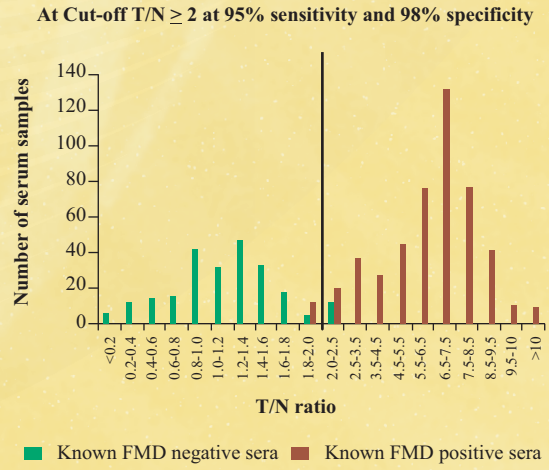
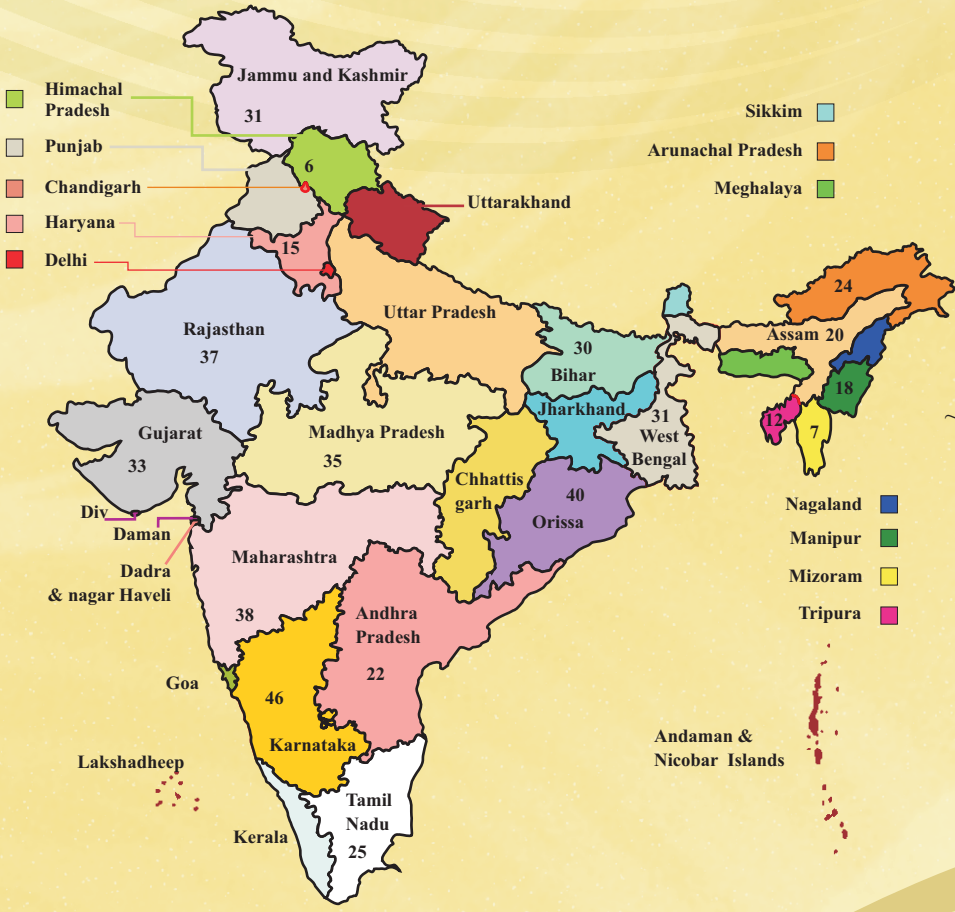
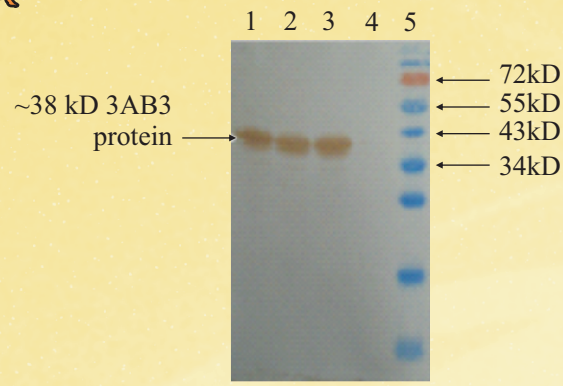
Annual Report
2008-09







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

Project Directorate on Foot and Mouth Disease

Annual Report 2008-09

IVRI Campus, Mukteswar-263 138
Nainital, Uttarakhand, India

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Project Directorate on Foot and Mouth Disease, Mukteswar

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Front Cover Page

Prevalence of FMD in the country based on DIVA test, Frequency distribution of normalizes test to positive control(T/P) ratio of sera from animals with known FMD status and western blot analysis with anti His antibodies of expressed 3AB3

Back Cover Page

Snow covered view of Himalaya (Top) and rear view of Central FMD Laboratory, Mukteswar

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Contents

PDFMD
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1.0 Project Director's Report

FOOT and Mouth Disease (FMD) has for centuries been known as a serious threat to the health and welfare of the domestic and wild ruminant animals, elephant and swine population of the world, with negative impacts on the livelihoods of animal keepers. The production, performance and use of large ruminants for ploughing and traction are seriously diminished when infected with FMD. Production and production efficiency is further diminished in terms of quality and quantity of dairy products and weight gain ratios. As per OIE and FAO, countries having FMD are more prone to food insecurity as a result of the impact of FMD at household level through reduced access to local, national and international markets and animal draught power for agriculture. Seventy countries in the world are already 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. Globalization of trade and movement of people and animals opens the door for any virus strain to infect any part of the world. There is need of a strong commitment of all countries at a high political level to harmonise global, regional and national policies for FMD control (OIE/FAO). The Global FMD Conference 2009 in Paraguay, which involved OIE national delegates, stakeholders, representatives of FAO and other partner international organisations, key global donors, non-governmental and farmers' organizations recommended a Globally Coordinated Approach to Control FMD. OIE official recognition of zonal freedom from FMD is an important element in the drive towards the global control of FMD and the facilitation of trade in animals and animal products. Regional long term efforts are needed to address the threats of FMD viruses, animal reservoirs and environmental persistence. India/ICAR have decided upon demand from OIE/FAO to establish an International Center for FMD for the SAARC region during the 11th Five-year period to support and facilitate launching of South Asian FMD Control Campaign.

Quality vaccine that is fit for the prevailing field strains in each serotype and in each relevant species is an important tool for control of FMD. Effect of regular active and passive surveillance and vaccination in the states of Himachal Pradesh, Punjab, Haryana and Delhi in achieving near to zero incidence of FMD is a success story for zonal control within an endemic country that can be replicated in the southern peninsular India and elsewhere. These four northern states sharing border contiguity can be focused for attaining zonal freedom from the disease, and a zonal freedom model can be developed in the country for application elsewhere. Though India produces suitable FMD vaccine, there is an urgent need in the country for research in vaccines to improve duration of immunity and thermo stability to meet challenging environmental conditions.

The Project Directorate on Foot and Mouth Disease (FMD), is 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 providing the technical support and scientific input information to the planners and strategy making agencies in making the FMD control programmes currently undergoing in the country a success, and opens the road towards control of the disease in the SAARC region with India as the leader.

During this period, a total of 511 outbreaks were recorded/reported as against 2962, 1467 and 1211 outbreaks during the year 2005-06, 2006-07 and 2007-08, respectively. The details of the outbreaks are shown in Table 1. Gradual decline in the incidence has been due to effective surveillance and practice of strict vaccination regime in more productive areas in the country. Maximum outbreaks were recorded in southern parts of the country followed by Eastern region almost similar to last year. There seems to be a visible positive effect of regular vaccination under FMD Control Programme and other programmes in the country.

A total of 660 clinical specimens were collected from 511 outbreaks and some of these are 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 cases of the disease in the country. Using sandwich ELISA, virus could be diagnosed in 166 samples. A total of 37 samples negative on ELISA could be diagnosed by mPCR. Retrospective diagnosis was made in a number of outbreaks also.

No FMD cases were reported in Punjab and Himachal Pradesh, whereas Haryana recorded a single case of serotype Asia1 outbreak from Bhiwani district in March 2009 in cattle. Tamil Nadu that experienced a rampant epidemic last year recorded only two outbreaks this year due to infection immunity. Though there is seasonal variation in occurrence of FMD in different parts of the country during 2007-08, maximum

Table 1. Number of FMD outbreaks reported during last three years

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

Table 2. Number of FMD outbreaks diagnosed and serotypes involved (Centers/Unit)

		Serotype detected in sandwich ELISA				Percent typed/ diagnosed	NVD/ undiagnosed
		O	A	Asia1	Total		
Number of tissue samples	640	334	26	16	376	58.7	264
Number of Outbreaks	511	198	19	24	241	47.2	270

incidence of disease was reported in the month of March. During 2008-09, FMD outbreaks occur round the year also with maximum incidence in the months of November and January 2009 (Fig.1).

region, whereas type A was absent in central and Northern India. Type O dominated the outbreak scenario followed by Asia1 and A. The incidence of Asia 1 has drastically reduced in the Eastern

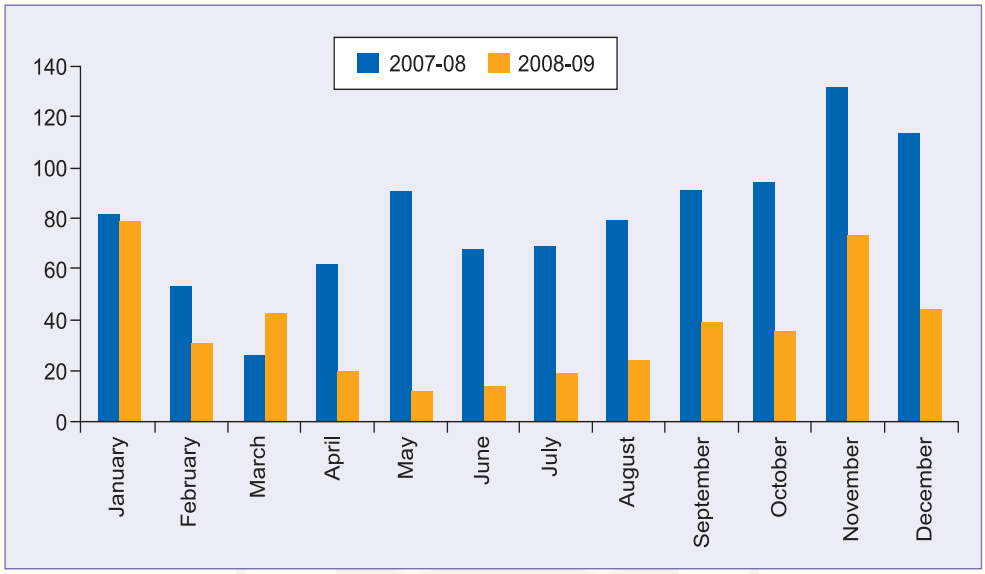


Fig. 1 Month wise occurrence of FMD outbreaks

During 2007-08, in all the geographical regions, other than central India, serotype O was most prevalent. The trend is similar during 2008-09 also, except type Asia1 replaced type A in causing outbreaks in central India alongwith type O. There is complete absence of Asia1 in Sothern and North Eastern parts of the country and there was an increase in the circulation of type O (Table 3). Though majority of the outbreaks involved cattle, disease was also reported in buffaloes, pigs, sheep and goats.

