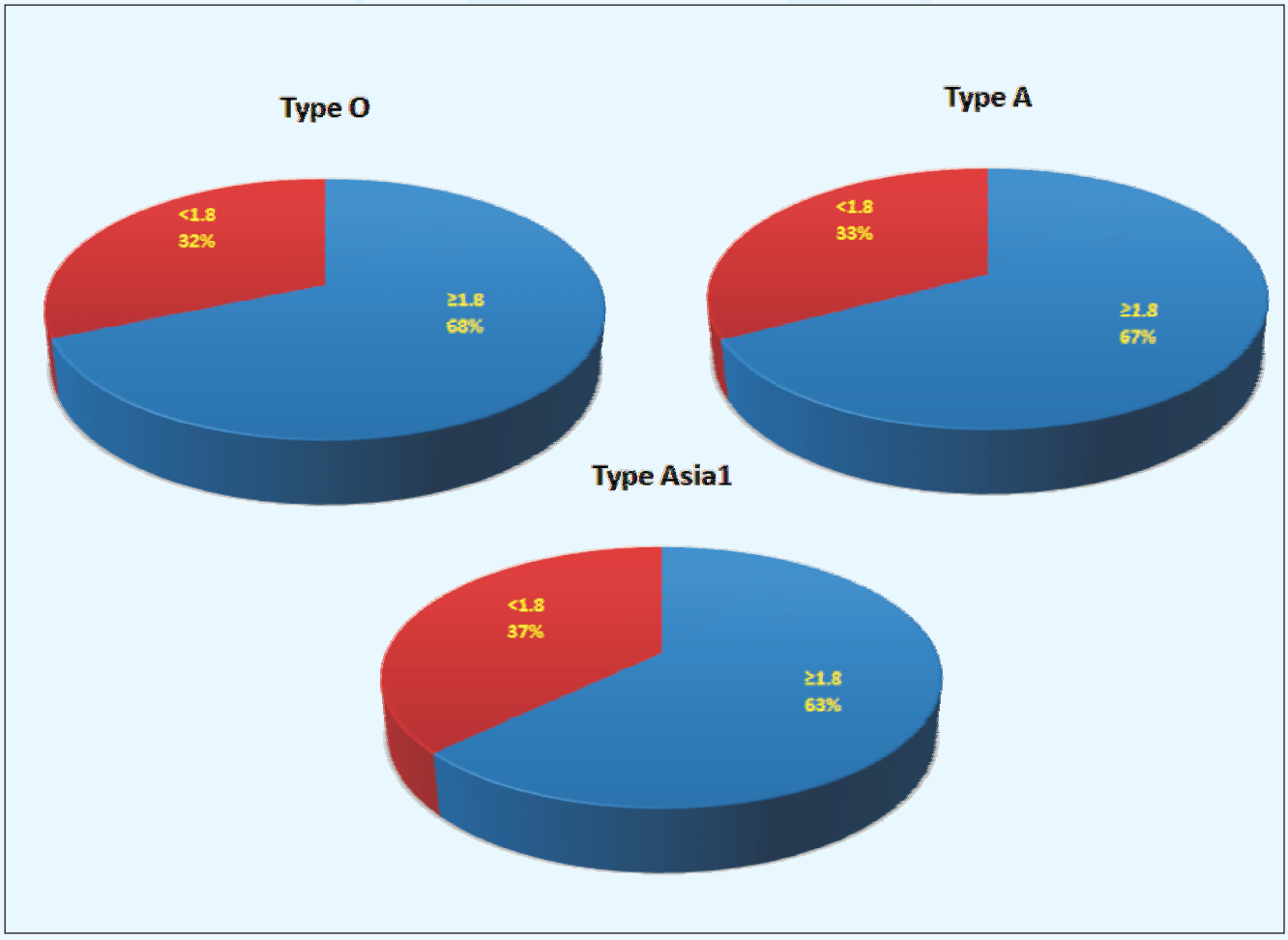


Fig 47: Average post vaccinal seroconversion in Phase VII

Phase VIII

State	Species	Number & % animals showing titres e"1.8 log_{10} against FMD Virus					
		Type O		Type A		Type Asia 1	
		Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
Andaman & Nicobar	Cattle+Buff.	Serum testing is in progress					
Andhra Pradesh	Cattle+Buff.	356 (44.5)	594 (74.0)	415 (51.8)	624 (78.0)	333 (41.6)	527 (65.8)
Delhi	Cattle+Buff.	92 (92)	100 (100)	66 (66)	86 (86)	83 (83)	98 (98)
Gujarat	Cattle+Buff.	191 (23.9)	394 (49.3)	197 (24.6)	357 (44.6)	264 (33.0)	403 (50.4)
Haryana	Cattle+Buff.	949(61.3)	1289 (83.7)	877 (56.6)	992 (64.4)	765 (49.4)	1101 (71.4)
Kerala	Cattle+Buff.	Serum testing is in progress					
Maharashtra	Cattle+Buff.	646 (64.6)	904 (90.4)	574 (57.4)	848 (84.8)	198 (19.8)	452 (45.2)
Punjab	Cattle+Buff.	Serum testing is in progress					
Tamil Nadu	Cattle+Buff.	Serum testing is in progress					
Uttar Pradesh	Cattle+Buff.	Serum testing is in progress					

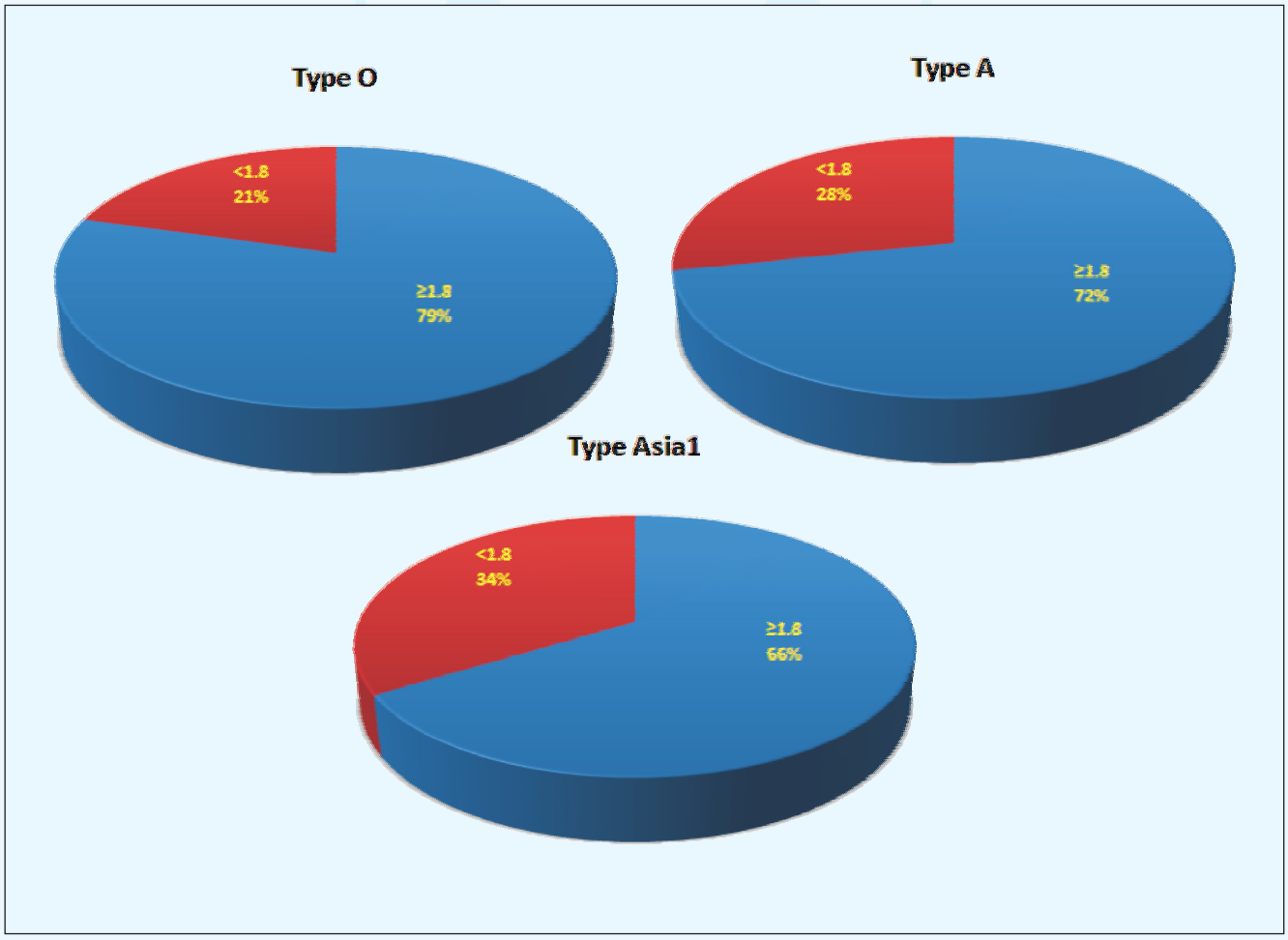


Fig 48: Average post vaccinal seroconversion in Phase VIII

10.2.1 Summary of overall sero conversion in different phases of vaccination against each serotype and impact of vaccine

Table 18. Percent animals showing post vaccinal antibody titers of e"1.8 log_{10} against FMD virus

Phase	Type O		Type A		Type Asia 1	
	Pre-vac	Post-vac	Pre-vac	Post-vac	Pre-vac	Post-vac
I	27.3	53.5	22.0	49.5	23.8	57.6
II	36.7	60.2	23.3	48.4	36.8	63.5
III	43.7	64.3	43.7	61.5	39.1	62.6
IV	41.2	62.3	42.4	67.5	36.2	61.1
V	38.0	39.3	46.3	65.6	40.8	59.4
VI	38.9	67.9	46.6	73.9	36.8	62.6
VII	39.7	68.5	39.4	67.1	35.1	62.8
VIII	57.3	79.5	51.3	71.6	45.4	66.2

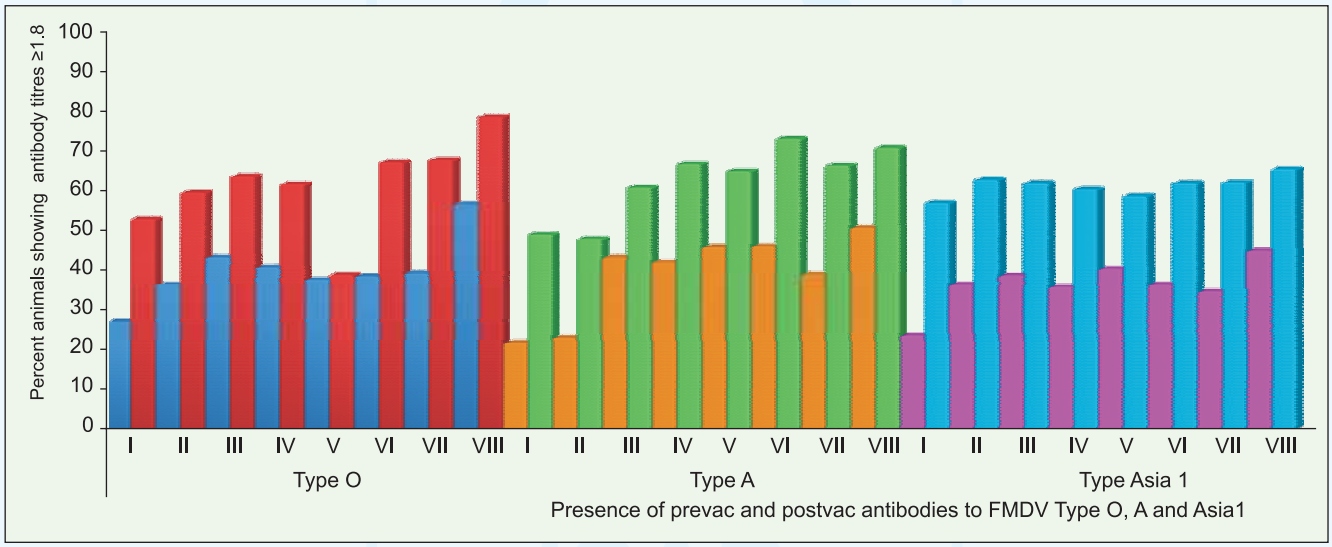


Fig 49: Overall percentage of pre and post vaccinal antibody titers from Phase I to VIII

10.3 SERO-EPIDEMIOLOGY

1.3.1 Testing of Random serum samples

During the period under report, a sum total of 11,560 random serum samples from the states of Jammu & Kashmir, Madhya Pradesh, Manipur, Maharashtra, Orissa, Uttar Pradesh, Tamil Nadu, Uttaranchal, Kerala, Karnataka, Punjab, Himachal Pradesh and West Bengal were subjected to LPB ELISA for determination of antibody level against structural proteins of serotypes O, A and Asia1. Along with the Central Laboratory (Mukteswar), selected Regional Centers and Network Units participated in the exercise. The exercise was aimed at understanding the herd immunity status and disease susceptibility in bovine population of the country by screening randomly collected serum samples. The results of the serum testing are given in Table 19 and Figure 50 and 51.

Table 19. Random serum samples showing protective antibodies against different serotypes of FMD virus (April 2009 to March 2010)

State	Total no. of samples tested	Number and percent of animals showingtitre e"1.8 log_{10} against FMDV		
		Type O	Type A	Type Asia 1
Assam	519	220 (42.4)	175 (33.7)	51 (9.8)
Jammu & Kashmir	1038	349 (33.6)	323 (31.1)	202 (19.5)
Madhya Pradesh	2487	882 (35.5)	849 (34.1)	768 (30.8)
Manipur	538	397 (73.8)	328 (61)	115 (21.4)
Maharashtra	2990	1526 (50.0)	1668 (55.8)	1371 (45.9)
Nagaland	232	140 (60.3)	70 (30.2)	44 (19)
Orissa	1195	490 (41.0)	545 (45.6)	419 (35.1)
Uttar Pradesh	484	175 (36.2)	142 (29.3)	177 (36.6)
Tamil Nadu	168	6 (3.6)	14 (8.3)	23 (13.7)
Uttaranchal	132	10 (7.6)	6 (4.5)	24 (18.2)
Kerala	37	25 (67.6)	22 (59.5)	24 (64.9)
Karnataka	62	8 (12.9)	5 (8.0)	28 (45.2)
Punjab	544	351 (64.5)	197 (36.2)	157 (28.9)
Himachal Pradesh	35	31 (88.6)	29 (82.6)	30 (85.7)
West Bengal	1099	285 (25.9)	229 (20.8)	82 (7.5)
Total	**11560**	**4895 (42.3)**	**4619 (40.0)**	**3523 (30.5)**

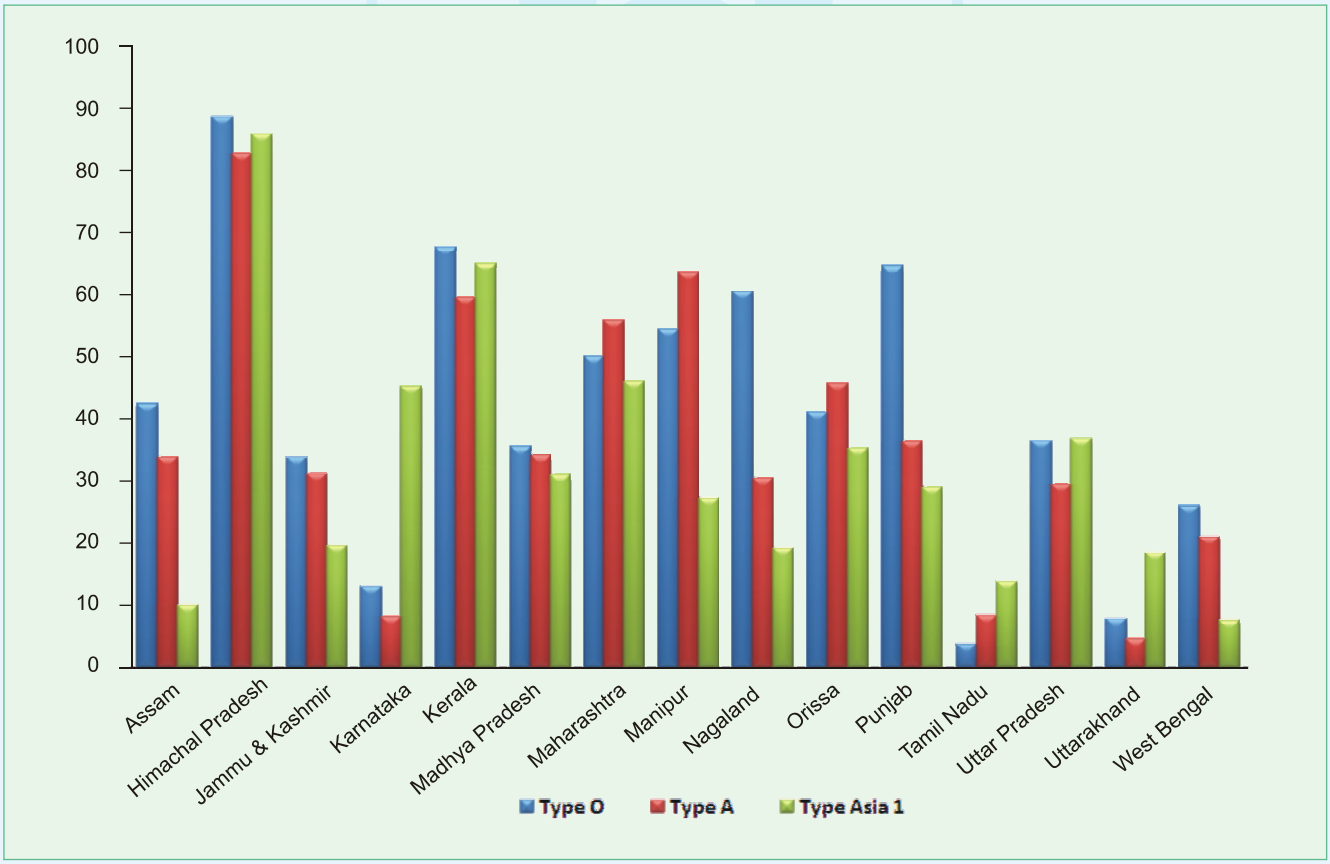


Fig 50: Percent animals showing antibody titre (log_{10}) of e"1.8 in random samples (April 2009 to March 2010).

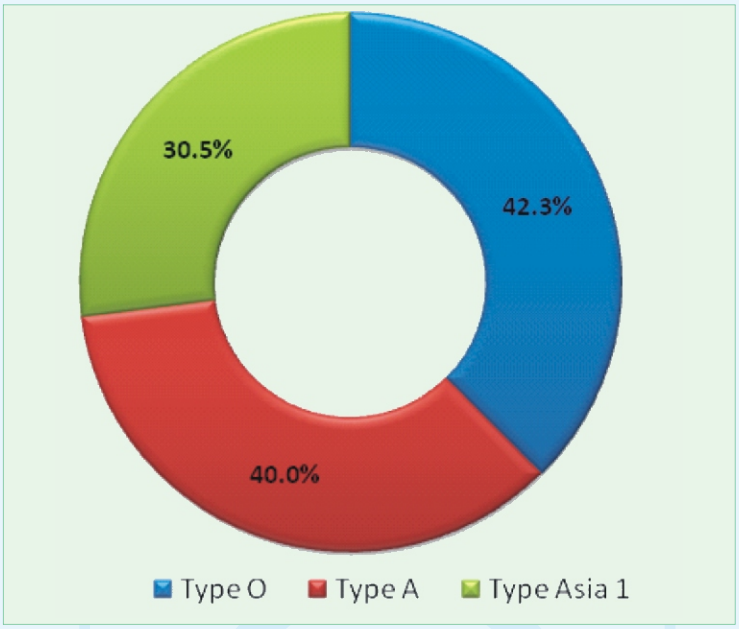


Fig 51: Overall average percent of animals showing antibody titre (log_{10}) of e″1.8 in random samples from different states of the country

11.0 Production, Standardization and Supply of Diagnostic Reagents

For production of reagents, the vaccine virus starins {O (IND R2/75), Asia1 (IND 63/72),) and A (IND 40/00)} were bulk produced in roller culture vessels and purified by density gradient centrifugation. Antibodies against purified virus was raised and titrated against homologous as well as heterologous virus. Freeze dried and standardized serum antibodies and known positive antigen (killed) of all three serotypes were supplied to all the centers and network units for use in virus typing ELISA and LPB-ELISA. Viruse serotyping Kits for testing 7000 clinical materials were supplied to 14 centers/network units. LPB-ELISA Kits for testing 80,000 serum samples supplied to the 15 centers/ network units and other government agencies and industry to ensure uniformity in test results.

r3AB3 DIVA Kit for FMD to test a total number of 75,000 serum samples was produced and reagents to test 54,485 samples have been supplied to the Centers and Network units for testing random serum samples for estimation of FMD sero-prevalence, for capacity build up and internal cross validation of the kit.

All the three diagnostic kits were also supplied to FMD vaccine industry on demand.

12.0 International Collaboration

12.1 OIE/FAO Global FMD vaccine matching exercises

2009 Global Inter-Laboratory Comparative Testing Exercise for FMD Vaccine Matching

The Project Directorate on FMD, a Regional Reference Laboratory within the OIE/FAO Network of FMD Reference Laboratories, has participated in 2008 Global Inter-Laboratory Comparative Testing Exercise for FMD vaccine matching between members and observers within the OIE/FAO Network of FMD Reference Laboratories. This was organized by the European Community Reference Laboratory for FMD and the OIE and FAO-World Reference Laboratory (WRL) for FMD at the Pirbright Laboratory of the Institute for Animal Health (IAH-P) as per the discussion during Network Meeting held in Botswana in June 2007. This is the first step taken by the OIE/FAO Network in working towards establishing equivalence in the vaccine matching methods that are done in different laboratories.

Purpose

Safeguarding the international trade of animals or their derived products requires an efficient global surveillance for foot-and-mouth disease (FMD) including constantly updated information on antigenic and genetic characteristics of FMD virus (FMDV) involved in current outbreaks. The exchange of FMDV isolates and data relating to them is also desirable for the development and selection of vaccines and other tools for surveillance and control of FMD, as well as for harmonization of such approaches.

Objectives

1. To gather, generate, analyze and make available laboratory information on the global occurrence and spread of FMD and on the characterization of FMD viruses.
2. To provide recommendations on vaccine strain selection for implementation of control schemes and for vaccine antigen reserves.
3. To offer expertise to OIE, FAO and Member Countries to assist in the control of FMD.
4. To harmonize approaches to the characterization of FMD viruses.
5. To increase the competence of reference laboratories, to identify constraints to the functioning of the network and to propose solutions.

Aim

To evaluate whether similar vaccine matching results are obtained by laboratories using their own methods but with the same FMD vaccine virus, bovine vaccine sera (BVS) and field isolates.

Benefits

1. Vaccine matching data produced in different labs is comparable and can therefore be integrated to produce a more reliable and complete set of recommendations on vaccine selection for different settings.
2. There will be reduced requirement for international exchanges in the future.

Material received

1. 1:4 pre-diluted pooled BVS (heat inactivated) against A Iraq 24/64
2. BEI inactivated vaccine virus A Iraq 24/64
3. 9 coded BEI inactivated FMD serotype A field isolates (ICLT 19-27).

Methodology

1. One way antigenic relationship was calculated using coating and tracing serum against both IND 17/77 and Iraq 24/64. Reference serum was used in one plate per test per day.
2. Optimized dilution of coating (1:5000) and tracing (1:4000) serum against IND 17/77 and predetermined dilution against Iraq 24/64 (as provided by (WRL), was used to

Table 20. Day-to-day variation in Iraq 24/64 bovine vaccinate serum (BVS) titre (Log_{10} SN_{50}) against serotype A FMD virus field isolates in LPB-ELISA

VIRUS	IND17/77 COATING AND TRACING SERUM		
	DAY 1	DAY 2	DAY 3
IND17/77	2.88	2.78	2.60
IND490/97	1.88	1.78	1.75
IND40/00	1.80	1.55	1.50
IRAQ24/64	2.70	2.70	2.80
ICLT-19	2.70	2.85	2.87
ICLT-20	2.63	2.65	2.85
ICLT-21	1.80	1.93	1.73
ICLT-22	2.10	2.05	1.80
ICLT-23	1.80	1.55	1.53
ICLT-24	1.80	1.55	1.20
ICLT-25	1.80	1.94	1.20
ICLT-26	1.80	1.33	1.20
ICLT-27	1.98	1.83	1.35
VIRUS	**IRAQ24/64 COATING ANDTRACING SERUM**		
	DAY 1	**DAY 2**	
IND17/77	2.75	2.48	
IND490/97	1.65	1.67	
IND40/00	1.20	1.20	
IRAQ24/64	2.55	2.15	
ICLT-19	2.52	2.25	
ICLT-20	2.48	2.16	
ICLT-21	1.20	1.20	
ICLT-22	1.50	1.42	
ICLT-23	1.20	1.20	
ICLT-24	1.72	1.48	
ICLT-25	1.75	1.54	
ICLT-26	1.20	1.20	
ICLT-27	1.35	1.28	

Table 21. One-way antigenic relationship (average of different test days) of serotype A FMD virus field isolates in relation to Iraq 24/64 using bovine vaccinate serum

VIRUSES	r-VALUE USING IND17/77 COATING AND TRACING SERUM			
	DAY 1	DAY 2	DAY 3	AVERAGE
IND17/77	1.51	1.20	0.63	>1.00
IND490/97	0.15	0.12	0.09	0.12
IND40/00	0.13	0.07	0.05	0.08
IRAQ24/64	1.00	1.00	1.00	1.00
ICLT-19	1.00	1.41	1.17	1.19
ICLT-20	0.85	0.89	1.12	0.95
ICLT-21	0.13	0.17	0.09	0.13
ICLT-22	0.25	0.22	0.10	0.19
ICLT-23	0.13	0.07	0.05	0.08
ICLT-24	0.13	0.07	0.03	0.08
ICLT-25	0.13	0.17	0.03	0.11
ICLT-26	0.13	0.04	0.03	0.07
ICLT-27	0.19	0.13	0.04	0.12
VIRUSES	**r-VALUE USING IND17/77 COATING AND TRACING SERUM**			
	DAY 1	**DAY 2**	**AVERAGE**	
IND17/77	>1.0	>1.0	>1.0	
IND490/97	0.13	0.33	0.23	
IND40/00	0.04	0.11	0.08	
IRAQ24/64	1.00	1.00	1.00	
ICLT-19	0.93	1.26	1.10	
ICLT-20	0.84	1.02	0.93	
ICLT-21	0.04	0.11	0.08	
ICLT-22	0.09	0.19	0.14	
ICLT-23	0.04	0.11	0.08	
ICLT-24	0.15	0.21	0.18	
ICLT-25	0.16	0.25	0.20	
ICLT-26	0.04	0.11	0.08	
ICLT-27	0.06	0.13	0.10	

Table 22. One-way antigenic relationship of serotype A FMD virus field isolates in relation to individual and pooled bovine vaccinates serum against Iraq 24/64

	BVS06104	BVS0610	BVS0637	BVS0645	BVS0679	POOLED
	r1	r1	r1	r1	r1	
IND17/77	0.91	0	0.52	0.47	0.35	1.12
IND40/00	0.16	0	0.18	0.15	0.11	0.11
IRAQ24/64	1.00	0	1.00	1.00	1.00	1.00
A61	0.19	0	0.19	0.12	0.11	0.14
A62	0.25	0	0.25	0.19	0.08	0.35
A63	0.09	0	0.07	0.06	0.04	0.09
A65	0.71	0	0.63	0.52	0.29	0.71
A66	0.40	0	0.25	0.20	0.12	0.13

determine the one way antigenic relationship of type A field isolates with Iraq 24/64.

3. Antigens of field isolates were titrated by making dilutions ranging from 1:2 to 1:14 as determined by checker board titration. Indian vaccine strains of type A were also included in this study. At dilutions where an OD of 1.0 obtained was used in LPB-ELISA.
4. The dilution of anti-guinea-pig conjugate (1:2000) was also determined by checker board titration.

The one way antigenic relationship (r value) was calculated as follows.

$$\text{r value} = \frac{\text{Serum titer with heterologous virus}}{\text{Serum titer with homologous virus}} \times 100$$

Conclusion

- ICLT 19 and 20 viruses are antigenically related (re"0.40) to A Iraq 24/64 and IND 17/77.
- ICLT 21 to 27 viruses are antigenically divergent (rd"0.40) from A Iraq 24/64 and IND 17/77.
- No variation in r value was observed for the isolates between the tests where A Iraq 24/64 and IND 17/77 coating and tracing serum used.
- Due to paucity of antigen the field isolates could not be tested using BVS against IND 17/77.

12.2 Collaboration with USDA-ARS (Under GFRA)

A project entitled "Effective Molecular Vaccines against Foot-and-Mouth Disease" was initiated in collaboration with USDA-ARS under Global FMD Research Alliance. The overall objective of this project is to identify molecular determinants responsible for antigenic variation of Indian FMDV field strains. This project will establish a collaborative research project between the USDA ARS and two Institutes of Indian Council of Agricultural Research (ICAR). The Indian Veterinary Research Institute (IVRI) at its Bangalore campus will be responsible for FMD vaccine research and production, and the PDFMD, Mukteswar will be responsible for detailed antigenic and genetic characterization of FMD field virus isolates, surveillance, and monitoring and vaccine strain selection for FMD control programs in India. At the end of the three-year project, it is expected that newer and effective molecular vaccines will be available for use in endemic countries including India.

13.0 FMD Vaccine Matching Exercises in Collaboration with Indian Vaccine Industry

Though all Indian vaccine manufacturers were contacted for this collaborative study to develop partenership in R & D, one of the vaccine manufacturer (IIL) came forward to participate the programme.

Selection of new field isolates as alternate candidate vaccine strains

FMD situation due to type O in India is not different from that found in the Middle-East and as mentioned earlier PanAsia is the predominant genotype causing outbreaks. Keeping in mind the changing scenario where there has been upsurge in type O outbreaks due to "IND2001" strains and co-circulation of PanAsia genotype, 7 FMDV type O strains from a pool of nearly 150 previously characterized FMDV type O field isolates were short listed for evaluation as alternate candidate vaccine strains based on the criteria of infectivity titer in cell culture and genetic relationship with presently circulating groups (Fig 52 and 53). Experimental monovalent vaccine batches were prepared by the industry partener and BVS gainst each of the 7 virus was raised. This work was undertaken with the objective to have a panel of strains that can be used as vaccine strain when the need arises and also to examine whether we have a better strain in the nature/ repository compared to the present vaccine strain, IND R2/75.