Table 3. Distribution/incidence/percent of FMD virus serotypes in different geographical regions as revealed by serotyping of clinical materials diagnosed during 2007-08 (numerator) and 2008-09 (denominator)

Geographical region	Serotypes			Number of clinical materials serotyped	
	O	A	Asia-1	2007-08	2008-09
North	21/12(67.7%/80%)	07/0 (22.5%/0)	03/03(9.7%/20%)	31	15
West	98/18 (88.2%/69.3%)	01/02 (0.9%/7.7%)	12 /06(10.8%/23%)	111	26
South	493/80 (91.8%/86%)	38/13 (7.1%/14%)	06/0 (1.1%/0%)	537	93
East	315/104 (80.5%/91.2)	47/04 (12%/3.5%)	29/06 (7.5%/5.3%)	391	114
North East	83/60 (70.3%/88.2%)	04/07 (3.3%/10.3%)	31/01 (26.3%/1.5%)	118	68
Central	32/60 (39.5%/70.6%)	39/0 (48%/0%)	10/25 (12.5%/29.4%)	81	85
Total	1042/334(82.1%/83.3%)	136/26(10.7%/6.5%)	91/41(7.2%/10.2%)	1269	401

Studies of antigenic relationship of the field outbreak strains with in-use vaccine strains is a regular exercise to monitor antigenic variation, if any, occurring in the field. This year, a total of 59 virus isolates including 35 type O, 16 type Asia1 and 7 type A field isolates were subjected to one-way antigenic relationship study. Serotype O and Asia1 isolates causing extensive outbreaks in the country are antigenically related to and covered by the current vaccine strain. Inspite of subtle variations (within group antigenic heterogenicity) in the antigenic makeup of type A field isolates, the new vaccine strain IND 40/00 offers optimum antigenic coverage to the recent field isolates. The National FMD Virus Repository is upgraded annually with latest/new virus isolates. This year, a total of 53 virus isolates including 41 type O, 3 type Asia1 and 9 type A field isolates were added to the repository. At present this National Repository contains a total of 1455 (934-O, 264-Asia 1, 242-A, 15-C) well characterized field isolates.

Molecular Epidemiological analysis of forty serotype O isolates, drawn from 14 states revealed complex epidemiological situation and genetic diversity of the field isolates in the country. This year, 'Ind2001' strain has re-emerged in major parts of Northern India and in some of the Southern states (Andhra Pradesh, Karnataka and Kerala). Besides, the PanAsia II strain, which dominated serotype O outbreaks in previous year was restricted to only few states, while the parental PanAsia strain (PanAsia I) is still detectable in Bihar, West Bengal and Orissa. In type A, all the isolates were found to cluster within genotype VII and precisely in the VP3^{59} deletion group. VP3^{59} deletion group after its resurgence in 2007 continues to dominate the field outbreak scenario even in 2008 and 2009 and is genetically diverging with time giving rise to three lineages. In case of Asia 1, lineage C continued to dominate during this year also as observed during 2007-08. This lineage dominated the (Asia 1) outbreaks between 1998 and 2002 and reemerged as dominating lineage since 2005.

This recent reemergence of VP3^{59} deletion group and genetic diversification has evoked interest and urgency to analyze regions beyond structural protein coding region of this group to verify the earlier prediction of its uniqueness and within group heterogeneity. The complete L^{pro} and 3A region of field isolates from VP3^{59}-deletion group and ten isolates belonging to other genotypes/lineages spanning a period of three decades were resolved for comparative analysis purpose which is supposed to assist the ongoing epidemiological investigations. The deletion group maintains genetic monophyly even at the nonstructural protein coding regions. Akin to the 1D region, this deletion group is gradually diverging genetically even at L and 3A region forming more number of lineages.

We have developed a recombinant nonstructural protein (3AB3) based ELISA test for differentiation of FMD infected from vaccinated animals (rDIVA-FMD) at Central FMD laboratory, Project Directorate on FMD, Mukteswar. This DIVA kit is at least 4 times cheaper than the imported kits. A total number of 18,326 random serum samples collected at the rate of 100 per district from 234 districts covering 20 different states of the country were tested in DIVA ELISA in an exercise to estimate FMD prevalence in the country. This revealed 27.94 % of the bovine population in the country to be FMD infected during 2008-2009, which might fluctuate consequent upon inclusion of data from bigger states like Uttar Pradesh.

This Directorate is extending full technical and logistic support to the FMD Control Programme 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. 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 64.2

percent of animals tested were having protective antibody level (log_{10} 1.8 and above) against serotypes O, A and Asia-1, respectively. Serum samples of phases 7 & 8 are under process.

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.

I am happy to share that PDFMD is now a member of the Global FAO/OIE Network of FMD Reference Laboratories that constitutes of ten other FMD laboratories in the world. We participate in Global FMD Vaccine matching exercise. The institute is also now a member of GFRA (Global Foot and Mouth Disease Research Alliance). International Center for FMD for SAARC/South Asia (South Asia Regional Reference Laboratory for FMD) to cater to the demand of the South Asian (SAARC) countries is also to be established during 11th Plan period (2007-2012). Creation of this international laboratory with state-of-the-art features of bio-safety and bio-containment (BSL 3+) will facilitate in Global participation. 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.

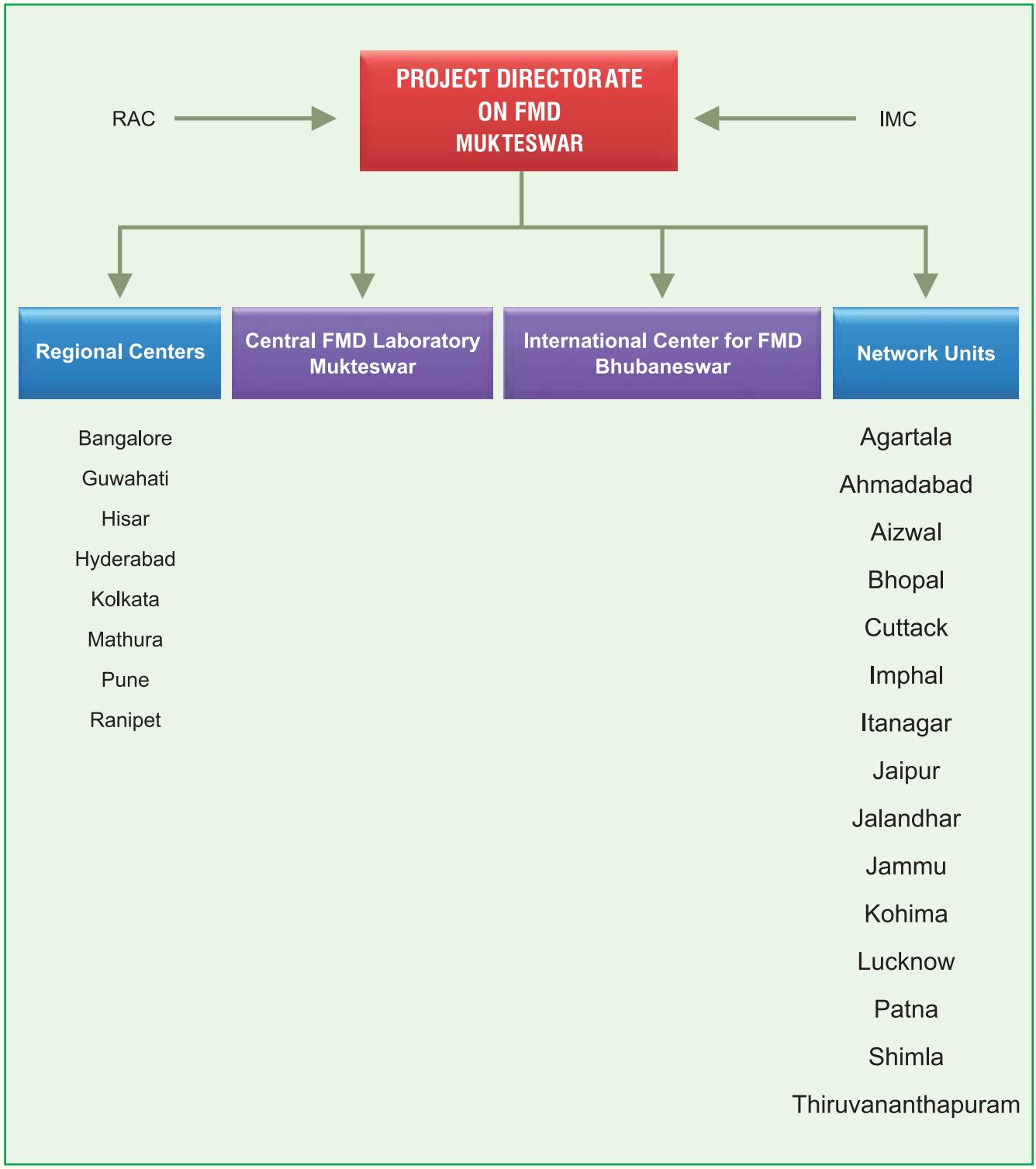
Objectives

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 strains 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 1D 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 (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.

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	Sr. Scientist	August 1996	Continuing
3.	Dr. D. Hemadri	Sr. Scientist	August 1996	December, 2008
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. P. Rameshkumar	Scientist	June 2008	Continuing
9.	Dr. K. Muniswamy	Scientist	June 2008	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 1997	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 D.S. Ginwal	S. S. Gr. IV (Lab)	December 1985	Retired on 30-04-2008
2.	Shri Gobind Singh	S. S. Gr. IV (Lab)	December 1985	Continuing
3.	Shri J.P. Bhan	S. S. Gr. IV (Machine)	February 2008	Continuing

5.0 Epidemiology Report

Table. 4 FMD outbreaks reported/recorded during 2008-09 and serotype involved (Reported by centers and network units)

States	Reporting Centre/Unit	No. of outbreaks recorded	No. of Samples tested	Serotyping Results				% Typed
				O	A	Asia1	NVD	
Southern Region								
Tamil Nadu	Ranipet	2	8	-	-	-	8	0
Andhra Pradesh	Hyderabad	13	28	19	-	-	9	67.8
Karnataka	Bangalore	190	71	46	1	-	24	66
Kerala	Thiruvananthapuram	58	59	15	12	-	32	45.7
	Total	263	166	80	13		65	
Central Region								
Madhya Pradesh	Bhopal	33	119	60	25	-	34	71.4
	Total	33	119	60	25	-	34	
Western Region								
Gujarat	Ahmedabad	9	29	13	-	-	16	44.8
Maharashtra	Pune	10	19	5	2	6	6	68.4
	Total	19	48	18	2	6	22	
Northern Region								
Punjab	Jalandhar	Nil outbreaks						
Haryana	Hisar	1	3	-	-	2	1	66.6
Jammu&Kashmir	Jammu	35	15	10	-	-	5	66.6
Uttar Pradesh	Mathura	6	13	2	-	1		15.3
Himachal Pradesh	Shimla	Nil outbreaks						
	Total	42	31	12	-	3	6	
Eastern Region								
Orissa	Cuttack	23	35	13	-	-	22	37.1
West Bengal	Kolkata	79	139	91	4	6	38	72.6
	Total	102	174	104	4	6	60	
North Eastern Region								
Assam	Guwahati	15	20	14	-	-	6	70
Mizoram	Aizawl	2	3	2	1	-	-	100
Nagaland	Kohima	6	8	4	-	-	4	50
Meghalaya	Guwahati	4	8	3	-	-	5	37.5
Tripura	Agartala	19	20	16	-	-	4	80
Manipur	Imphal	1	4	2	-	-	2	50
Arunachal Pradesh	Itanagar	5	39	19	6	1	13	66.6
	Total	52	102	60	7	1	34	
	Grand Total	511	640	334	26	16	197	

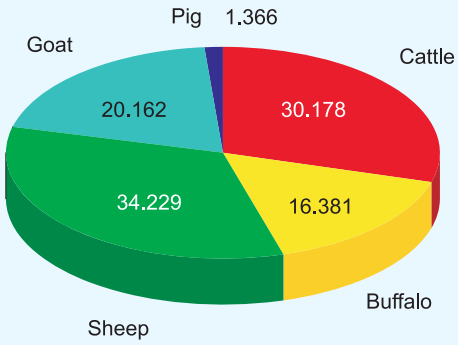
5.1 Southern Region

Karnataka (Bangaluru): During the year under report, a total of 190 Outbreaks have been reported with 3736 attacks and 79 deaths. A total of 71 clinical samples were processed for virus typing by Sandwich ELISA. FMD virus type 'O' was recovered from 46 samples and FMD virus type 'A' from 1 samples. From the remaining 24 samples virus could not be recovered. FMD virus type 'O' was diagnosed from 23 outbreaks, FMD virus type 'A' from 1 outbreak. The total vaccination coverage in the state was around 45.2%. The highest number of outbreaks was reported during the month of November 2008 (58) followed by September 2008 (25), December 2008, February 2009 (16) and October (15). The highest number of outbreaks was reported from Uttar kannada (35) followed by Chikkaballapur (24) and Gulbarga (22) and Hassan (22).

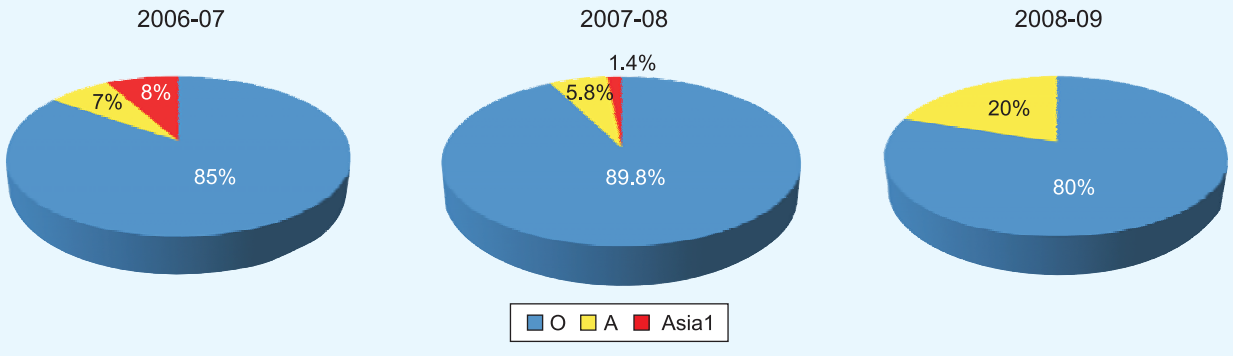
Kerala (Thiruvananthapuram): During the year the state recorded 58 outbreaks (45 outbreaks and 13 sporadic incidences) with 399 animals affected and 4 deaths. From the outbreaks, 59 samples were received/collected for FMD serotyping and were subjected to Sandwich ELISA. Of these, serotype O was present in 15 samples, A in 12 samples and no virus was detected (NVD) in 21 samples. 11 samples were unsuitable material (serum, blood etc) for virus typing. Most of the outbreaks were mild and number of animals affected per outbreak was below five. Outbreaks were mostly sporadic in nature. Spread of the infection was also very limited. More number of outbreaks was reported in southern districts such as Thiruvananthapuram, Kollam and Pathanamthitta. Thiruvananthapuram experienced 25 outbreaks and Kollam and Pathanamthitta had 16 and 6 outbreaks,

Scenario of FMD in Southern Region

Susceptible animal population
(in millions)



Serotype-wise prevalence



respectively. Maximum number of outbreaks were recorded in the month of January. In the previous years also outbreaks were more during December and January and could be due to increased animal movement from outside for slaughter purpose in the festival months. Cattle, goats and pigs were the only species affected.

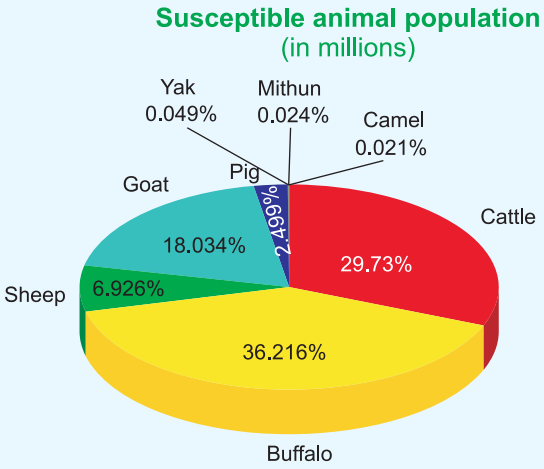
Tamil Nadu (Ranipet): During 2008–09, only 2 FMD outbreaks were recorded in the whole State. One FMD outbreak from Omalur Taluk in Salem District and another FMD outbreak from Polur Taluk in Tiruvannamalai District were reported. Both the outbreaks were reported in July, 2008. During the period from April 2008 to March 2009, only a total of 8 clinical materials were received/collected. All the received specimens could be processed by Sandwich ELISA and no virus could be detected from any of the samples.

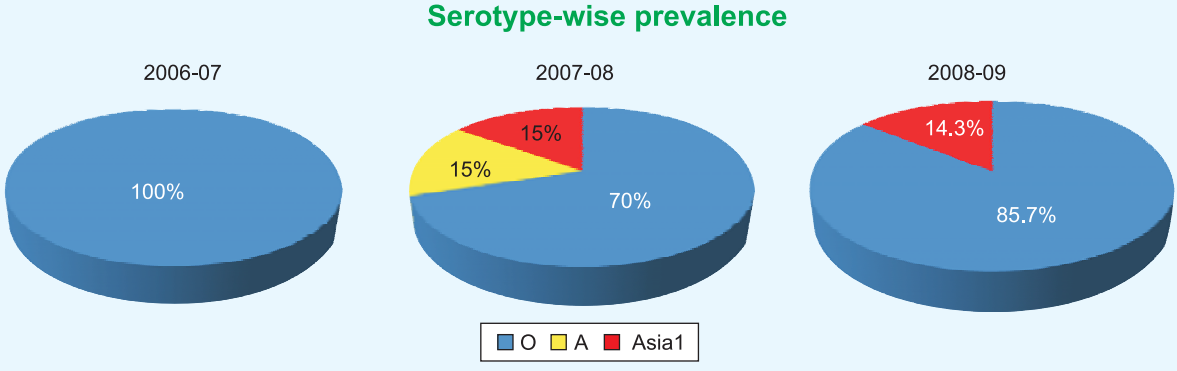
Andhra Pradesh (Hyderabad): During the year under report 13 outbreaks of FMD were reported with a total of 549 cases. The disease was reported throughout the year. The highest number of outbreaks (4) was recorded during the month of March. The highest number of outbreaks (5) were recorded in the district of Vizianagaram. A total of 28 samples from 13 outbreaks were tested and 19 were found to be positive for O serotype.

5.2 Northern Region

Haryana (Hisar): A single FMD outbreak (from Haryana) was recorded from Bhiwani district in March 2009 in cattle. A total of 3 FMD specimens were collected. Of these two samples were typed using ELISA and found to be serotype Asia1.

Scenario of FMD in Northern Region





Punjab (Jalandhar): The state has not recorded any FMD outbreak during 2008-09.

Jammu and Kashmir (Jammu): A total 35 outbreaks were reported throughout the State. Seven outbreaks were reported from Jammu Division, 17 from Kashmir division and 11 outbreaks from Ladakh. All outbreaks have been reported in bovines only. Total 14,494 animals were affected and 17 deaths were reported. Outbreaks have been reported in Rajouri (5) & Jammu district (2) of Jammu Division, Budgam (2), Anantnag (2), Baramulla (3), Pulwama (1), Srinagar and Kupwara (4) districts of Kashmir Division and Leh (1), Kargil (10) in ladakh Division. Outbreaks have been reported during April to October. Out of 15 samples processed, 10 samples were found to be positive for O type.