A total of 18 FMDV serotype O field isolates from different parts of country were used to study antigenic relationship by micro-neutralization test. All the field isolates selected showed an r-value of >0.3 with the currently used vaccine strain (IND R2/75) (Fig 54), while it was 94.4% (17 out of 18) for IND 320/07(Fig 58). In case of IND 271/01 (Fig 56) and 120/02 (Fig57), 88.35% (14 out of 17) and 88.24% (15 out of 17) of the isolates, respectively showed an r value of >0.3. The other candidate vaccine strains (IND489/97 (Fig 55), IND331/07 (Fig 59) and IND408/07 (Fig 60)) showed poor antigenic coverage. The study revealed that the current vaccine strain, IND R2/75 continues to provide good antigenic coverage even in the current situation followed by IND 320/07.

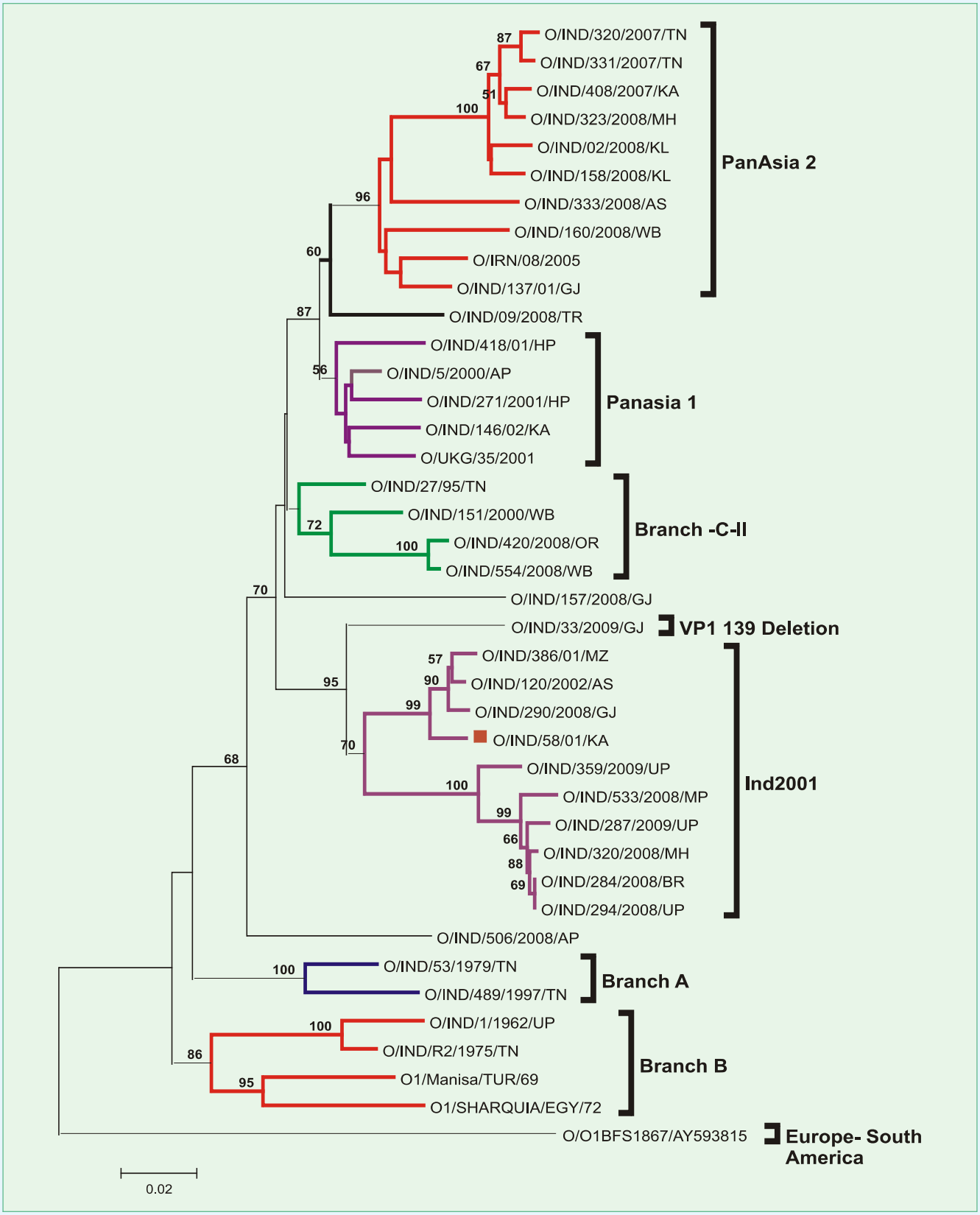


Fig 52. Neighbour-joining tree depicting phylogenetic relationship of type O field isolates with vaccine candidates and currently used vaccine strain(INDR2/72) at 1D region. Candidate vaccine strains are underlined

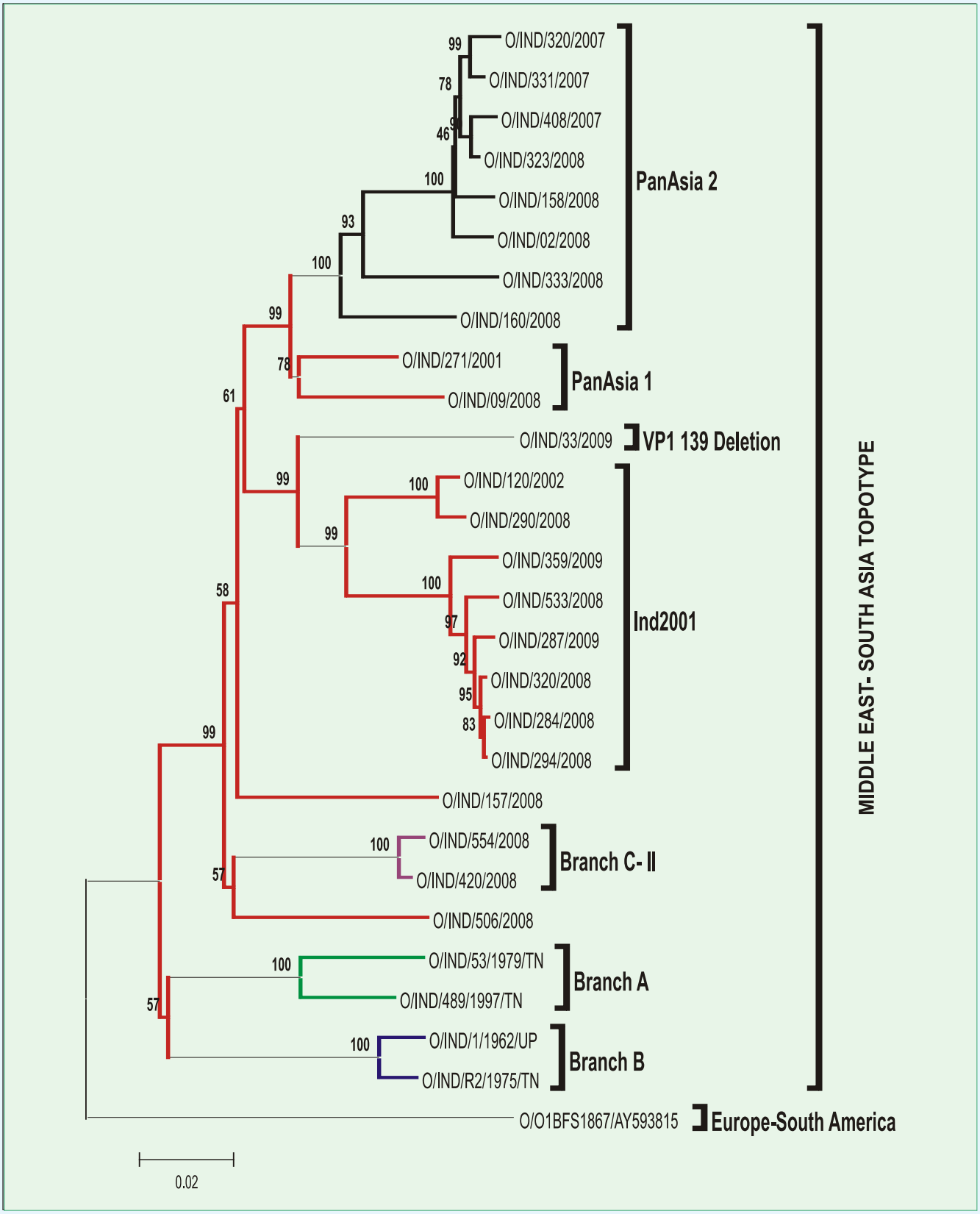


Fig 53. Neighbour-joining tree depicting phylogenetic relationship of type O field isolates with vaccine candidates and currently used vaccine strain(INDR2/72) at P1 region. Candidate vaccine strains are underlined.

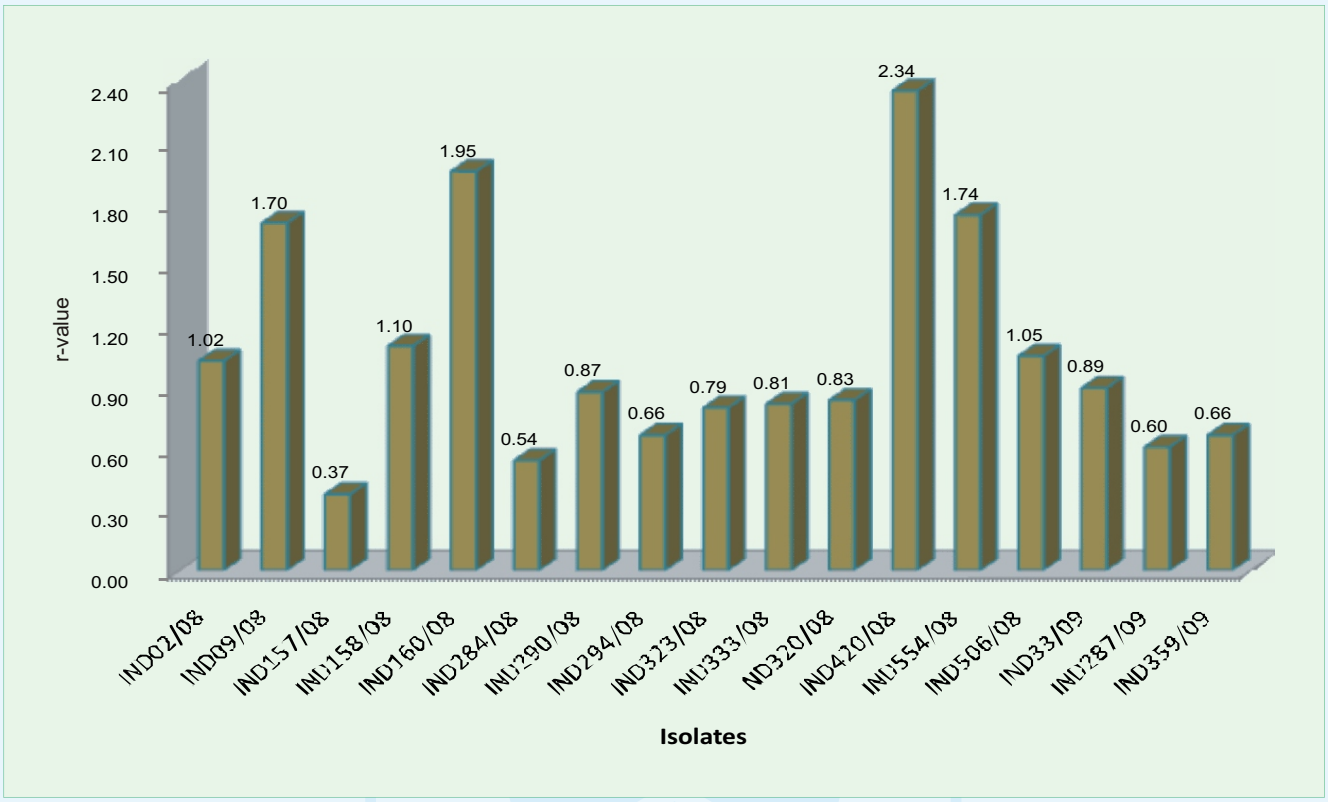


Fig 54. Antigenic relationship of type O isolates with the vaccine strain IND INDR2/1975/TN (Branch B) BVS raised against INDR2/75 was used in 2DMNT

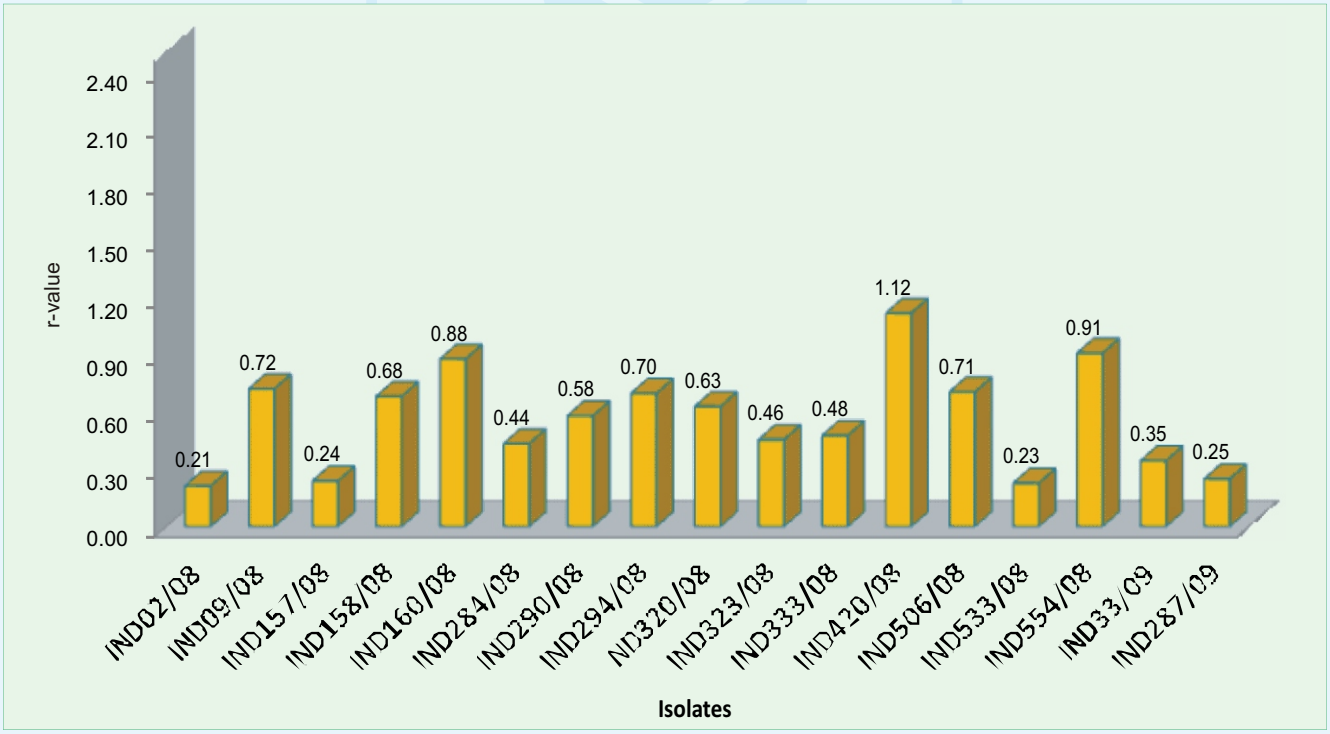


Fig 55. Antigenic relationship of type O isolates with candidate vaccine strain IND 489/1997/TN (Branch A) BVS raised against IND 489/1997/TN was used in 2DMNT