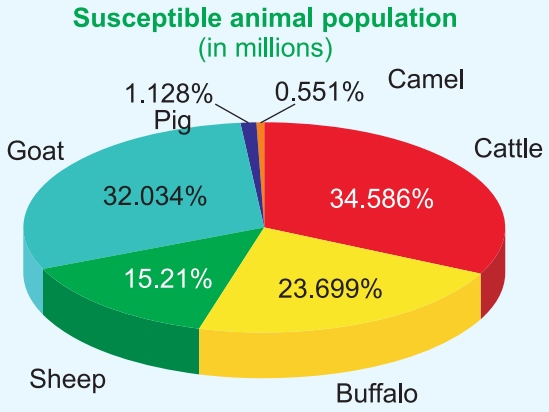
Himachal Pradesh (Shimla): During the year 2008-09 there was no incidence of the FMD in our State.

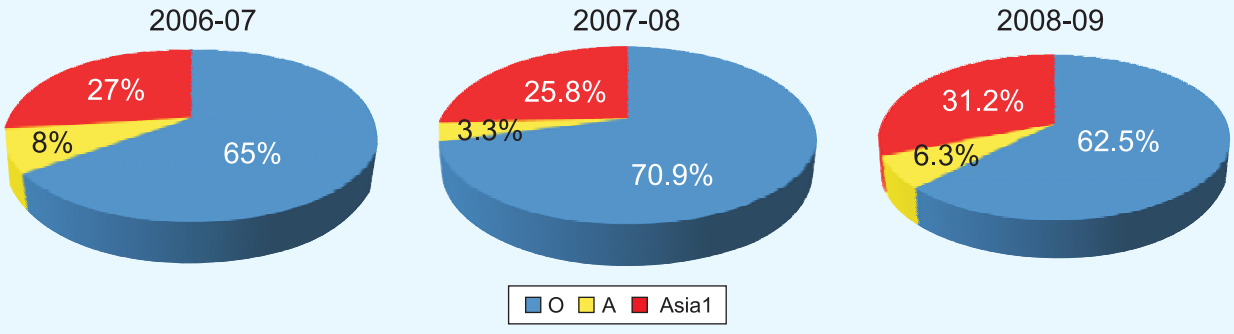
Uttar Pradesh (Mathura): During the period under report, a total of 13 FMD specimens (vesicular tongue epithelium/oeso-pharyngeal Fluid) were collected from clinically affected cattle and buffaloes from 6 outbreaks. Out of 13 specimens processed, 02 were typed as FMDV type 'O', 01 as type 'Asia-1' and no outbreak was detected due to FMD type A. Seasonal analysis of the FMD outbreaks revealed that all the recorded outbreaks were in the months of winter and spring (October, February and March). Six outbreaks were recorded from Mathura, Aligarh, Etawah, Basti and Gorakhpur districts of Uttar Pradesh.

5.3 Western Region

Gujarat (Ahmedabad): During the year 2008-2009, a total 9 outbreaks of Foot and Mouth Disease were recorded. All the outbreaks recorded during the period of September-2008 to March-2009. These outbreaks were mostly

Scenario of FMD in Western Region





recorded in cattle and buffaloes. Outbreaks of FMD were not recorded in Vadodara region during the year 2008-09. Highest number of outbreaks was recorded in the Kutchh district. Out of the total 29 samples typed, 13 samples were of type O. Six outbreaks could be typed and found to be serotype O.

Maharashtra (Pune): A total of 10 outbreaks were recorded during the period**.** Out of 19 tissue samples received, 13 samples (68.42%) were positive in typing ELISA. The type distribution noted for O type is 5 samples (38.46%), A type 2 samples (15.39%) and Asia-1 type 6 samples (46.15%). The outbreaks have been reported in the months of April, August, September, December, January, February & March.

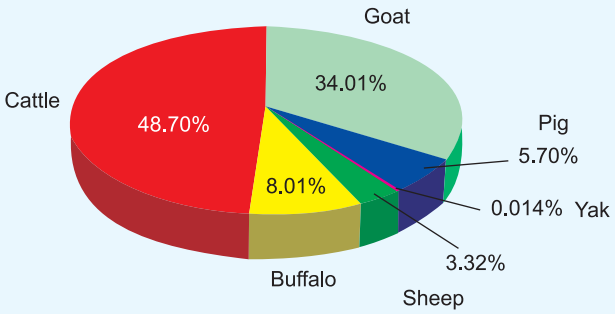
5.4 Eastern Region

West Bengal (Kolkata): The center received 139 field samples from 79 reported F.M.D outbreaks of different field units of the districts of West Bengal. Out of 139 samples received / collected from different outbreak, 91 samples were typed as O, 04 samples were confirmed as type A and 06 samples were typed as Asia-1 FMD Virus. The highest number of outbreaks reported in South 24 Parganas district (15), followed by Burdwan (11) and Nadia (09). Involvement of O type FMD virus was detected in 46 outbreaks, Asia 1 type in 04 outbreaks and A type FMDV in 03 outbreaks. Maximum numbers of outbreaks (20) were reported during the month of December'08 followed by March' 09 (17), Feb' 09 (12) and Jan' 09 (10).

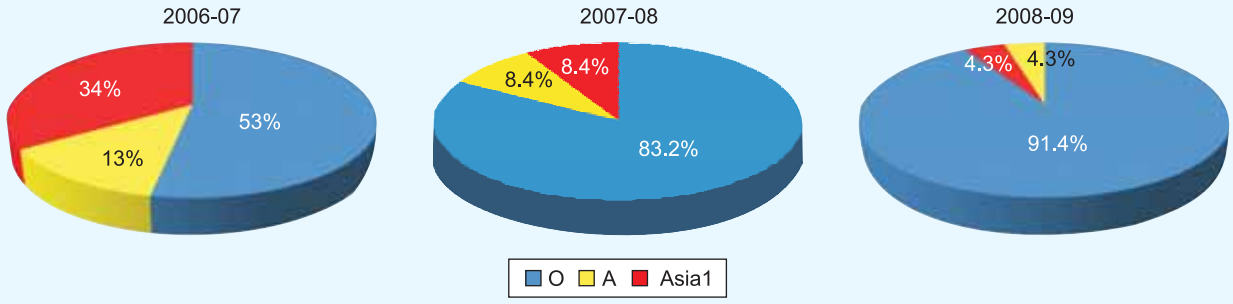
Orissa (Cuttack): During the year under report, 23 outbreaks were recorded with 777 attacks and 12 deaths mostly in young calves. Out of these 23 outbreaks, 7 were diagnosed by sero-typing and 11 by retrospective diagnosis and rest 5 reported on the basis of clinical

Scenario of FMD in Eastern Region

Susceptible animal population
(in millions)



Serotype-wise prevalence



symptoms. No outbreak was reported in Sheep, Goats and Pigs. Of the typed samples, FMD virus could be identified in 13 samples and no virus could be detected in 22 samples. From 13 positive samples, FMD virus type O was detected in all samples. Maximun number of outbreaks was reported from Cuttack (6) district. The outbreaks were mostly limited to a particular area of town and village of the districts.Highest numbers of outbreak was recorded in the month of August and October. Mostly Cattle consisting of local non-descript and crossbred animals were affected. No outbreaks in Caprine, Porcine and Ovine species was reported during the period under report

5.5 North Eastern Region

Assam (Guwahati): A total of 27 outbreaks of FMD were recorded and studied in Assam and other North Eastern States. The highest number (15) of outbreaks was in Assam followed by 6 outbreaks in Nagaland, 4 outbreaks in Meghalaya and 2 outbreaks in Mizoram. FMD outbreaks have been recorded and studied only in 6 districts of Assam. The highest number (4) of outbreaks was in Kamrup district. A total of 39 samples collected/ received from those outbreaks were processed for typing of FMD virus. FMD virus type O could be detected in 23 samples and type A in one sample. Nine isolates of FMD virus type O could be isolated in BHK-21 cell line.

Mizoram (Aizawl): A total of two outbreaks were recorded in the month of April and March. Three samples were collected and subjected to sandwich ELISA and in which two were found to be type O and one was type A.

Tripura (Agartala): Nineteen outbreaks were recorded during the period under report. Seven outbreaks in the West District, 9 outbreaks in North District, 2 outbreaks recoded in south Tripura and one in Dhalai district. Maximum outbreak was recorded in the month of April (05). Out of 20 samples processed for serotyping, 16 samples were positive for FMD virus type O. Virus type could be identified as type O in 13 outbreaks (ten outbreaks were identified using typing ELISA and 3 were detected retrospectively from serum samples in LPB ELISA). A total of 611 cattle & pigs were involved in the outbreaks.

Arunachal Pradesh (Itanagar): A total of 5 outbreaks were recorded from five different districts of the state. The first outbreak of the year was reported from the plains of Lower Subansiri district adjoining to lakhimpur and Dhemaji district of Assam. Probably from theses districts, it spread to the other nearby districts of the state. The disease started in the months of April, 2008 and lasted till Jan, 2009. A total of 39 samples were subjected to typing ELISA and of which 19 samples were typed to be O, 6 were type A and 1 type Asia1.

Manipur (Imphal): During the year, only one FMD outbreak was recorded in Chandel district during the month of December. A total of 87 cattle were affected in the outbreak with a mortality of 1 adult and 8 young calves. The source of infection was due to illegal import of cattle from Myanmar which were transported through these villages. A total of 4 clinical materials could be collected from these outbreaks and only 2 clinical samples were sufficient and suitable for FMD virus typing. These two samples were found positive for O type virus. Further, 20 sera samples were collected from these outbreak after 5 days and out of these samples, 18 sera samples were tested by LPB-ELISA and 15 serum samples found to have antibodies against O type and 11 serum samples against A type virus.

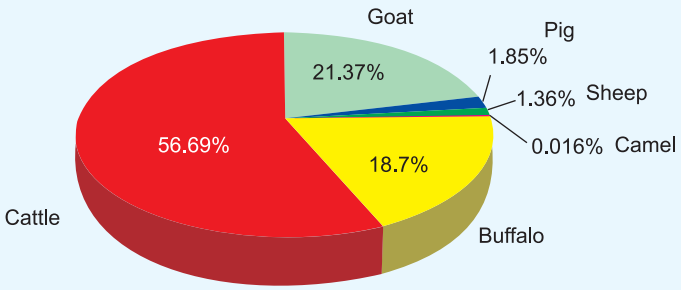
5.6 Central Region

Madhya Pradesh (Bhopal): Thirty three outbreaks were recorded from twelve districts of MP viz. Bhopal (6), Balaghat (5), Gwalior (4), Seoni (4), Chindwara (3), Jhabua (3), Mandla (2), Shivpuri (2), Betul (1), Rajgarh (1), Sagar (1) & Tikamgarh (1). Twenty eight outbreaks involved both cattle & buffaloes, five outbreaks were reported solely in cattle, while no outbreak

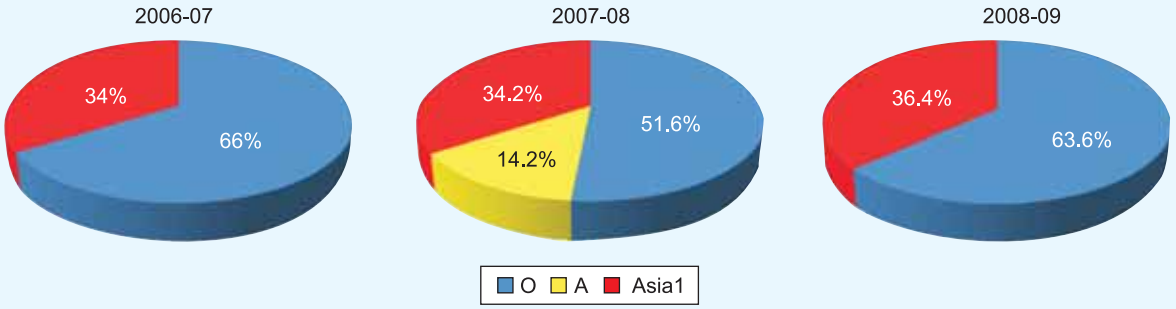
was reported in goat, sheep & pigs. Outbreaks were recorded in all the month barring July and maximum number of outbreaks were reported in the month of April (5) and June (5). One outbreak was reported from JCB Farm, Bhopal and was diagnosed to be type O. One hundred nineteen clinical samples were processed and tested by sandwich ELISA and of which 85 samples were typed (60 type O and 25 type Asia1). Of the 33 outbreaks, 21 outbreaks were due to type O and 12 outbreaks were caused by type Asia1.

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

The Central Laboratory of the Project Directorate received 436 clinical samples from centers/network units for serotype confirmation and detailed analysis during the year (Table 5 and 6). The samples were processed and subjected to sandwich ELISA for type identification and multiplex PCR in case of ELISA negative samples. Using sandwich ELISA, virus serotype could be diagnosed in 166 samples, of which 144 samples were typed as type O, 9 samples were confirmed as type Asia1, and 13 samples were typed as type A. Samples were also processed in BHK 21/ IBRS-2 cells and virus could be recovered.

6.2 Detection of FMDV in ELISA negative clinical samples by multiplex PCR

During this year, a total of 59 ELISA negative tissue samples (each from different outbreak) from different states were subjected to mPCR. The samples were selected randomly from undiagnosed outbreak clinical materials received during 2008 and 2009. Positive and negative controls were kept strictly in order to avoid false positive and negative results. FMD serotype could be identified in 59 samples and by this 63% of the outbreaks which went undiagnosed using ELISA were diagnosed by mPCR (Table 7). Both serotype O and A could be identified in one outbreak samples.

Table 5. Number of outbreaks samples referred to PD FMD and serotypes involved

		Diagnosed by sandwich ELISA				Diagnosed by m-PCR				Total	NVD/ undiagnosed
		O	A	Asia1	Total	O	A	Asia1	Total		
Number of samples referred	436	144	13	9	166	29	8	-	37	203	233*
Number of outbreaks	242	115	10	6	131	29	8	-	37	168	74

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

Table 6. Details of the field materials received at PD FMD collected during the year 2008-09

S. No.	Centre	Samples Received	Virus types detected at PD on FMD				Source host species for FMD positive samples						Vaccination status for positive samples		
			O	A	Asia1	NVD	Cattle	Buffalo	Sheep\ Goat	Pig	Wild life	NA	V	UV	NA
1.	Agartala	2	1	-	-	1	2	-	-	-	-	-	-	2	-
2.	Ahmedabad	27	8	-	-	19	18	9	-	-	-	-	14	3	10
3.	Bangalore	105	34	2	-	69	94	2	9	-	-	-	50	32	23
4.	Bhopal	39	7	-	-	32	37	1	-	-	1	-	-	39	-
5.	Bhubaneswar	35	13	-	-	22	35	-	-	-	-		31	4	-
6.	Guwahati	15	14	-	-	1	12	-	-	3	-	-	3	12	-
7.	Mizoram	2	1	-	-	1	2	-	-	-	-	-	-	2	-
8.	Nagaland	2	1	-	-	1	1	1	-	-	-	-	-	1	1
9.	Meghalaya	3	2	-	-	1	3	-	-	-	-	-	1	2	-
10.	Hisar	2	-	-	2	-	2	-	-	-	-	-	-	-	2
11.	Hyderabad	16	7	3	-	6	8	4	-	-	-	4	-	-	16
12.	Itanagar	12	-	-	-	12	-	-	-	-	10	2	-	10	2
13.	Jaipur	31	7	-	1	23	24	7	-	-	-	-	10	21	-
14.	Jalandhar	3	-	-	-	3	2	-	-	-	-	1	-	-	3
15.	Jammu	5	3	-	-	2	5	-	-	-	-	-	-	-	5
16.	Kolkata	60	19	2	5	34	56	-	1	3	-	-	17	43	-
17.	Mathura	4	1	-	1	3	3	1	-	-	-	-	4	-	-
18.	Patna	25	13	-	-	12	21	4	-	-	-	-	8	17	-
19.	Pune	1	-	-	-	1	1	-	-	-	-	-	-	-	1
20.	Ranipet	5	-	-	-	5	2	-	2	-	1	-	-	-	5
21.	Shimla	3	-	-	-	3	3	-	-	-	-	-	-	-	3
22.	Trivandrum	19	-	5	-	14	15	-	-	-	-	4	4	6	9
23.	GADVASU	6	4	1	-	1	-	-	-	-	-	6	-	-	6
24.	CUL, Chennai	3	-	-	-	3	2	-	-	-	1	-	-	1	2
25.	Meerut cant	8	7	-	-	1	8	-	-	-	-	-	8	-	-
	Sub Total	**436**	**144**	**13**	**9**	**270**	**358**	**30**	**12**	**6**	**13**	**17**	**153**	**195**	**88**
	Total			**166**											

Table 7. Results of mPCR on ELISA negative clinical samples

No of ELISA negative samples*	Type O	Type A	Type Asia1
59	29	08	-

*from different outbreaks

6.3 Genetic and antigenic characterization of field isolates

6.3.1 Type O

Molecular epidemiology

Forty serotype O isolates, either in the form of infected tongue epithelium or cell culture supernatants were subjected to phylogenetic analysis using partial/complete 1D genomic region sequence. The isolates were drawn from West Bengal, Karnataka, Kerala, Bihar, Maharashtra, Gujarat, Assam, Meghalaya, Uttar Pradesh, Punjab, Nagaland, Andhra Pradesh, Rajasthan and Orissa for the said purpose. Phylogenetic analysis revealed presence of many sub-lineages, thus indicating the complex epidemiological situation and genetic diversity of the field isolates in the country (Fig.2). This year, 'Ind 2001' strain has re-emerged in major parts of Northern India and in some of the Southern states (Andhra Pradesh, Karnataka and Kerala). PanAsia II strain, which dominated serotype O outbreaks during 2007-08 was restricted to only few states (Orissa, Assam and Maharashtra), possibly due to infection acquired immunity and extensive vaccination that has been practiced in almost all parts of the country. The parental PanAsia strain (PanAsia I) is still detectable in Bihar, West Bengal and Orissa. Interestingly, more than one genetic group is found to be present in some of the states such as in Orissa (both PanAsia I and II) and Bihar (PanAsia I and 'Ind 2001'). Interestingly, some of the isolates recovered from the state of Gujarat were found to lack an amino acid at $VP1_{138}$. Based on grouping of the field isolates on the phylogenetic tree, it can be said that these isolates have probably originated from 'Ind 2001' strain. Nevertheless, it is heartening to note that the current vaccine strain (IND R2/75) may still offer protection against the said strain, as evident from 2D-MNT results. Besides the above strain, outbreaks were also caused by viruses of some minor sub-lineages and 'Ind 2001' strain in Gujarat.

Antigenic characterization

Thirty five representative serotype O isolates recovered from FMD outbreaks in Andhra Pradesh, Gujarat, West Bengal, Uttar Pradesh, Meghalaya, Assam, Rajasthan, Orissa, Nagaland, Kerala, Bihar, Tripura, Maharashtra and Karnataka were subjected to two dimensional-micro neutralization test (2D-MNT) and the results are shown in Fig 3 and 4. From the results, it can be seen that 33 of the 35 isolates (94%) show close antigenic relation ($r \geq 0.4$) with the current vaccine strain. The remaining two isolates (IND 157/08 and IND 91/09) gave r values between 0.20 and 0.39 (6%). Thus it can be concluded that the serotype O isolates causing extensive outbreaks in the country are antigenically related to and covered by the current vaccine strain.

As there is always scope for increasing the antigenic coverage and for inter and intra-laboratory comparison of test results, vaccine matching studies have been initiated by the Project Directorate with industrial partners. To this end, five field isolates belonging to different genetic group, along with in-use vaccine strain (IND R2/75) were used for raising bovine vaccinate serum (BVS). Homologous and heterologous serum titer against the selected panel is in progress.