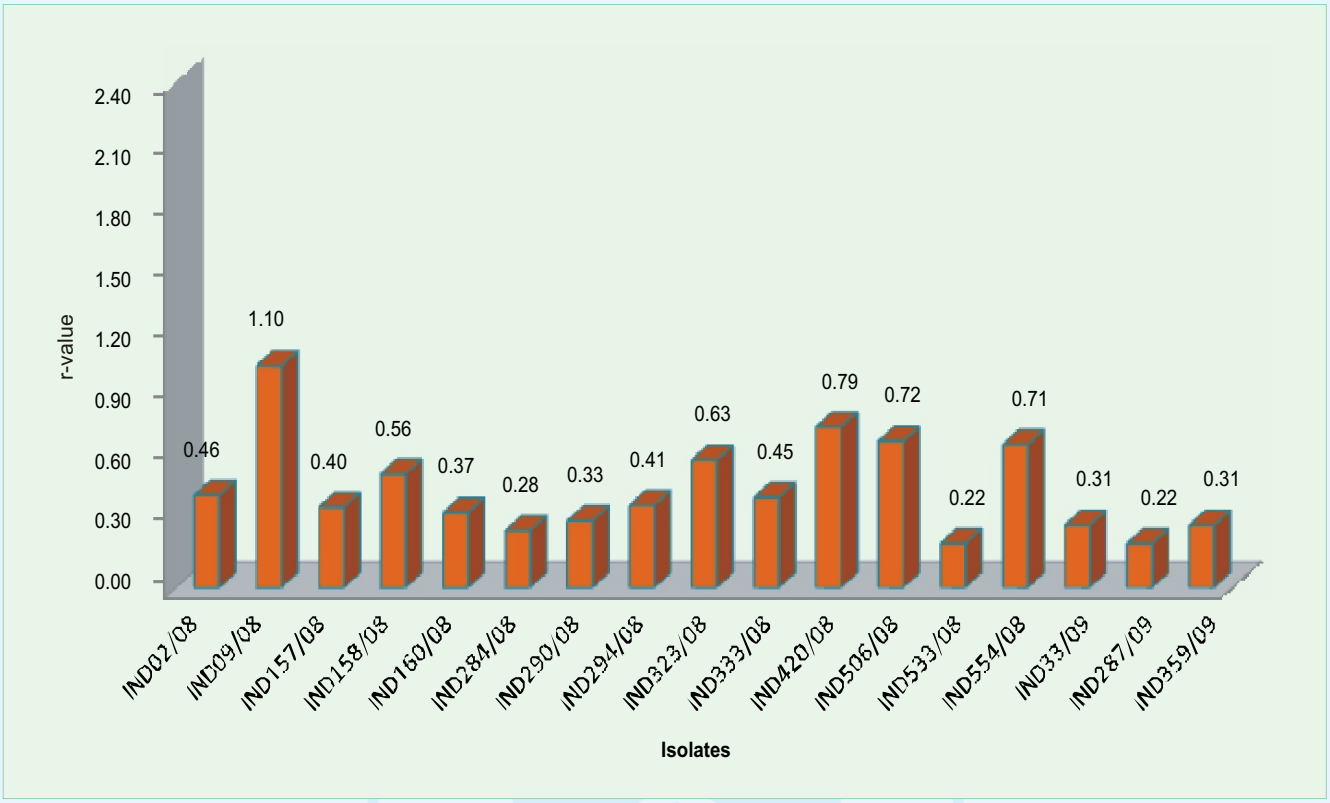


Fig 56. Antigenic relationship of type O isolates with candidate vaccine strain IND 271/2001/HP (PanAsia1) BVS raised against IND 271/2001/HP was used in 2DMNT

Fig 57. Antigenic relationship of type O isolates with candidate vaccine strain IND 120/2002/AS (Ind2001) BVS raised against IND 120/2002/AS was used in 2DMNT

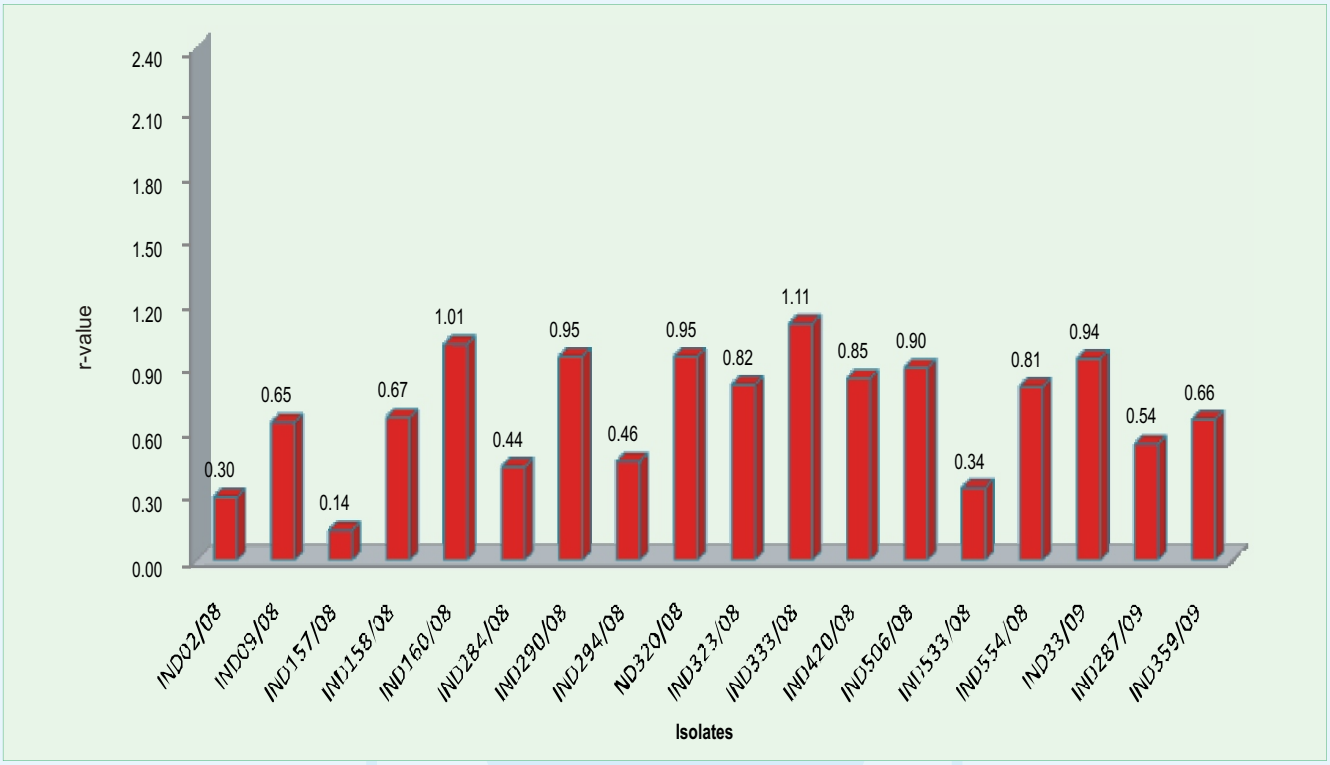


Fig 58. Antigenic relationship of type O isolates with candidate vaccine strain IND 320/2007/TN (PanAsia2) BVS raised against IND 320/2007/TN was used in 2DMNT

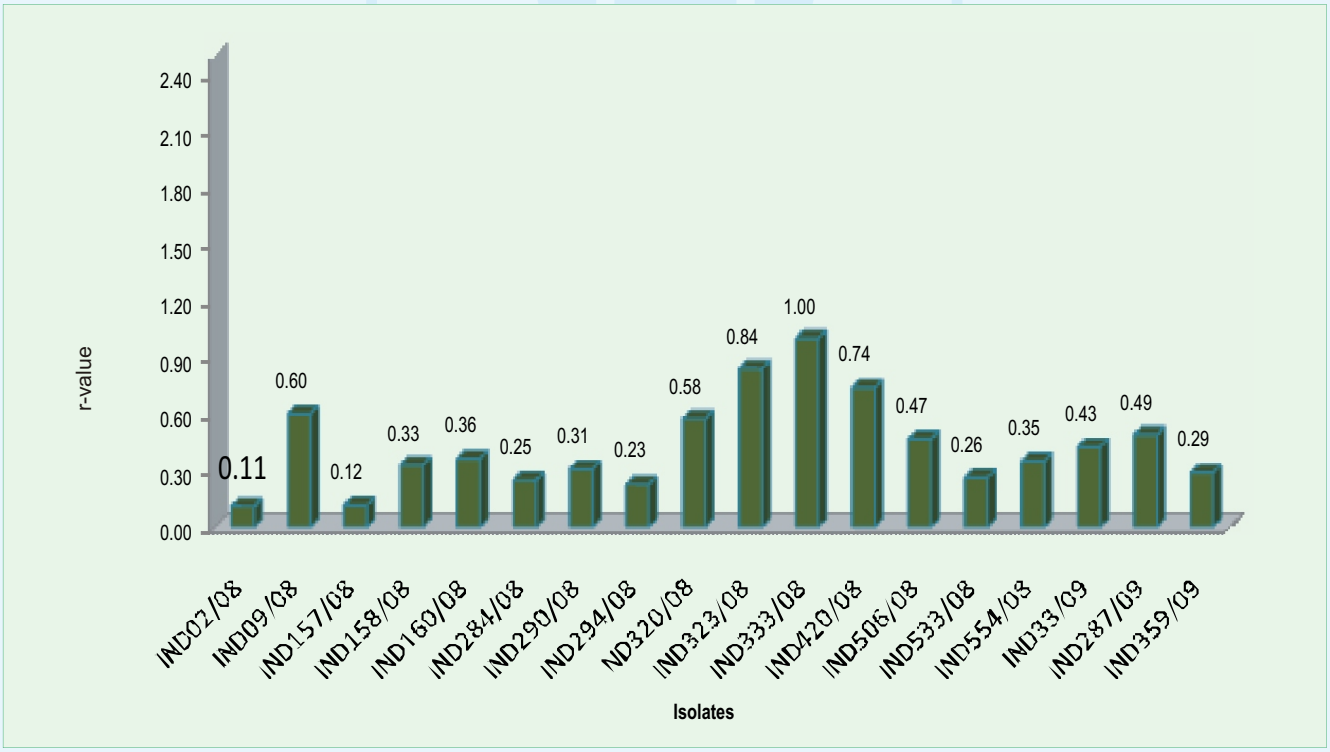


Fig 59. Antigenic relationship of type O isolates with candidate vaccine strain IND 331/2007/TN (PanAsia2) BVS raised against IND 331/2007/TN was used in 2DMNT

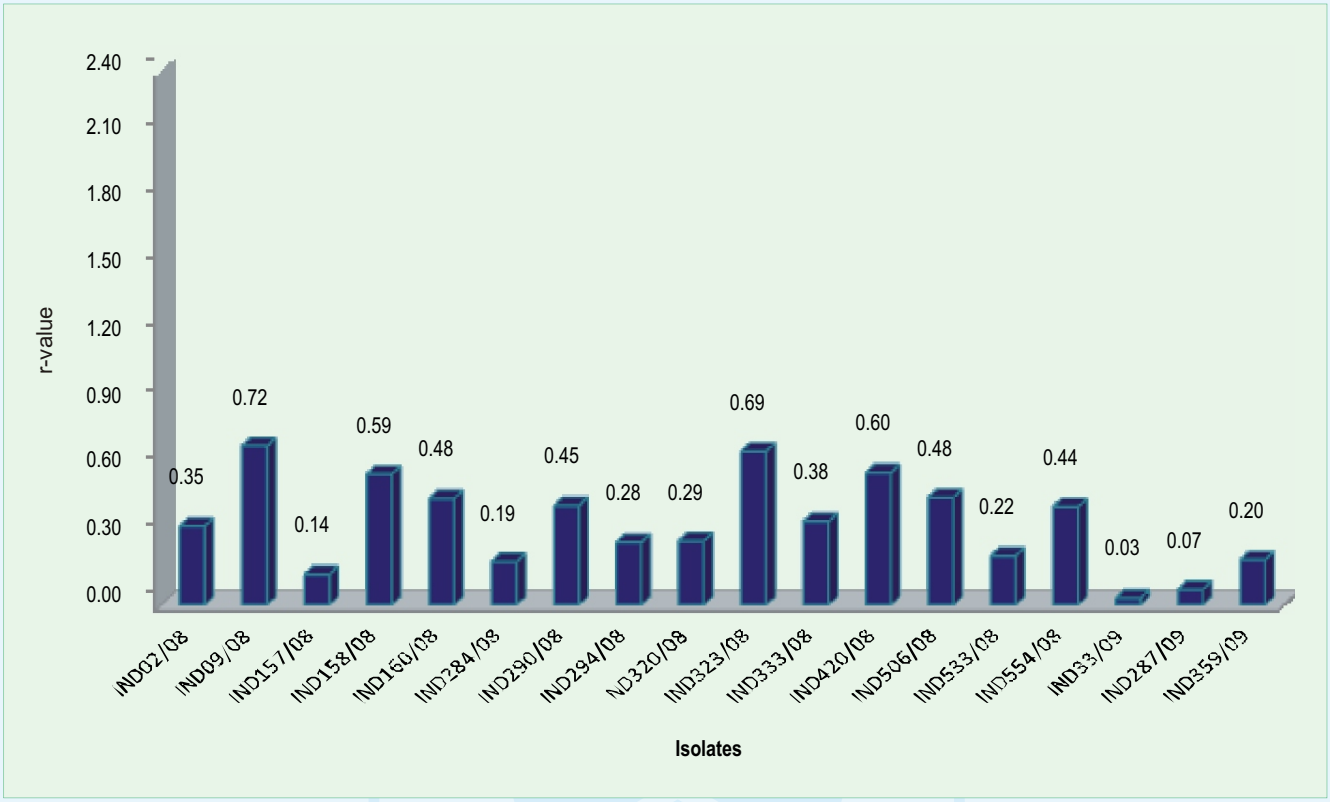


Fig 60. Antigenic relationship of type O isolates with candidate vaccine strain IND 408/1997/KA (PanAsia2) BVS raised against IND 408/1997/KA was used in 2DMNT

14.0 Epidemiological Investigations

14.1 Epidemiological Investigation of FMD in Mithuns in Arunachal Pradesh

Scientists (Dr. A. Sanyal & Dr. S. Pawar) were deputed from the Project Directorate on FMD for epidemiological investigation of FMD in Mithun in the affected districts of Arunachal Pradesh. Dr. Krishna Sharma from the Regional FMD Center, Guwahati was also a member of the team.

Dr. Sanyal, Scientist from PDFMD briefed the members about the purpose of the visit and discussed the importance of controlling FMD by regular vaccination. He informed the officers of the Arunachal Pradesh Government that Project Directorate on FMD under ICAR is responsible for epidemiology and surveillance of FMD in the country and based on the information generated strategies are developed for controlling FMD in the country by the Government of India. Dr. Sanyal informed the officers that scientists of FMD Network Unit, Nirzuli have reported outbreaks of FMD in Arunachal Pradesh since (last) a few months and in recent past there were many cases of FMD in Mithun and these have been already diagnosed and necessary recommendations were made through the FMD Network Unit at Nirjuli. Dr. Taku informed that in the year 2006-07, 100% vaccination was made in Assam-Arunachal Pradesh border as a result there was no disease during this period. But in the years 2007-08 and 2008-09, no vaccination was done at the border of Assam-Arunachal Pradesh and the cases of FMD occurred. Dr. H Tama, DVO, Papum Pare, informed that more than 400 Mithuns died in the affected area and FMD vaccination in the area has been very poor. He stated that it is very difficult to administer vaccine in these semi-domesticated/wild animals. Dr. Sanyal enquired that is there any organized Mithun farm where disease was noticed. The concerned DVO told that the state is having Government Mithun Farm at Sagalee where regular vaccination is done and there is no case of FMD. Dr. Sanyal asked if the disease was noticed in cattle and pigs in those villages and if there is any movement of animals into the district. Dr. H Tama, DVO, Papum Pare, informed that in the affected villages disease was noticed initially in cattle and pigs, and on regular basis animals are brought from Assam for meat purpose through Kimin check post which shares the boundary with Lakhimpur district of Assam. After a lengthy discussion it was decided that the officers from the state along with Scientists form PDFMD will visit the affected villages for detailed investigation and to assess the situation.

In the afternoon Scientists form PDFMD along with state government officers went in and around Itanagar to investigate the FMD situation there as well as in neighbouring areas. They went to District Veterinary hospital near Nirjuli. The Doctor informed that a case of suspected FMD in cattle was observed on 26th October 2009. The team went to the concerned farmer and found that all the three cattle he is maintaining were affected with FMD. Clinical materials were collected from these animals for laboratory investigation. To avoid further spread of the disease, the DVO was advised to carry out immediate vaccination in the area.

On 28th morning the team left for detailed epidemiological investigation in Papaum Pare district. Details of FMD susceptible animals in the district are shown in Table 1. First case was noticed in Khil village. The concerned Veterinary Surgeon Dr. Tow Tagu informed that sporadic

cases were noticed there since last one and half months in Mithun, cattle and pigs. He told that in three villages under Taru Panchayat namely, Taru, Sero and Mowa, more than 30 deaths were noticed in Mithuns. Mithuns in the villages namely, Laptap, Tashi and Peach under Pabar Panchayat were also affected with FMD. He informed that Mithuns in this area were never vaccinated. The team examined many Mithuns in these villages (in forest) and on clinical examination (about 30 Mithuns), severe maggot infestation and secondary infection following FMD in nostrils, muzzle and inter-digital spaces of feet was observed. Animals were not in a position to breath properly due to blockade in the nostrils. Photographs of ailing Mithuns with severe secondary complications are appended for reference. The owners (Mrs. Teli Yaga, Mr. Tama Taya, Mr. Jiban Debnath, Mr. Tana Tezi, Mr. Gambura, Mr. Tana Nido, Mr. Tana Takar, Mr. Chetu etc.) informed that the disease was noticed in the month of September 2009. The animals live in the jungle and inspite of the disease they are unable to treat them because it is difficult to restrain them. The affected Mithuns remain in jungle for several days and as they cannot eat anything, they become weak and die. Many ailing Mithuns in the jungle were treated by the team for secondary complications. The owners were advised regarding how to take care of the affected animals. They were asked to clean the oral wounds frequently and administer medicines to the affected animals with the help of local veterinarian. The team of PDFMD conducted FMD awareness meeting in all the villages visited for the Mithun owners and explained the animal owners about the importance of vaccinating Mithuns against FMD.

After visiting several FMD affected villages the team went to the Government Mithun Breeding Farm at Sagalee. There was no case of FMD in all the 40 Mithuns maintained in this farm. These animals are vaccinated regularly at 6 months interval.

On 29th morning a meeting was arranged with the Director, Animal Husbandry after he was back from Guwahati. He was briefed with the real situation and advised to take necessary measures in controlling FMD. He narrated the problems of FMD vaccination in Mithuns like accessibility, semi domestic nature of the animals, and superstition of the animal owners that quality of meat deteriorates after FMD vaccination. He stressed that additional assistance should come from the ICAR/Central Government for strengthening interstate and international check posts and the state share should be reduced from 25 percent to at least 10 percent. They want FMD vaccine should be made available free of cost to the state, so also vehicles for each DVO and free medicines. Dr. Sanyal informed that the Project Directorate on FMD under ICAR is responsible for epidemiology and surveillance of FMD in the country and based on the information generated strategies are developed for controlling FMD in the country by Government of India, and PDFMD/ ICAR has no role to play for regular FMD vaccination and other supports as demanded. The Animal Husbandry department was advised to trace all the ailing Mithuns in jungle and treat them for severe maggot infestation and secondary bacterial infection which is the cause of death in Mithuns. He stressed for immediate vaccination of not only apparently healthy Mithuns but also the in contact cattle and pigs in the area. Clinical materials from affected Mithuns were collected by the team for laboratory investigation.

Salient Epidemiological Finding

1. No FMD vaccination is being done in Cattle and Pigs since last two years in the state. Due to wild habitat of Mithuns it has not been possible to access, restrain and vaccinate them against any disease, at any time, by the state department; thereby these animals remain highly susceptible to all infectious animal diseases.

2. FMD has been prevalent from September 2009 in 'Sagalee' and 'Itanagar' under District Papum Pare and serotype 'O' virus has been responsible. FMD serotype 'O' virus was also diagnosed earlier during the year in the district of Lower Subansiri, East Siang, and Changlang, mostly in cattle.
3. Clinical materials collected from Mithuns and cattle during investigation were confirmed to be serotype O FMD virus.
4. The virus was isolated in cell culture and 2D-MNT test of serotype O viruses from Mithun revealed close antigenic relationship with in-use vaccine virus.
5. PanAsiaI lineage of Type O virus was responsible for disease outbreaks in Mithuns.
6. Initially cattle and pigs had the disease that spread to semi domesticated/wild Mithuns in Sagalee causing mortality.
7. Deaths in Mithun were due to severe maggot infestation and secondary bacterial infection, following FMD, in nostrils, muzzle and inter-digital spaces of feet and concurrent starvation. Animals were not in a position to breath properly due to blockade of the nostrils, and were lying here and there in the jungle. No treatment was given to the ailing Mithuns due to difficulty in restraining them in forest, thereby, increasing the number of deaths.
8. Mithuns in Jungle lives in groups and they identify their kith and kin only by smelling muzzle which helps in faster spread of FMD in the population.
9. Low/no awareness of Mithun owners and superstition of the owners that quality of meat deteriorates after FMD vaccination.
10. There were no cases of FMD in organized Government Mithun Farm at Sagalee that follows regular 6 monthly vaccination schedule.

14.2 Epidemiological Investigation on reported cases of FMD in Yaks at Nykmadung Farm of NRC on Yak

The scientists of PDFMD (Dr. A. Sanyal and Dr. S Pawar) reached NRC on yak in the night of 29th October, and visited the Yak Farm for FMD investigation alongwith the Director of the Institute. After visiting the Farm and examining some yaks, a meeting was conducted in the Farm premise at Nykmadung for discussion on the subject.

Examination of clinically infected animals by the team showed recovery and there was complete healing of the tongue, so no clinical tissue material could be collected for virus type identification. However, clinical materials collected earlier by the Guwahati Regional FMD Center were found negative for FMD virus. Serum samples from 33 apparently healthy animals were collected by the team for retrospective diagnosis. Animals were also found to take food normally.

The Director, NRC on yak presented the history of the suspected cases of FMD in Yak Farm. The farm experienced FMD for the first time. The disease was first noticed in the farm on 13th October, 2009. The officer-in-charge of Regional FMD Center, Guwahati had attended the reported cases and collected clinical materials and serum samples for diagnosis of FMD. The disease spread slowly and continued upto 23rd October. The animals were kept in 13 different shades and disease was observed at 4 adjacent shades. A total of 42 animals out of 204 animals were affected and showed clinical sickness. 12 animals below 6 month age (not vaccinated), 19 animals between 1 and 2 years and 11 adult animals showed clinical disease and death of one adult animal were also noticed. The animals are regularly vaccinated twice in a year and last vaccination was done in the month June 2009. The disease was noticed first in an around village of the farm where cattle was introduced from local market. The workers of the farm came in

contact with these affected animals and might have spread the infection in the Yak firm.

The Director asked for suggestion to take preventive measure in the event of FMD. Necessary suggestions were given and several preventive measures to be taken were discussed. It was felt that once the serum samples were tested at PDFMD, Mukteswar, necessary suggestions/ steps for keeping the yak herd free from FMD will be provided. It was also advised that after every round of vaccination serum samples should be collected between 21-28 days post vaccinations for evaluation of sero-conversion.

FMD affected Mithuns in Jungles

Examination of FMD affected Mithuns

Treatment of FMD affected Mithun by the team

FMD awareness programme by the team to the Mithun owners

Team along with the Director, NRCY Dirang

Examination of Yaks at NRCY for FMD lesions

15.0 Meetings, Reports and Recommendations

15.1 Proceedings of the 19th Annual scientists meet of the PDFMD (ICAR)

The 19th Annual Scientists Meet of the Project Directorate on Foot and Mouth Disease (PD on FMD) for the year 2007-08 was held on 18 – 20 June 2009 at Shimla, Himachal Pradesh. Scientists of all the regional centers (8) and network units (15) participated. In the plenary session, following recommendations were made, concerned have been communicated and action has been taken.

Recommendation of 19th ASM

1. As incidence of FMD has come down in HP and other Northern states (Punjab Haryana and Delhi), Disease Free Zone can be created in these states to boost export. **[Action: AHC/ State AH Departments/ PDFMD]**
2. No reagent/ biological originating from FMD virus shall be imported without prior permission of ICAR. **[Action: PDFMD/ Regional FMD Centers and Network Units]**
3. There should be no collaborative (external) research programme without prior permission of ICAR. **[PDFMD/ Regional FMD Centers and Network Units]**
4. The 3AB3 DIVA Kit developed indigenously and validated by PD-FMD should only be used for differentiation of infected from vaccinated animals, and PDFMD should undertake DIVA training programme for all concerned laboratories and scientists at the earliest for extending the technique to district level. **[Action: AHC/ CDDL/ RDDLs/ PDFMD/ Regional FMD Centers and Network Units/Joint Director, IVRI, Bangalore/ Head, Div. of Standardization, IVRI / Director, NRC Meat/ Director, NRC Pig/ Director, NRC Yak/ Director, NRC Mithun/ PD_ADMAS/ Director, CIRG/ Director, NRC Buffalo/ Director, PD-Cattle/ All FMD vaccine manufacturer]**
5. There should be harmonization and uniformity in FMD vaccine quality, and vaccination schedule between different programme (FMDCP, ASCAD, and RKVY), FMD vaccination should be twice a year irrespective of the vaccination programme till herd immunity is developed, there should be contiguity in area/ districts under vaccination irrespective of funding source (between FMDCP/ ASCAD/ RKVY), and 100% vaccination should be targeted as in FMDCP in all the vaccination programme. **[Action: AHC/ PDFMD/ Regional FMD Centers and Network Units/ Director, State AH Departments/ PD_ADMAS]**
6. Areas covered under FMD vaccination under ASCAD and RKVY should also be screened for level of protective antibody (sero-monitoring) as in FMDCP, so that effect of vaccination can be assessed and successful areas can be considered for maintaining zonal freedom from FMD. **[DAHD&F/ ICAR/ PDFMD]**
7. There is a need to harmonize FMD vaccine quality in the country at the earliest. **[DAHD&F/ PDFMD/ IVRI, Bangalore/ All FMD vaccine manufacturer]**
8. All FMD vaccine manufacturers in India should participate in extended FMDCP as a tool to harmonize vaccine quality. **[Action: DAHD&F/ AHC]**

9. Collaboration to be initiated with NRC's on Yak, Mithun or Pig for FMD surveillance in these species. [**Action: PDFMD/ Director, NRC Pig/ Director, NRC Yak/ Director, NRC Mithun]**

10. Efforts need to be strengthened and research programme initiated to increase the vaccine-protection time to at least 8-9 months. **[Action: AHC/ IVRI, Bangalore/ Head, Div. of Standardization, IVRI/ PDFMD/ All FMD vaccine manufacturer]**

11. A National FMD Random Seromonitoring program need to be initiated to estimate prevalence of FMD in cattle and buffalo in all districts of the country that will yield valuable information for taking policy decisions related to FMD control programme, and creating disease free zones. [**Action: PDFMD/ Regional FMD Centers and Network Units/ Director, State AH Departments/ AHC]**

12. No scientist working in the Regional centers and network units and trained in FMD diagnosis and epidemiology should be transferred without concurrence of ICAR. **[Action: AHC/ Director, State AH Departments/ In charge, Regional FMD Centers and Network Units]**

13. FMD situation in Kerala in spite of vaccination has to be investigated, and a team be sent for random serum sampling for detailed investigation.

[Action: AHC/ PD on FMD/ Director IAH&VB, Bangalore/ Director, Department of AH & VS, Kerala]

14. Southern states, West Bengal and North Eastern states need to be focused in FMD control programme. **[Action: DAHD&F/ ICAR/ AHC/ Director, State AH Departments]**

15. FMD reported by ICAR to DAHD&F should not be overlooked while compiling country status. **[DAHD&F]**

15.2 Proceedings/Recommendations of the 7th Meeting of the IMC held at Bhubneswar 0n 06-07-2009

The project Director presented scientific achievement and Targets of Directorate before the members. He informed the members that Project Technical Committee (PTC) & Project Monitoring Committee (PMC) has been

Recommendations of the 7th Meeting of the IMC held at Bhubaneswar, on 06/07/09

Item no.	Agenda/ recommenadation	Comments of the members of the imc	Comments of the director	Comments of the council
1	**Approval of the proceeding of the 6th Meeting of the Institute Management committee of the Project Directorate on FMD**	It was noted by all the members and were satisfied with the action taken	Agreed with IMC	**Noted**
2	Purchase of colour photocopier under Non-Plan	Agreed with the condition that unless the old photocopier is condemned, the new copier will not be purchased as replacement	Agreed with the IMC. The existing photocopier is B/W and has become obsolete. Colour photocopying facility is required for making reports and documents, and this facility is not available at this remote location (in the market)	**Agreed, subject to observance of council instructions issued in this regard from time to time and other codal formalities**
3	Appointment of full time Doctor on remuneration /term & conditions at par with RAs	Appointment of a Doctor on contractual basis was agreed by the Hon'ble members. Considering the remote location of the Institute, it was agreed as an important welfare measure for the employees of PDFMD, IVRI and CITH Mukteswar	3rd QRT strongly recommended improving medical facility for Scientists, Officers and staff of PDFMD that was agreed to by ICARGB. Appointment of a Doctor as AMA from Haldwani/ Nainital was also recommended by 4th IMC. Accordingly, effort was made to engage a Doctor/ lady Doctor on par-time basis but due to remote location of the institute it could not materialize. Now after lot of efforts, a lady Doctor has been engaged (on contractual basis) as a welfare measure who will cater to the needs of Staff of PDFMD & IVRI sister institute located at Mukteshwar	**Agreed, per the instructions issued by the council from time to time**

constituted by ICAR for establishment of International center for FMD.

Scientific achievement of the Directorate was appreciated by all the members. All agenda items were discussed at length & recommendation tabulated below. Action taken report of the previous IMC was also presented, and appreciated by the members. The hon'ble members visited the site of International Center for FMD on 5th July 2009 and attended the Foundation stone laying ceremony of the International center.

15.3 Proceedings of The 20th Annual Scientists Meet

The 20th Annual Scientists Meet of the Project Directorate on Foot and Mouth Disease (PD on FMD) for the year 2008-09 and six monthly progress for the period April 2009 to September 2009 was held on 8 –10 February 2010 at Imphal, Manipur. Scientists of all the Regional centers (8) and Network units (13) participated in this meet. The Network Unit, Patna could not attend the meet due to administrative problem. Lucknow Network Unit also did not participate. There were special invitees, Chairman and Members of RAC and other dignitaries participated in the meeting.