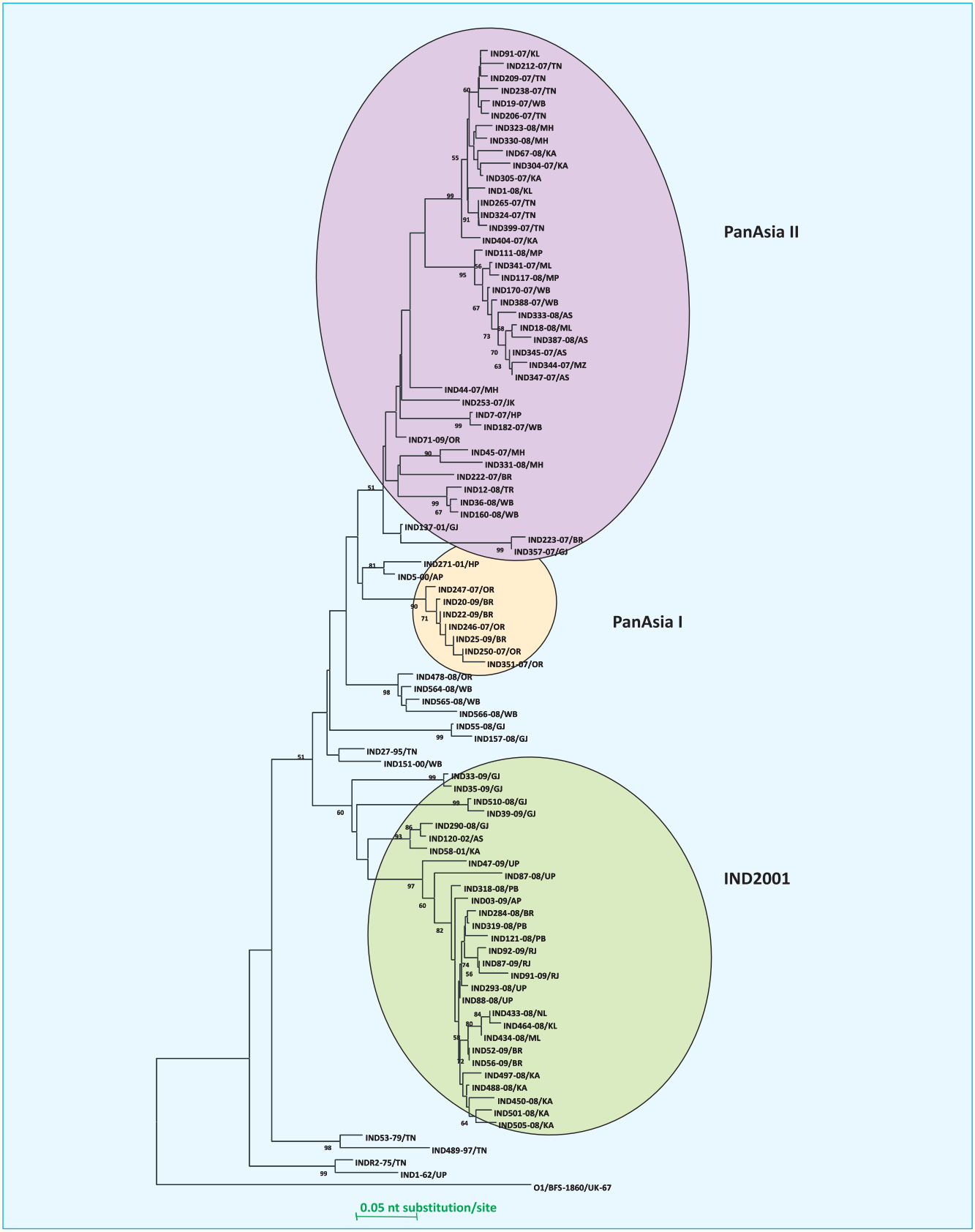


Fig. 2 Neighbour-joining tree depicting phylogenetic relationship among type O isolates at 1D region collected during 2008-09

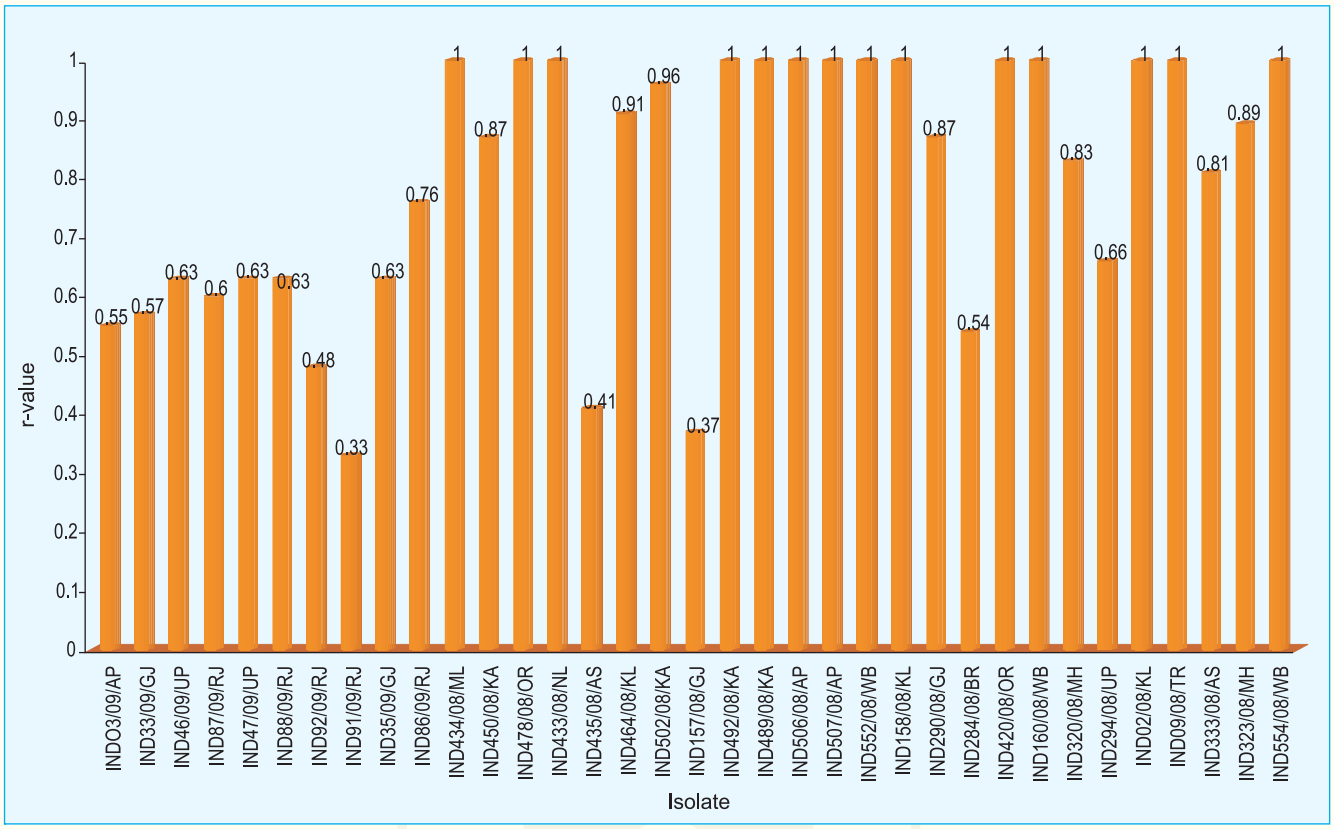


Fig. 3 r-value of Serotype O isolate recovered during 2008-09

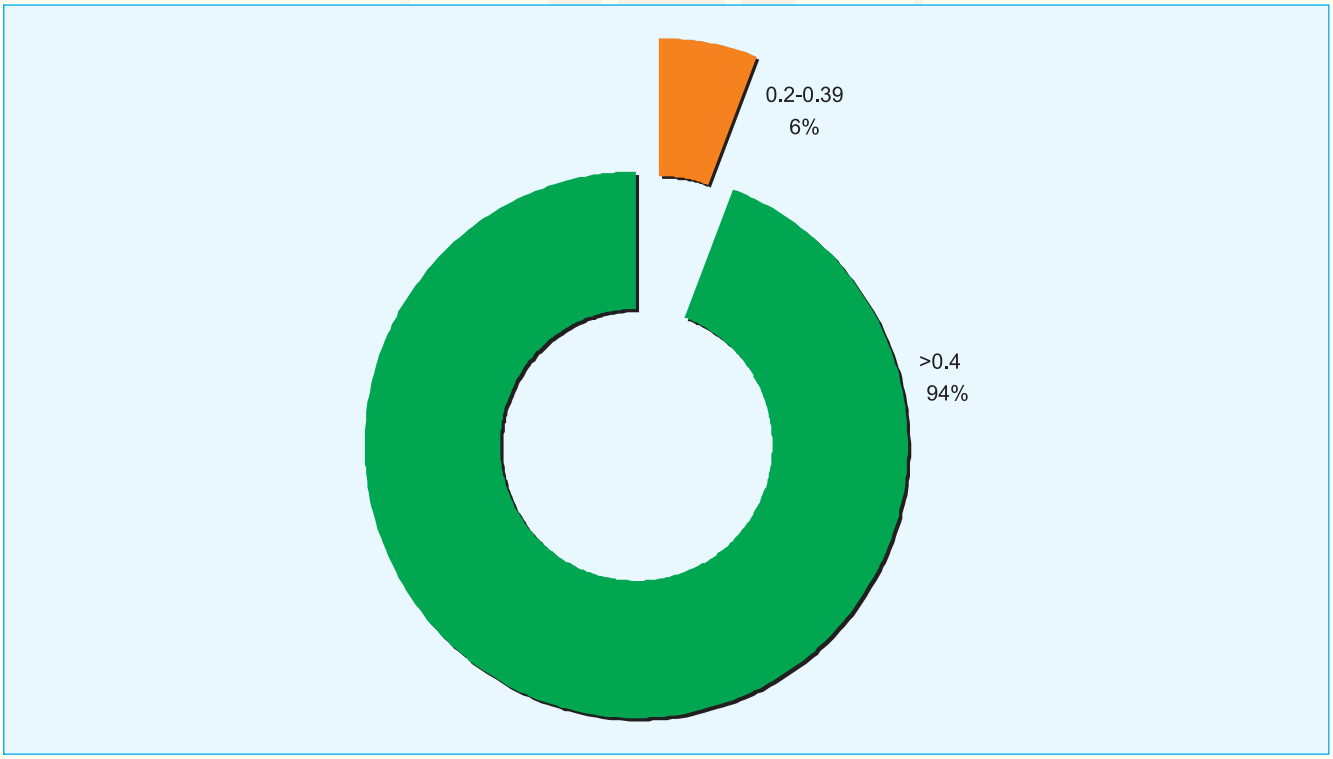


Fig. 4 Pie Diagram showing Percentage of FMD virus serotype O isolates having r-value between 0.2-039 (orange) and >0.4 (green) with in-use vaccine virus (IND R2/75)

6.3.2 Type A

Genetic analysis at VP1 coding (1D) region

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. Recently in 2007-08, there is once again an upsurge in incidence of outbreaks due to this lineage. This single aa deletion is at an antigenically critical position in structural protein VP3, which is considered to be a major evolutionary jump probably due to immune selection.