Shri I.S Laishram, IAS, Commissioner (Veterinary and Animal Husbandry), Manipur in his well come address highlighted that there is always a threat of FMD virus introduction from Myanmar border to Manipur. He emphasized that special attention has to given to the areas in NE region. Dr. M P Yadav, Chairman RAC and Guest of Honour stressed that India should be made free of FMD which results in economic loss to the tune of Rs. 20,000 crore/annum. Though there is reduction in FMD incidence due to FMD CP programme covering 54 districts, entire country needs to be covered for which sufficient fund is required. He informed that Sheep, gaots and Pigs need to be vaccinated including cattle and buffalo. Dr. SN Puri, Vice-Chancellor, CAU, Manipur as Guest of Honour informed that one pair of bullock per day work fetches Rs 150 and once they suffer from FMD they can not work for two weeks causing substantial loss to the farmer. He also recollected his childhood memories of treatment of FMD foot lesions with fish water. He also informed that pure bred Manipuri cows are resistant to certain important diseases. Dr. AK Srivastava, Director, NDRI as Guest of Honour, informed that livestock sector in India (annually) is losing substantially due to FMD. He informed that India contributes 0.2% Milk of the Global Production where as FMD free countries like New Zealand producing 1.5% of total global milk production with 37.5% global market share. He stressed that we should have a certification system of FMD free status to livestock farms having no outbreak of FMD during last three years to create Disease Free Zones (DFZ) to facilitate the export of milk and meat products. He stressed that message of two vaccination per year has to reach to each dairy farmer to control this disease. He emphasized that FMD is much of a managemental failure in elite herds in the country. The Minister for Veterinary and Animal Husbandry, Tribal Development and Hills, Govt. of Manipur emphasized about the important role of livestock farming in rural economy. He informed the economy of the state is mainly based on livestock production and incidence of FMD has reduced in Manipur due to the efforts of scientists with the use of advance scientific knowledge and technologies. He expressed his hope that FMD will be eradicated from India in near future.

Dr. Lal Krishna, Animal Husbandry Commissioner and ADG (AH), ICAR, in his presidential address thanked the Govt. of Manipur to hold this meeting. He informed the house about the negative impact of FMD on livestock economy with reduction of socio economic status of the farmer. He emphasized that FMD is an impediment to global food security. He elaborated the functioning of PD on FMD and the inbuilt AICRP component with 23 regional centers/ network units spread across the country for real time FMD surveillance and diagnosis. He informed that PDFMD has achieved the milestone in surveillance and molecular epidemiology of FMD with development of suitable vaccine candidates and diagnostics. He informed that FMD CP programme will be extended from 54 districts to cover more areas in the country. He further informed that International Center for FMD with BSL3+ containment facility will come up soon at Aragul, near Bhubaneswar to cater to the demand of South Asia and generate unified epidemiological data for formulation of Regional

(SAARC) FMD Control Programme using Indian experience. He also informed the house that ICAR successfully hosted the 3rd Annual meeting of the "OIE/FAO Global Network of FMD Reference Laboratories" during November 2009 in Delhi and will hold the Global Meet on FMD in India during December 2010. He congratulated scientists of PDFMD for developing a DIVA Kit for differential diagnosis of FMD infected and vaccinated animals. Dr. Th. Jeevan Singh, Director, AH, Govt. of Manipur extended the vote of thanks.

The first technical session was chaired by Dr. M P Yadav, Chairman, RAC, and co-chaired by Dr. Lal Krishna, AHC and ADG (AH), ICAR.

In this session, Dr. B. Pattnaik, Project Director, PD on FMD presented the overall progress and achievements of the institute and scenario of FMD in the country and impact of FMD control program (FMDCP). At the outset, he shared with the house that PD on FMD is now a member of the FAO/OIE Global Network of FMD Reference Laboratories that constitutes of ten other FMD laboratories in the world. He informed that there is gradual decrease in number of outbreaks since 2006-07, due to the effect of vaccination with appropriate vaccine strains and regular surveillance including sero monitoring. He added that, after phase 6 vaccination under FMDCP, 63.5, 64.4 and 54.2 percent of animals vaccinated/ tested were having protective antibody level (log_{10} 1.8 and above) against serotypes O, A and Asia-1, respectively, and this result is quite encouraging. He informed the house that disease free zone can be created in Northern states comprising 335 districts. Further, an indigenous recombinant- 3AB3 DIVA test has been developed just at the right time by the Central FMD Laboratory when there is immediate necessity to differentiate infected and vaccinated animals as the FMDCP progresses and validated using commercially available DIVA kit and reference positive and negative bovine serum. This kit has been found as sensitive as other commercial DIVA kit available internationally, and has made the country self sufficient in another FMD diagnostic and will save foreign exchange. Random sero-surveillance of bovine serum samples collected during 2008-09 revealed 27.9% of the bovine population in the country to be FMD virus positive. The incidence varied from 5.1% in Himachal Pradesh to 56% in Rajasthan depending on the extent/regularity of vaccination against FMD and geographical location/ advantage. Having an indigenously developed LPB-ELISA kit to monitor protective antibody response to prophylactic vaccination has also made the country self sufficient (and also economical) in evaluating of vaccinal antibody response under FMDCP. The project director advised all the centers/ network units to complete random seromonitoring at district level by November 2009 using the DIVA kit supplied by the Central FMD Laboratory and present the report during next ASM in the month of December 2009. He further stressed that all centers/ network units should present the scenario of FMD in their respective states for the period 2004-09 along with the extent of vaccination coverage at district level in the December meet. Dr. A. Sanyal, Principal Scientist presented detailed technical achievements of Central FMD Laboratory.

Summary of the progress made during 2008-09 and April 2009 to September 2009 is summarized below:

- A total of 511 outbreaks during 2008-09 and 421 outbreaks during April to September 2009 were recorded/ reported as against 1211, 1467 and 2962 outbreaks during the years 2007-08, 2006-07 and 2005-06, respectively. There is visible positive effect of regular vaccination under FMD Control Programme and other programmes in the country.
- During 2008-09, maximum numbers of outbreaks were reported in the southern parts of the country followed by Eastern region almost similar to last year. The four

states of South India contributed to 50% of the FMD outbreaks; almost same as that of 2007-08.

- Investigation of the outbreaks revealed low level of protective antibody (log_{10}<1.5) in affected animals at the time of infection as the predominating factor.
- No FMD case was reported in Punjab and Himachal Pradesh, whereas Haryana recorded a single case of Asia1 from Bhiwani district in March 2009 in cattle. During April to September 2009, no FMD cases were reported in the states of Punjab, Haryana, Himachal Pradesh, Rajasthan, Tamilnadu and Manipur.
- During 2008-09, a total of 1377 clinical samples were collected from 932 FMD outbreaks through the network of laboratory and subjected to virus typing. Virus could be identified in 895 samples viz. type O 834, type A 34, type Asia1 27 and the remaining samples were negative in ELISA.
- Multiplex PCR (mPCR) was applied on ELISA negative samples and by which another 55 outbreaks could be diagnosed.
- There was no incidence of type C FMD virus during this year also. Type O dominated the outbreaks scenario followed by types A and Asia1. During 2008-09, in all the geographical regions, other than Central India, serotype O was most prevalent. In central India type Asia1 replaced type A in causing outbreaks.
- There is complete absence of Asia1 in Southern region, whereas type A was absent in central and northern India during 2008-09.
- The incidence of Asia1 has drastically reduced in the Eastern and North-Eastern parts of the country during 2008-09 and there is an increase in the circulation of type O. During April to September 2009 a high proportion of outbreaks is recorded in the Eastern parts of the country followed by North-eastern region. Over the years, status of FMD in West Bengal has direct bearing on the incidences of the disease in NE states.
- Northern region had outbreaks due to serotypes O and Asia1. In Western region there was preponderance of serotype O, followed by Asia 1 and A. In Southern region also, there was dominance of serotype O like the previous year, and no incidence of type Asia 1 was recorded.
- Though majority of the outbreaks involved cattle, disease was also reported in buffaloes, pigs, goat and sheep.
- Though there is seasonal variation in occurrence of FMD in different parts of the country during the years 2008 and 2009, FMD outbreaks occurred round the year also during 2008 with maximum incidence in the month of March and May. In 2009, maximum incidence occurred in the months of August and September.
- Molecular epidemiological analysis of type O isolates based on 1D sequence analysis collected during 2008-09 revealed the re-emergence of Ind2001 strain in major parts of North India and some of the Southern States (Andhra Pradesh, Karnataka and Kerala). During this year also, PanAsia II strains was restricted to only few states and PanAsia1 was still detectable in Bihar, West Bengal and Orissa. During April to September 2009 Ind2001 lineage was predominant and this lineage is diverging over time. In addition PanAsia I and II were also responsible for disease outbreaks.
- PanAsiaI was responsible for disease outbreaks in Mithun in Arunachal Pradesh.
- Co-circulation of two different lineages of type O was recorded in the states of Assam and Bihar.

- During 2008-09, all the serotype A isolates were found to cluster within genotype VII, precisely in the VIIb-VP3[59] deletion group and this group is diverging over time giving rise to three different lineages. During April to September 2009, type A viruses responsible for disease outbreaks belonged to both deletion and non-deletion groups.
- In case of serotype Asia1, Lineage C continued to dominate also during 2008-09. This lineage was in circulation during 1998-2000, and has reappeared since 2005.
- All of field isolates of serotypes O, A and Asia 1 demonstrated close antigenic relationship with respective vaccine strains indicating good antigenic coverage by them.
- The nucleotide sequence analysis of complete L^{pro} and 3A region of type A field isolates revealed genetic divergence, leading to the formation of more number of lineages.
- The Project Directorate ensured regular supply of diagnostic kits (Virus serotyping ELISA and LPB-ELISA) to all AICRP Centers/ Network units and other government and non government agencies to maintain uniformity in the country.
- An indigenous recombinant- 3AB3 DIVA test has been developed and validated using commercially available DIVA kit and reference positive and negative bovine serum. A total of 29,763 random bovine serum samples collected during 2008-09 and April to September 2009, respectively revealed 27.9% of the bovine population in the country to be FMD virus positive. The incidence varied from 5.1% in Himachal Pradesh to 56% in Rajasthan depending on the extent/regularity of vaccination against FMD and geographical location/advantage.
- The project directorate continued to extend full technical and logistic support to the FMD Control Programme for post vaccination seromonitoring after each round/phase of vaccination being run by the Department of Animal husbandry, Dairying and Fisheries, GOI in selected 54 districts of the country. Reagents and consumables for LPB-ELISA along with test protocol, produced and developed by this Project Directorate, were supplied to all the testing centers to determine the serotype specific protective antibody level following vaccination. Seven regional FMD centers and the Central FMD Laboratory participated in the programme. Gradual increase in protective antibody response was observed subsequent to phase 1 vaccination. After phase 6 vaccination, 63.5, 64.4 and 54.2 percent of animals vaccinated/ tested were having protective antibody level (log_{10} 1.8 and above) against serotypes O, A and Asia-1, respectively. Testing of serum samples from phase 7 and 8 is under progress.
- The National Repository of FMDV was upgraded during this year also and now it comprises a total of 1548 (999-O, 279-Asia 1, 255-A and C-15) well characterized field isolates.
- Regular training and refresher courses for the scientific staff of Regional Centers and Network units were conducted on use/ application of virus typing ELISA, LPB-ELISA and DIVA ELISA. Overall performance of the regional centers and network units were monitored periodically and any technical difficulties faced by the centers were removed instantly through electronic guidance.

Subsequent technical sessions on 9th and 10th were chaired by Dr. Lal Krishna, AHC and ADG (AH), and Project Director, PDFMD co chaired. The scientists of the Regional centers and Network units presented the report for the year. Dr. R. Sharma of Hisar Regional center informed that there was an incidence of FMD in Kuruskhetra district of Haryana and it could not spread due to presence of herd immunity. It was suggested that they should perform DIVA to understand

virus circulation in that area. Bangalore center presented the report and showed that 48.4% of the animals are DIVA positive. Hyderabad center recorded FMD outbreaks only in coastal region and PDADMAS was requested to map this outbreaks. All the regional Centers and network Units should submit monthly report in time so that it is conveyed to ICAR and DAHD&F. It was decided that no irrelevant research work should be carried out by the regional centers and network units, and all clinical materials (portion of tissue samples) collected for diagnosis should invariably be transmitted to the Central FMD Laboratory. It was also decided that if there is FMD incidence in FMD CP region, detailed report will be submitted by the respective centers. In Arunachal Pradesh, there was outbreaks recorded in Mithun and only 20% of Mithun are vaccinated due to semi-domesticated nature. It was decided that North India Centers will be supervised by Mathura and Hisar center and Guwahati center will supervise all the North eastern states. Network unit of Kerala will be supervised by Bangalore center.

The plenary session, on 10th, was chaired by Dr. Lalkrishna and co-chaired by Project Director, PDFMD and co-chaired by Director, AH, Manipur and following recommendations were made. Dr. Lal Krishna appreciated the progress of the institute and contribution made by the Scientists. He congratulated the Scientists for developing an indigenous DIVA assay kit at the right time that will save valuable foreign exchange. He said that this kit will soon be released by ICAR in a suitable function, and the scientists developing this will be awarded. The following recommendations were drawn in this Annual Scientist Meet:

1. All the Regional Centers and Network Units should submit monthly report by 5th of the month, so that the information is conveyed to ICAR and DAHD&F **[Action: Regional FMD Centers and Network Units / PDFMD]**
2. E bulletin has to be brought out on monthly basis **[Action: PDFMD/ Regional FMD Centers and Network Units]**
3. Guidelines for collection and preservation of clinical material and epidemiological information to be circulated to all the Regional Centers and Network Units **[Action: PDFMD/ Regional FMD Centers and Network Units]**
4. Trained scientist working in the Regional centers and network units and trained in FMD diagnosis and epidemiology should not be transferred without concurrence of ICAR **[Action: ICAR/PDFMD]**.
5. The PI of Regional Research Units are responsible to monitor the activities of the units in the respective zone **[Action: PDFMD/ Regional FMD Centers]**
6. Network units of North India to be supervised by Hisar and Mathura Regional Centers on regular basis. Similarly Network units of North East to be supervised by Guwahati Center. Network unit of Kerala will be supervised by Bangalore center **[Action: PDFMD/ Regional FMD Centers]**.
7. Status report on Jammu Center has to be made and submitted in 3 months time by Hisar center **[Action: PDFMD/ Hisar Regional FMD Center]**.
8. As incidence of FMD has come down in HP, Punjab and Haryana, Disease Free Zone can be created in these states to boost export **[Action: DAHD&F/ICAR/PDFMD]**.
9. Instead of haphazard/scattered vaccination in all the districts in each state under ASCAD, 100% vaccination should be done in selected districts with a view to develop FMD free zone as a future strategy **[Action: DAHD&F/ ICAR]**.
10. Areas covered under FMD vaccination under ASCAD and RKVY should also be screened for level of protective antibody (sero-

monitoring) as in FMDCP, so that effect of vaccination can be assessed and successful areas can be considered for maintaining zonal freedom from FMD **[Action: DAHD&F/ ICAR/PDFMD]**..