During the period under report 7 field isolates of serotype A recovered from outbreaks in Karnataka and Kerala were sequenced at complete 1D (VP1) region for molecular epidemiological analysis. For the isolate IND 314/08 from Punjab partial 1D sequence at the 3'end of VP1 was resolved. The determined sequences were aligned with other Indian sequences available in the data base of PD on FMD. All the isolates were found to cluster within genotype VII in the N-J tree (Fig. 5) and precisely in the VP3^{59} deletion group. Hence after its resurgence in 2007, VP3^{59} deletion group continues to dominate the field outbreak scenario even in 2008 and 2009. 1D region based phylogeny has also revealed that this deletion group is genetically diverging with time giving rise to three lineages (VIIb, VIIf & VIIg). All the isolates sequenced here grouped in a single lineage VIIg alongwith the isolates responsible for Tamil Nadu and Karnataka outbreaks during 2007. Within lineage VIIg a maximum nucleotide divergence of 2% was observed which confirms the isolates from Kerala outbreaks are epidemiologically related to those from Karnataka and Tamil Nadu and the type A viruses circulating in the southern peninsula share common ancestry. This VIIg lineage shows 6% nucleotide divergence from other lineages (VIIb and VIIf) of deletion group. A third amino acid in VP1 region was found to be serine in all deletion group isolates in place of threonine or alanine and hence this could be considered an amino acid signature for the deltion group.

Genetic analysis at *nonstructural protein coding regions*

L protease (L^{pro}) coding region

This recent reemergence of VP3^{59} deletion group and genetic diversification has evoked interest and urgency to analyze regions beyond structural protein coding region of this group to verify the earlier prediction of its uniqueness and within group heterogeneity. The 5'-end component of the FMD virus polyprotein is the leader (L^{pro}) protease. L^{pro} is a papain-like protease that has unique cation concentration and pH range requirements. L^{pro}, being involved in cleavage of viral precursor proteins and of host translation factors, prove to be not only important in fundamental virus biology but also crucial for pathogenesis and virulence. Keeping the above facts in view, the complete L^{pro} region sequence of seven isolates from VP3^{59}-deletion group and ten isolates belonging to other genotypes/lineages spanning a period of three decades were resolved for comparative analysis purpose which is supposed to assist the ongoing epidemiological investigations.

Genotype inclusive grouping of Indian type A isolates as observed in 1D region based phylogeny was distorted at complete L^{pro} region

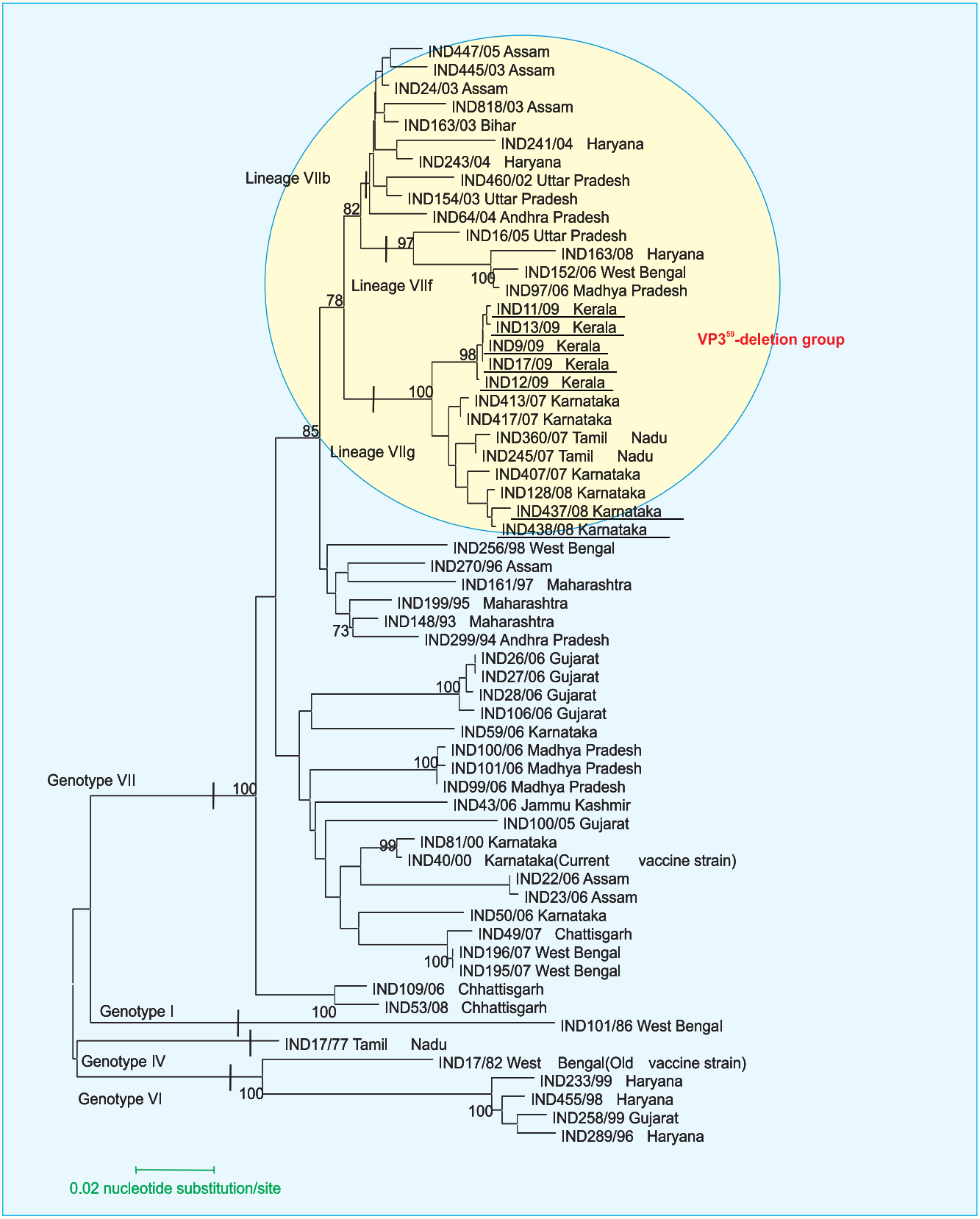


Fig. 5 Neighbour-joining tree depicting phylogenetic relationship among Type A FMD virus isolates at 1D region. Isolates sequenced during the reported year are underlined.

(Fig. 6), where the VP3[59]-deletion group lineages of genotype VII clustered away from both genotypes VII and VI, confirming its uniqueness and independent evolution of L^{pro} and 1D region. Akin to the 1D region, this deletion group is gradually diverging genetically even at L region forming more number of lineages and inter-lineage distance at L region is considerably more than that for 1D region. For instance, IND 288/07 formed a separate lineage (VIIg) from rest of the older isolates within deletion group (VIIb) with more than 0.1 nt substitution/site at L region, while this value was 0.06 for 1D region. The deletion group is restricted to India only as none of the exotic sequences clustered within this group. Notably, L protein exhibited variability comparable to external capsid proteins as evident from its high *d*N/*d*S ratio (0.105), number of variable amino acid positions (41%), lowTs/Tv ratio (3.47) and alignment revealed N-terminal region, β2 sheet and C-terminal extension to be extremely variable. Basic residues at P1, P3 and only leucine at P2 were predicted to provide an optimum autocatalytic cleavage site at L/P1 junction.

All of the eight sites identified to be under positive selection (Table 8) revealed aa substitutions of varied physicochemical properties and at two positions lineage specific signatures were observed (Table 9), which supports the contention that lineages are evolving under differential selection pressure to adapt to the varied ecological environment. Majority of the aa positions identified here are either in the hyper-variable region between the two alternate initiation codons or in the connecting loops or in helices on the surface of the enzyme. Lack of structural constraints imposed by enzyme architecture at the identified regions is reasonably consistent with the radical aa substitutions observed. Though none of these positions were predicted to be critical in terms of functionality of L proteinase in the structural analysis, by virtue of their location either in the N-terminal hyper-variable region thought to be involved in choice of start codon and crucial for IRES activity or in proximity to the catalytic residues/active sites in some of the structural elements, are presumed to be contributing to the enzyme's stability and catalytic function through some hitherto unknown molecular interactions in turn imparting replicative fitness to the virus.

The rate of evolution (synonymous substitution per synonymous site per year) for type A FMDV Lb region was estimated to be 3.6×10^{-2} synonymous substitution/site/year with an R^2 value of 0.9463. The total rate of nt substitution at Lb region was found to be 9×10^{-3} nt substitution/ site/year with an R^2 value of 0.8602 (Fig. 7). The rates estimated for Lb region of serotype A in this study are found to be on a higher side than those for type Asia 1 estimated in an earlier study tempting us to presume that serotype A is evolving faster than type Asia 1 in India at L region. This is supported by the observation that rapid and extensive speciation has been observed for type A in the course of evolution leading to circulation of multiple genotypes and lineages in India. The rate of evolution for VP1 region was estimated to be 1.75×10^{-2} synonymous substitution/site/year and the total rate of nucleotide substitution was found to be 5.6×10^{-3} substitutions/site/year. Hence L region in type A virus population in India is evolving at a rate twice than that of VP1 region in contrary to type Asia 1 study, where a similar rate was predicted for both the regions. This narrow two-fold difference in the rate of evolution of L and VP1 region needs to be evaluated further through rigorous statistical tests.

Table 8. Sites identified to be under positive selection by Datamonkey web interface (SLAC and FEL) of the Hyphy package and CODEML programme (NEB and BEB) of the PAML v.4 package. Position in bold face was detected in all the methods; '–' indicates site not detected by that particular method and ND indicates value not determined.

Sites under positive selection	SLAC method		FEL method			NEB analysis		BEB analysis	
	d_N/d_S	p-value	d_N/d_S	Normalized d_N/d_S	p-value	d_N/d_S (ω)	Posterior probability (p-value)	d_N/d_S (ω)	Posterior probability (p-value)
22L/F/R/S/T	-	-	-	-	-	1.052	0.904	1.316	0.728
23S/L/K/T/P/A	3.268	0.25	3.070	ND	0.372	1.107	0.991	1.493	0.953
25T/P/H/A/I/R/V	1.738	0.39	-	-	-	1.087	0.959	1.343	0.762
49N/K/S/T	3.005	0.396	ND	0.055	0.276	-	-	-	-
57T/A	1.5	0.393	-	-	-	-	-	-	-
86R/K/E/N/G/Q	-	-	-	-	-	0.973	0.778	-	-
114H/Q/N	3.761	0.29	-	-	-	-	-	-	-
156V/I/N	1.488	0.303	ND	0.029	0.173	-	-	-	-

Table 9. Identified genotype/lineage specific signatures in L region. Positions shown in bold are found to be under positive selection in our analysis.