11. There is a need to harmonize FMD vaccine quality in the country at the earliest. Efforts need to be strengthened to increase the duration of vaccine-protection time **[Action: DAHD&F/ICAR/PDFMD]**.
12. Uniformity has to be there in the schedule of vaccination as per epidemiological information **[Action: DAHD&F/ICAR/ State AH Departments]**.
13. Random Seromonitoring program need to be continued to estimate prevalence of FMD in cattle and buffalo in all districts of the country **[Action: DAHD&F/ICAR/ State AH Departments/PDFMD]**.
14. Rajasthan state need to be focused in FMD control programme which possess threat to neighboring states Punjab, Gujarat and Haryana **[Action: DAHD&F/State AH Departments]**.
15. There should be an uniformity in the presentation by all the Regional Centers and Network Unit **[Action: PDFMD/ Regional FMD Centers and Network Units]**
16. PDFMD should initiate research work to understand the early infection process of the disease. **[Action: PDFMD/Central FMD laboratory]**
17. There is need to develop pen side test for early and rapid diagnosis of the disease so that necessary emergency measures should be in place to contain the disease. **[Action: PDFMD/Central FMD laboratory]**

18. Monovalent vaccination policy based on epidemiological information. **[Action: DAHD&F/ICAR]**

19. Kerala seromonitoring should be done afresh **[Action: PDFMD/Bangalore center/ Kerala unit]**.

20. Each center and network unit select one village/ district under FMDCP/ASCAD/RKVY for seromonitoring.**[Action: PDFMD/ Regional FMD Centers and Network Units]**

15.4 Proceedings of the 2nd Meeting of the RAC held at Imphal, Manipur on 08.02.2010.

The second RAC meeting of the PD on FMD was held on 08.02.2010 at Imphal, Manipur under the chairmanship of Prof. M. P. Yadav, Ex-Vice-chancellor, S.V.B.P. University of Agricultural and Technology, Meerut.

The Project Director welcomed the chairman and members of RAC including special invitees to the second RAC meeting and requested the chairman Prof. M.P. Yadav to conduct the proceedings.Dr. Yadav stressed that India should be made free of FMD which results in economic loss to the tune of Rs. 20,000 crore/ annum. Though there is reduction in FMD incidence due to FMD CP programme covering 54 districts, entire country needs to be covered. He informed that Sheep, goats and Pigs need to be vaccinated including cattle and buffalo.

The chairman then requested Dr. B Pattnaik, Project Director, PD on FMD, Mukteswar, to present the research programmes and achievements of the Project Directorate including sero-monitoring of the animals covered under FMD-CP during the period from **April 2008 to September 2009**. Dr. Pattnaik presented salient achievements including active surveillance and molecular epidemiology of FMD, sero-monitoring of FMD-control program, etc. The salient achievements are as follows:

- A total of 511 outbreaks during 2008-09 and 421 outbreaks during April to September 2009 were recorded/ reported as against 1211, 1467 and 2962 outbreaks during the years 2007-08, 2006-07 and 2005-06, respectively. There is visible positive effect of regular vaccination under FMD Control Programme and other programmes in the country.
- During 2008-09, maximum numbers of outbreaks were reported in the southern parts of the country followed by Eastern region almost similar to last year. The four states of South India contributed to 50% of the FMD outbreaks; almost same as that of 2007-08.
- Investigation of the outbreaks revealed low level of protective antibody ($\log_{10}$<1.5) in affected animals at the time of infection as the predominating factor.
- No FMD case was reported in Punjab and Himachal Pradesh, whereas Haryana recorded a single case of Asia1 from Bhiwani district in March 2009 in cattle. During April to September 2009, no FMD cases were reported in the states of Punjab, Haryana, Himachal Pradesh, Rajasthan, Tamilnadu and Manipur.
- During 2008-09, a total of 1377 clinical samples were collected from 932 FMD outbreaks (511+421) through the network

of laboratory and subjected to virus typing. Virus could be identified in 895 samples viz. type O 834, type A 34, type Asia1 27 and the remaining samples were negative in ELISA.

- Multiplex PCR (mPCR) was applied on ELISA negative samples and by which another 55 outbreaks could be diagnosed.
- There was no incidence of type C FMD virus during this year also. Type O dominated the outbreaks scenario followed by types A and Asia1. During 2008-09, in all the geographical regions, other than Central India, serotype O was most prevalent. In central India type Asia1 replaced type A in causing outbreaks.
- There is complete absence of Asia1 in Southern region, whereas type A was absent in central and northern India during 2008-09.
- The incidence of Asia1 has drastically reduced in the Eastern and North-Eastern parts of the country during 2008-09 and there is an increase in the circulation of type O. During April to September 2009 a high proportion of outbreaks is recorded in the Eastern parts of the country followed by North-eastern region. Over the years, status of FMD in West Bengal has direct bearing on the incidences of the disease in NE states.
- Northern region had outbreaks due to serotypes O and Asia1. In Western region there was preponderance of serotype O, followed by Asia 1 and A. In Southern region also, there was dominance of serotype O like the previous year, and no incidence of type Asia1 was recorded.
- Though majority of the outbreaks involved cattle, disease was also reported in buffaloes, pigs, goat and sheep.
- Though there is seasonal variation in occurrence of FMD in different parts of the country during the years 2008 and 2009, FMD outbreaks occurred round the year also during 2008 with maximum incidence in the month of March and May. In 2009, maximum incidence occurred in the months of August and September.
- Molecular epidemiological analysis of type O isolates based on 1D sequence analysis collected during 2008-09 revealed the re-emergence of Ind2001 strain in major parts of North India and some of the Southern States (Andhra Pradesh, Karnataka and Kerala). During this year also, PanAsia II strains was restricted to only few states and PanAsia1 was still detectable in Bihar, West Bengal and Orissa. During April to September 2009 Ind2001 lineage was predominant and this lineage is diverging over time. In addition PanAsia I and II were also responsible for disease outbreaks.
- PanAsiaI was responsible for disease outbreaks in Mithun in Arunachal Pradesh.
- Co-circulation of two different lineages of type O was recorded in the states of Assam and Bihar.
- During 2008-09, all the serotype A isolates were found to cluster within genotype VII, precisely in the VIIb-VP3[59] deletion group and this group is diverging over time giving rise to three different lineages. During April to September 2009, type A viruses responsible for disease outbreaks belonged to both deletion and non-deletion groups.
- In case of serotype Asia1, Lineage C continued to dominate also during 2008-09. This lineage was in circulation during 1998-2000, and has reappeared since 2005.
- All of field isolates of serotypes O, A and Asia1 demonstrated close antigenic relationship with respective vaccine

strains indicating good antigenic coverage by them.

- The nucleotide sequence analysis of complete L^{pro} and 3A region of type A field isolates revealed genetic divergence, leading to the formation of more number of lineages.
- The Project Directorate ensured regular supply of diagnostic kits (Virus serotyping ELISA and LPB-ELISA) to all AICRP Centers/ Network units and other government and non government agencies to maintain uniformity in the country.
- An indigenous recombinant- 3AB3 DIVA test has been developed and validated using commercially available DIVA kit and reference positive and negative bovine serum. A total of 29,763 random bovine serum samples collected during 2008-09 and April to September 2009, respectively revealed 27.9% of the bovine population in the country to be FMD virus positive. The incidence varied from 5.1% in Himachal Pradesh to 56% in Rajasthan depending on the extent/regularity of vaccination against FMD and geographical location/advantage.
- The project directorate continued to extend full technical and logistic support to the FMD Control Programme for post vaccination seromonitoring after each round/phase of vaccination being run by the Department of Animal husbandry, Dairying and Fisheries, GOI in selected 54 districts of the country. Reagents and consumables for LPB-ELISA along with test protocol, produced and developed by this Project Directorate, were supplied to all the testing centers to determine the serotype specific protective antibody level following vaccination. Seven regional FMD centers and the Central FMD Laboratory participated in the programme. Gradual increase in protective antibody response was observed subsequent to phase 1 vaccination. After phase 6 vaccination, 63.5, 64.4 and 54.2 percent of animals vaccinated/ tested were having protective antibody level (log_{10} 1.8 and above) against serotypes O, A and Asia-1, respectively. Testing of serum samples from phase 7 and 8 is under progress.
- The National Repository of FMDV was upgraded during this year also and now it comprises a total of 1548 (999-O, 279-Asia1, 255-A and C-15) well characterized field isolates.
- Regular training and refresher courses for the scientific staff of Regional Centers and Network units were conducted on use/ application of virus typing ELISA, LPB-ELISA and DIVA ELISA. Overall performance of the regional centers and network units were monitored periodically and any technical difficulties faced by the centers were removed instantly through electronic guidance.

The Chairman applauded the achievements of the Directorate and the contributions made by the scientists of the Institute in the area of FMD research at global standards. He also congratulated Project Director and his team for successfully organizing the Global FMD meet in India.The Chairman subsequently invited suggestions/comments on the presentation made by Dr. Pattnaik. To the query made by Dr. Palaniswami, the Project Director informed that the incidence of FMD in the states of Punjab, Haryana, Himachal Pradesh and Delhi has become nil/ negligible due to regular vaccination and sero monitoring. Dr. Lal Krishna informed the house that the decision regarding the replacement of type A vaccine virus has already been made by the Government. He further added that vaccine strain updating exercise is a real time activity of Central FMD Laboratory.

S. No	Recommendation of the RAC	Comments of the Director	Comments of the council
1.	Inclusion of Pig, sheep and goat for FMD surveillance	A research project on this aspect has already been initiated. Random Serum samples from different region of the country are being collected to understand virus circulation and carrier status in these small livestock	Agreed & action initiated
2.	Area specific vaccination as per prevalence of serotype(s) in the region	It has been communicated to the DAHD&F. The matter of using monovalent Type O vaccine in areas having dominance of serotype O virus is under examination at DAHD&F since sometime (Recommendation 3 of first RAC refers)	The policy issue need to be undertaken with DAHDF
3.	Areas coved under FMD vaccination under ASCAD and RKVY should also be screened for level of protective antibody (sero-monitoring) as in FMDCP, so that effect of vaccination can be assessed and these areas can be considered for maintaining zonal freedom from FMD	Necessary diagnostic kit and service are being extended to the state governments on request. Both LPBE and DIVA kits are being provided on demand regularly. However, necessary directives required to be issued by the DAHD&F so that serum sampling of the ajnimals as in FMD CP areas is done by the respective state AH departments	Agreed & action initiated
4.	To initiate research work to understand the early infection process of the disease	A research project on the subject in mice model has already been initiated. Subsequently, it will be extended to cattle and buffalo	Agreed
5.	National FMD sero-surveillance should be initiated and continued on yearly basis	This activity is underway since last one year, in which 100 random serum samples (cattle and buffalo) from each district is being tested by DIVA. The results have provided important information on virus circulation in different states of the country. This activity is an annual exercise, as it will monitor level of virus clearance from areas under regular vaccination and areas where there is nil or sparse vaccination	Agreed and action initiated
6.	International collabo-rative research on FMD in frontier areas	A collaborative research project on alternate vaccine for FMD with USDA_ARS has already been finalized and initiated	Under conside-ration to the counci
7.	International center for FMD need to be completed and made functional at the earliest	The Project Technical Committee of the International center for FMD at Bhubanaswar (Arugul) has already submitted the detailed report, and this is under examination at the Council for administrative approval etc. The NDDB has already agreed to undertake this BSL3+ project on turnkey basis. Required land has already been acquired from Govt of Orissa near Bhubanaeswar (Arugul) and boundary wall is under construction to secure the land.	Action need be undertaken on Priority basis
8.	Diagnostic kits produced and supplied by the directorate should include all required reagents and components	The kits provided are complete with all required reagents and salts and ELISA plates	No comments

S. No	Recommendation of the RAC	Comments of the Director	Comments of the council
9.	Genomic and antigenic variation in field utbreak strains, particularly in case of serotypes O and A should be monitored in real-time	This activity is a continuous one, and virus isolate from each outbreak is analyzed in detail	Such study need be undertaken to assess the variance in subtypes, if any
10.	There is a need to generate complete genome nucleotide sequence data for the field outbreak strains to understand evolution of FMD virus at both translated sand untranslated genomic region in an endemic situation	Selected outbreak strains are being sequence in both the regions for generating necessary information	Agreed and need be undertaken
11.	Opening of new network centers at Sikkim, Jharkhand and Chhattisgarh	This will be projected in the EFC of 12th Plan	To be proposed during 12th Plan
12.	Developmint of pen- side diagnostic kits as a time bound programme	Work on this project is underway	Agreed
13.	National Information Network on FMD should be established	Necessary Help of DIPA and ERNET is being taken to establish this network through an interactive website. The officers /Scientists of DIPA and ERNET are formulating it	Agreed
14.	Efforts should be initiated to conduct sero-surveillance of FMD in the SAARC countries	A project on this aspect with FAO funding and guidelines is under examination in ICAR (Diagnostic laboratory network coordination for Surveillance and vaccine evaluation in South Asia)	Such proposal is under consideration

15.5 Proceeding of 5th Meeting of the PTC held at Hyderabad

The 5th meeting of the Project Technical Committee (PTC) was held between 27-01-2010 and 29-01-2010 in the Committee Room of Indian Immunologicals Limited (IIL), Hyderabad. The meeting was called for discussion regarding finalization of user specific requirement, layout and estimation of the cost of the Project.

The PTC continued the deliberations to finalize user specific requirements and other bio-safety and bio-security provisions and cost estimate of the project. The PTC decided that necessary optimal physical barrier has to be there in the main BSL3+ laboratory as cross contamination of Indian virus strains with any other from south Asian countries need to be eliminated. Therefore, the committee made necessary provisions in the layout of main BSL3+ laboratory.

For calculation of the costing, the following aspects/items were discussed and finalized by the members for calculation of the total requirement and cost of the project to make the facility fully functional ensuring bio-safety and bio-security norms as per international guidelines for safe handling of FMD virus.

- Land development, contour mapping and Land scaping
- Construction of boundary wall and main gate
- Main laboratory with BSL 3+ provision
- Experimental animal facility with BSL 3+ provision
- Diagnostic process laboratory with BSL 2 facility,
- Administrative block with conference facility,
- Small animal breeding unit
- Animal holding facility
- Effluent treatment plant
- Stores and laundry
- Service Building
- Security office
- International Trainee hostel
- Residential Housing
- Water supply and water harvest
- 33 KVA HT/LT electric substation
- Construction of internal road and Parking facility
- Internal electrification and street lighting
- Engineering services and structural equipments

The following decisions/ suggestions were made.

- The drawing of the main laboratory building made during 4th PTC meeting was discussed and finalized.
- It was decided the main laboratory building should have basement, ground floor and first floor. The total carpet area (all the floors) was worked out for the main laboratory building (civil works and modular partitions for making independent laboratory rooms).
- Use of Pre-Fabricated Insulated Modular Partitions for the main Laboratory instead or concrete wall partition was discussed. All the members went to visit the laboratory facility of IIL laboratory as well as the firm/ company who had supplied these modular partitions to IIL. The firm also informed that they are fabricating such modular partitions for CCMB, Hyderabad. To the queries of the team, the3 firm agreed to test the partitions at 150 pascals negative pressure.
- It was decided that the experimental animal facility should also have basement, ground floor and first floor,
- AHU (Air handling Unit) requirements to maintain graded negative air pressure in the containment area were calculated in detail for the main laboratory as well as experimental animal facility. Some modifications were also made at certain areas of these two buildings by the members. To maintain the negative pressure in animal house, air lock should be of 50 pascals negative pressure, main Laboratory at 50 pascals negative pressure, Clean corridor at – 50 Pascals, laboratories at (80) pascals negative pressure, dirty corridor in lab, at 100 pascals negative pressure and field sample handling area will be of 150 (120) pascals negative pressure. PM room in experimental animal facility, at 200 pascals negative pressure. So the system has to be tested accordingly for bearing maximum negative pressure.
- The exhaust air will be passed through UV decontaminator followed by heat treatment (heater/steam) before release to the atmosphere. It was decided to have Common ETP for both main laboratory and experimental animal house side.
- It was decided to have alarms/ walkie talky/ cell phone for emergency and pin pointing to the rooms of problem.
- Water requirement was also discussed and finalized. The water requirement for international center will be around 150 KL /

day to meet the requirements of 30 workers and 20 animals, out of which 75% water will be effluent.

- There will be two types of water system one for laboratory and diagnostic process laboratory and another for cell culture and water for injection.
- There will be generation of pure steam for use in autoclave whereas the black steam will come from boilers. All the members went to visit these facilities of IIL.
- It was decided to have two incinerators, one for large animals (280 Kg/ animal) and another for small animals and petty items/ laboratory wastages. In small animal house there has to be a bio-safety cabinet.
- There will be two numbers of security gates (one for main laboratory and one for experimental animal facility). There should be the provision of entry through thumb impression at the security gate for the laboratory workers for reasons of bio-security.
- Facilities for small animal breeding need to be created in experimental animal facilities.
- There should be a separate building for engineering section.
- Drainage facilities for rain water and rain water harvesting system should be in place.
- The ETP will constitute primary treatment heat based comprising holding tanks of 40 KL capacity and on-line cutters,-4 stainless steel sterilization vessels of 3KL capacity each for heating of the effluents at 125o fitted with agitators for uniform heating, 2 holding tanks each of 10KL capacity, and one un-insulated storage tank to hold the treated water. For the secondary treatment it is necessary to have TDS/ RO system/ sand bed filter system.
- ETP water will not be allowed to go outside the campus. Therefore, out of 100 acres of land, 50 acres will be used for erection of buildings and rest will be used for fodder cultivation and horticulture for rearing and maintaining FMD virus negative animals for use in experiments, and this is a must.
- There has to be UPS (Automatic) of 100KVA capacity (50KVA x 2) for total lighting inside the laboratory, all deep freezers and refrigerators in case of power failure/ interruption. The UPS should supply for a few minutes with power back up by DG set.
- The committee suggested to get clearances from Pollution control board, BARC, Boiler inspector (Nagpur), BDA / Panchayat (for building construction), CPSEA for laboratory animal testing, civil aviation for height of the building and chimney, electric inspector, labour commissioner etc.
- There should be provision of Architect and structural consultant, ETP Consultant, HBAC Consultant, GLP and bio-safety consultants, Laboratory accreditation and validation consultants. The service of one architecture has to be hired at the earliest for the purpose.
- Laboratory Equipment list will be provided by PDFMD which will include Gamma Chamber, Freeze dryers, bio-safety cabinets (Type II, B2), Fume cabinet, Fume hood etc.
- After all technical discussions, user specific requirement and layout of containment laboratory, containment animal house and diagnostic process laboratory along with other supporting requirements to run the facility were finalized and cost of the total project for establishment of International Center for FMD was worked out at Rs. 148 crore at the present cost. It was decided that DGM (AH), NDDB who is also a member of PTC to submit layout drawings and details of estimate to the convener of PTC for onward transmission to the chairman, PTC and the council.
- The committee suggested that as there is

not much time left in the 11th Plan period, to start with only selected civil works may be undertaken and completed in about 2 years time.

15.6 OIE/ FAO FMD Reference Laboratories Net Work Meeting 2009

FAO and OIE have launched a new initiative for global control of FMD, in which the OIE and FAO international Laboratories on FMD play the important role. OIE and FAO have an agreement in control of transboundary diseases. FMD control in endemic countries will require active partnership to overcome the limitation of insufficient epidemiological informations and limited capacity and resources to apply control measures. To that effect, the action of FAO and OIE aims to address the information gaps needed to develop effective national and regional strategies and to assist in emergency responses.

The OIE/ FAO FMD Reference Laboratories Net Work Meeting 2009 was held at NASC Complex, ICAR, New Delhi during 23rd to 27th November 2009. The FMD Reference Laboratories Network is a network of OIE and FAO reference laboratories for Foot and Mouth Disease (FMD) that has been established under the secretariat of the OIE Reference Laboratory and FAO World Reference Laboratory for FMD, IAH, Perbright, UK (WRL FMD)., in which Project Directorate on FMD, Mukteswar is an OIE/ FAO reference centre in India. The purpose of the global FMD network is to safeguard the international trade of animals or their derived products requires an efficient global surveillance for Foot and Mouth Disease (FMD) including constantly updated information on antigenic and genetic characteristics of FMD virus (FMDV), involved in current outbreaks. These informations are required for the development and selection of vaccines and other tools for surveillance and control of FMD, as well as harmonization of such approaches. Strengthening of reference laboratory capabilities supports regional FMD control schemes under the auspices of the OIE/FAO global frame work for the progressive control of Transboundary Animal Diseases (GF- TADS).

The objective of the Global FMD Reference Laboratory Network are -

(1) To gather, generate, analyse and make available of the laboratory informations on the global occurrences and spread of FMD and on the characterization of FMDV.

(2) To provide recommendations on vaccine strain selection for the implementation of control schemes and for vaccine antigen reserves.

(3) To offer expertise to OIE, FAO and member countries to assist in the control of FMD.

(4) To harmonize approaches to the characterization of FMD Viruses.

(5) To increase the competence of reference laboratories to identify constraints to the functioning of the network and to propose solutions.

Representatives from OIE / FAO FMD Reference Laboratories of UK, Botswana, Russia, Brasil, Argentina, South Africa, USA, Thailand, Belsium and India participated along with observers invited from other reference laboratories of China, Germany and Kenya.

FAO and OIE have a long road map for FMD control in West Euresia and South East Asia and are willing to partner India in developing a similar road map. The challenges are many in that road map including the cost of vaccine, short duration of immunity multiple serotypes of the virus and large number of live stock and wild life. The Network of OIE/ FAO Reference Laboratories for FMD are contributing for a clean understanding of the threats posed by FMDV. The global network meeting on FMD mainly envisaged upon the (1) update of the global and regional FMD situation during 2009, (2) regional FMD Vaccine strain recommendation, (3) conclusions on inter – laboratory vaccine matching studies conducted in n2009, (4) plans for future inter – laboratory

vaccine matching studies, (5) an update and draft report on regional quality assurance undertaken in support of FMD laboratory testing, (6) and a better understanding of the present FMD control programme in India.

In India the research on FMD has progressed from am unmonitored form to the understanding of the dynamics of disease maintenance through the surveillance and monitoring of FMD in network laboratories. The reference centers of FMD provides the services for FMD surveillance, assisting in risk management and threats, involved in training and development of specialized human resources, introducing improved laboratory methods and harmonizing the lab methods practiced all over the country. The Indian national programme for FMD control includes mass vaccination which started in 2003-04 covering 34 million cattle and buffaloes in 54 districts will be further increased step wise to achieve the goal of controlling FMD in 2020. The introduction of wide spread large scale vaccination has greatly reduced the number of reported cases of this disease. However it is required to increase the scale of vaccination and improve the thermostability and potency of the present vaccines with affordable costs to address the existing problems of FMD in India. At present there is substantial fall in the number of laboratory confirmed outbreaks due to effective surveillance in addition to vaccination programme in place. Several states now have a very low FMD incidence and this may enable the development of FMD free zones in India. Among the FMD cases reported, the serotype 'O' remained predominant and the prevalence serotypes 'A' and 'Asia 1' FMD virus varied temporally and geographically. However there is no report of the involvement of serotype 'C'of FMDV since 1995. In India an indirect ELISA test kit to detect antibodies against 3AB3 FMDV non – structural protein (NSP) using the recombinant protein expressed in *E.Coli* has been developed, validated and in use in the country. A survey of 32000 cattle in different regions of India had revealed an overall NSP sero-prevalence of 31%, with ranges from 6 – 46 %. The test kits found to have good levels of sensitivity and specificity. The test kit was a cheaper alternative to imported kits. In this meeting the participants from Russia and Botswana took the sample kits for validation in their laboratories and the participants from Brasil and South Africa showed interest to use this kit after completing official formalities. It was emphasized that ICAR has all the capabilities for surveillance and control of the disease in the South Asian Region. The establishment of an International Center for FMD to be built in the city of Bhubaneswar by 2012 will cater to the needs of the SARC countries. The global community considered that there is a gap to be filled in the coordination of South Asian Reference Centers and the Indian National reference Laboratory of FMD could play an important role in this area to bridge the gap.

The European communities seek the partnership of laboratories from India, China and South America as well as from Europe for a new European project, "Disconvac," that supports a range of research activities related to facilitating vaccine based FMD control measures.

The conclusions drawn in this global network meeting were as follows: –

(1) The members of the network should seek cooperative actions between the meetings.

(2) The PD- FMD, India is carrying out a wide range of reference laboratory activities at an advanced level with considering resources to take a lead in establishing a regional network for Southern Asia.

(3) Provision of proficiency testing schemes is essential to maintain quality assurance.

(4) The recent focus of the network on vaccine matching is justifiable, but reference laboratories should ensure that vaccines used in their regions are of adequate potency and encourage and promote effective delivery.

(5) The naming of the regional virus pools has

been changed to use the terms as "Southern Africa", "Eastern Africa", "Western Africa", "Southern Asia" and Eastern Asia".

(6) The network will evaluate the convenience of using the format "SAT2-KEN-2010-1-Wrl" for the designation of the samples.

(7) The network agreed to working towards production and sharing of reagents for vaccine matching methods and results or data.

(8) The PD-FMD, India will supply WRLFMD with a selection of viruses and VP1 sequences representing the main genetic lineages circulating in India in the last five years. The PD-FMD and WRLFMD will analyze antigenic and genetic findings jointly in comparison to their own data.

(9) The PD-FMD, India will supply their new DIVA test kit to other Network laboratories willing to assist in validation of the method.

(10) A spread sheet will be prepared to collate Network information on vaccine matching tests performed with training and reagent needs as a start to prioritizing capability improvement.

There will be a listing of vaccine strains and their manufacturers.

16.0 Education, Trainings and Awards

The Scientists of PD FMD are involved in teaching various courses for the M.V. Sc., and Ph.D. students of Division of Veterinary Virology for their degree at Indian Veterinary Research Institute. During the year the following courses were offered by the scientists of the project: Viral Pathogenesis, Immunity to Viral Infection, Viral vaccines, Diagnostic Virology, Virological Technique, Advanced Virological Technique, Slow Viruses and Virus like agents, Advanced General Virology, Avian viruses.

Nine training *cum* workshop sessions consisting of a total of 49 days on "recombinant 3AB3 nonstructural protein based indirect ELISA for differentiation of FMD virus infected from vaccinated animals (DIVA)" were organized, in which 60 researchers from network units/regional centres, private FMD vaccine manufacturing companies and universities participated.

Twenty five personnels from the Regional Centers and Network Units were given training on Liquid Phase Blocking ELISA for detection of antibodies to structural protein of FMDV.

Two students completed their MV.Sc. dissertation work in Microbiology and one student completed his M.Sc dissertation work in Biotechnology at central laboratory of the Project Directorate on FMD.

Participation in Trainings

1. Dr. B. Pattnaik and Dr. Sachin S. Pawar attended Brain Storming Session on "Vaccines & Diagnostics" organized at NDRI, Karnal (10 to 11-07-2009).
2. Dr. Sachin S. Pawar attended Workshop on PERMIS*net* II organized by IASRI at Delhi (22-07-2009).
3. Dr. B. B. Dash and Dr. Sachin S. Pawar attended training programme on "Web design methodologies and protocols" held at NAARM, Hyderabad (8 to 18-09-2009).
4. Dr. B. B. Dash attended training programme on "IT-based DSS for Geographical Information System for Rural Livelihood assessment", held at NIRD, Govt. of India, Hyderabad (11 to 20-03-2010).

Publications

Research articles in journals

1. Mohapatra, J. K., A. Sahu, S. K. Barik, A. Sanyal, B. Pattnaik (2009). Comparative analysis of the large fragment of the 5'

S.No	Name of the students and institute	Title of the Research Work	For Degree
1	Dr. Ajjaiah IVRI, Deemed University, Izatnagar	Selection of serotype A FMDV candidate vaccine strain in context of emergence of VP3[59] deletion group in India	MVSc
2	Dr. Amit Khutia W.B.U.A.F.S, Kolkata	Comparative sequence analysis and structural variability at 5' LFUTR region of Foot and Mouth Disease Virus serotype O isolates	MVSc
3	Mr.Biswajit Das HND, Garhwal,	Vaccine matching exercise and molecular epidemiology of type O FMDV in India Uttarakhand	MSc

untranslated region (LF-5′ UTR) of serotype A foot-and-mouth disease virus field isolates from India. ***Virus Genes*** Vol. 39, 81-89.

2. Mohapatra, J. K., P. Priyadarshini, L. K. Pandey, S.Saravanan, D. Hemadri, A. Sanyal, B. Pattnaik (2009). Phylogenetic analysis of 3C protease ($3C^{pro}$) coding region of Foot-and-mouth disease virus type A. ***Acta Virologica*** Vol. 53, 175-183.

3. Sanyal. A., S.Saravanan, J. K. Mohapatra, R.P. Tamilselvan, N.K.Singh, D.Hemadri, B. Pattnaik. (2010). Phylogenetic analysis of Indian serotype Asia1 foot-and-mouth-disease virus isolates revealed emergence and reemergence of different genetic lineages. Veterinary Microbiology. doi:10.1016/j.vetmic.2009.12.034

Abstracts/Presentations in Conferences

1. Mohapatra, J. K., L. K. Pandey (2009). Recombinant 3AB3 NSP-ELISA: A handy technique for differentiation of Foot and Mouth Disease virus infected from vaccinated animals (DIVA). ***OIE/FAO reference laboratories network meeting 2009,*** 23-27 Nov. 2009, ICAR, New Delhi.

2. Tamilselvan R.P, Ankan De, Sanyal A and Pattnaik B (2009).Genetic transitions of Indian serotype O foot and mouth disease virus isolates responsible for field outbreaks during 2005-2009: A Brief Note. ***OIE/FAO reference laboratories network meeting 2009,*** 23-27 Nov. 2009, ICAR, New Delhi.

3. Mohapatra, J. K., S. Pawar, P. Ramesh Kumar (2009). Molecular epidemiology of serotype A FMD virus in India. ***OIE/FAO reference laboratories network meeting 2009,*** 23-27 Nov. 2009, ICAR, New Delhi.

4. Saravanan. S and A.Sanyal (2009). Molecular epidemiology of serotype Asia1 FMD virus in India. ***OIE/FAO reference laboratories network meeting 2009,*** 23-27 Nov. 2009, ICAR, New Delhi.

5. Mohapatra, J. K., L. K. Pandey, B. Pattnaik (2010). r3AB3 DIVA-FMD Kit: Application in Foot-and-Mouth Disease Surveillance. ***XXIV Annual Convention of Veterinary Microbiologists, Immunologists and Specialists in Infectious Diseases (ICIAVMI)*** and International Conference on "Protecting Animal Health: Facilitating Trade in Livestock and Livestock Products", January 27-29, 2010, Raipur, Chhattisgarh.

6. Saravanan.S and B. Pattnaik (2010). Significance of molecular diagnosis of Foot and Mouth Disease. XIX National conference on recent trends in viral disease problems and management. **VIROCON-2010**. Indian Virological Society. March 18-20, 2010, Tirupati, A.P.

Chapters in Training Brochures

Mohapatra J. K. (2009). Recombinant 3AB3 NSP-ELISA: A handy technique for differentiation of Foot and Mouth Disease virus infected from vaccinated animals (DIVA). Short term training course on *"Serological and PCR based diagnosis of economically important infectious diseases of domestic animals".* 10-30 Nov. 2009, Sponsored by DBT, GOI, Organized by CADRAD, IVRI, Izatnagar.

Awards

Dr. J. K. Mohapatra received **"Professor LSS Kumar Memorial Award 2009"** in Agricultural Sciences group by Indian National Science Academy (INSA), New Delhi, India.

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OIE
ANNUAL MEET OF OIE / FAO GLOBAL NETWORK OF
FMD REFERENCE LABORATORIES, 2009