Location	Consensus aa Residue	Lineage specific change	Signature for Group
24	R	K	Lineage VIIb & VIIj
75	D	D	Genotype VII except VP3[59]-deletion group & VIIj
114	H	N	Lineage VIc
145	L	M	Lineage VIIb
156	V	I	Lineage VIc
159	N	D	Genotype VII except VP3[59]-deletion group & VIIj

3A coding region

The 3A region of foot-and-mouth disease virus has been implicated in host range and virulence. Alterations in 3A protein, including point mutations and deletions, have been linked to host specificity, adaptation, attenuation and virulence and some evidence points to its relevance in complex virus–host interactions. This protein colocalizes into the infection induced intracellular membranous RNA replication complex in several picornaviruses and disrupts the normal cellular secretory pathways, thereby offering an immunological advantage to the virus. Virus with truncated forms of 3A had defects in initiating RNA synthesis in specific cell types. Hence 3A is considered one crucial element among the multiple virulence determinants of foot-and-mouth disease virus (FMDV).

Here the 3A region of serotype A virus was sequenced and analyzed in view of the emergence of a variant group in India with an amino acid deletion at an antigenically critical position of capsid protein, VP3. The 3A region exhibited extreme variability with 38% of the amino acid positions showing substitutions and the C-terminal third (127–151) region wasmost flexible. Genotype inclusive grouping of type A foot-and-mouth disease virus as observed in 1D region based phylogeny was much less apparent at 3A region possibly due to independent evolution of nonstructural and structural protein coding regions. Akin to the 1D region, the VP3[59]-deletion

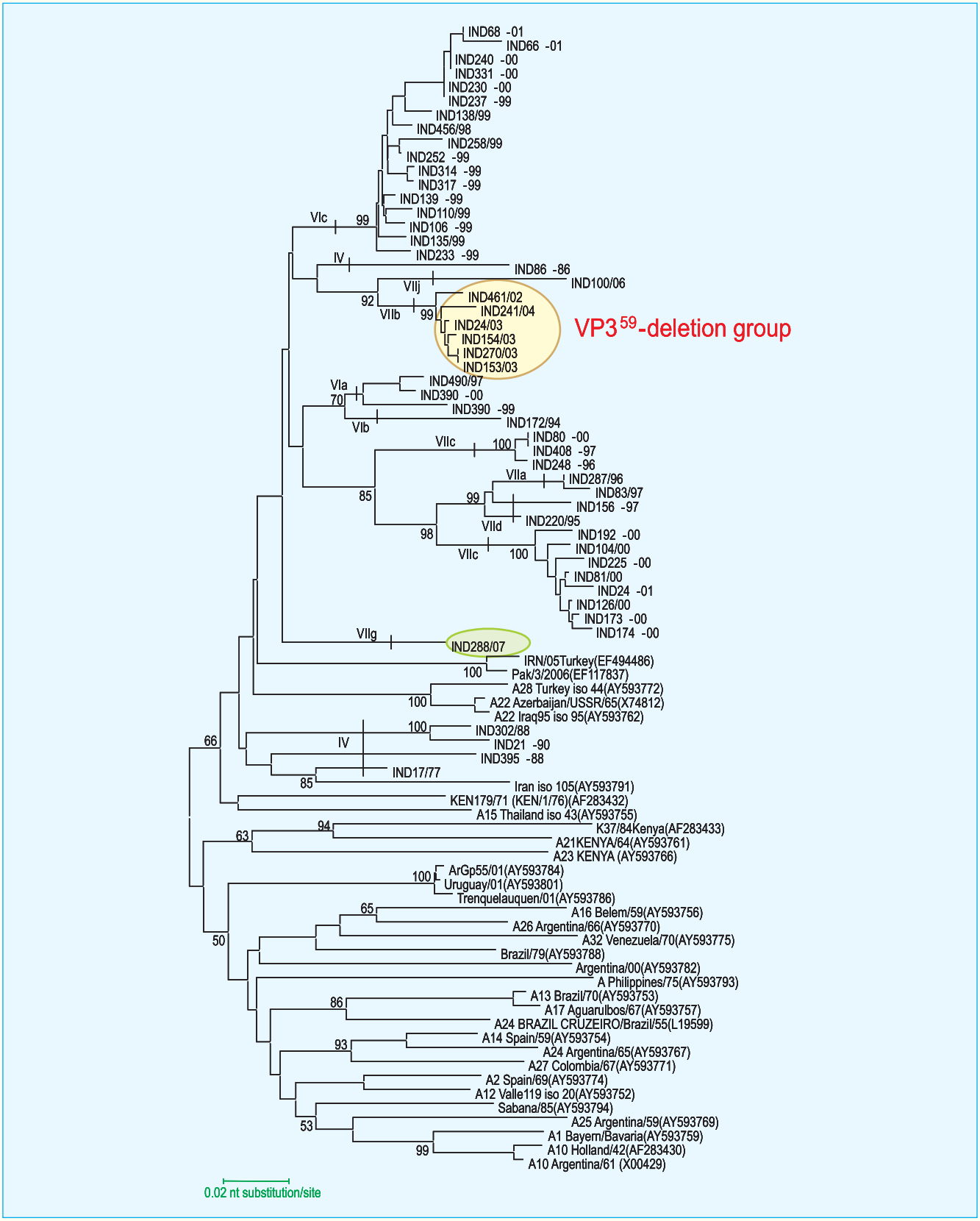
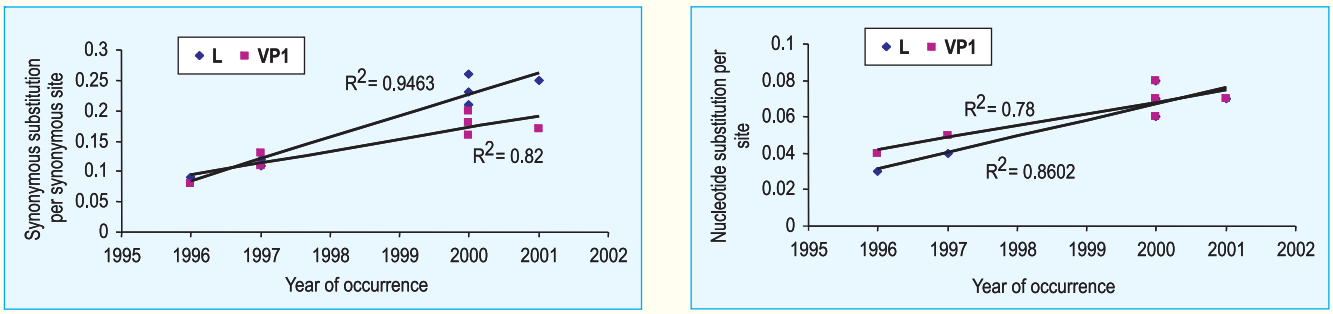


Fig. 6 Neighbour-joining tree depicting phylogenetic relationship among Type A viruses and genotype/lineage distribution at Lb region



(A) (B)

Fig. 7 Linear regression analysis and rate of evolution estimated for type A isolates at L and VP1 region

group maintained its phylogenetic distinctness even at the 3A region and was found to be diverging with time.

Twelve lineage specific signature amino acid residues (Table 10), of which four were identified to be experiencing positive selection, indicates fixation of advantageous mutations in a lineage specific manner. Six positions, all located in the hypervariable C-terminal third, were identified to be under positive selection (Table 11) and were presumed to be imparting the virus certain advantage accounting for its adaptability to wide host spectrum and rapid dissemination. Although factors related to FMD pathogenesis remains an unsolved conundrum, by virtue of their location in the C-terminal hypervariable region experiencing extensive deletions and radical substitutions, these sites under adaptive evolution are presumed to be contributing to 3A's assumed role in virulence, host and cell tropism, viral RNA replication, virus yield and transmission through some unknown molecular interactions with host cellular factors in the context of never-ending dynamics of mutant generation and selection driven fitness testing. A significant change of Q44H was noted only in the older

Table 10. Identified genotype/lineage specific signatures in 3A region

Location	Consensus aa Residue	Lineage specific change	Signature for Group
31	H	S	Lineage VIIc
34	I	V	
114	L	Q	
118	G	S	
144	Q	E	
147	E	R	
84	K	R	Lineage VIc
128	T	V	
145	P	T	
44	Q	H	Lineage VIIb
57	E	D	
138	V	M	Lineage VIIg

Table 11. Site identified to be under positive selection by Datamonkey web interface of the Hyphy package and CODEML programme of the PAML v.4 package

Position	SLAC method		FEL method		FEL method		NEB analysis		BEB analysis	
	dN/dS	*p-value*	*dN/dS*	*p-value*	*dN/dS*	*p-value*	*dN/dS (ω)*	*Posterior probability (p-value)*	*dN/dS (ω)*	*Posterior probability (p-value)*
132H/Q/R/N	-	-	-	-	-	-	1.409	0.965	1.436	0.875
134a/V/M	3.93	0.11	2.70	0.15	2.78	0.19	-	-	-	-
138/V/E/M/A	1.49	0.30	0.05[a]	0.10	0.08[a]	0.06	-	-	-	-
143A/V/I/T	1.49	0.37	-	-	-	-	-	-	-	-
144Q/R/K/G/E/N/V	-	-	-	-	-	-	1.433	0.992	1.494	0.942
151Q/R/H/K	1.88	0.36	0.06[a]	0.10	0.08[a]	0.10	-	-	-	-

[a]Normalized dN-dS values; Positions in bold are detected in earlier analyses; '-' indicates position not detected

lineage (VIIb) of the deletion group at a position where Q44R mutation is associated with guinea pig adaptation. As this site has been detected to be under positive selection, such a lineage specific substitution is thought to have imparted certain temporary advantage to the virus during its possible adaptation in wild or some understudied domestic hosts and must not have seriously compromised fitness upon readaptation in bovines.

A conserved hydrophobic transmembrane domain from position 59 to 76 could be predicted (Fig. 8) which possibly anchors 3A to intracellular membranes for successful interaction with RNA

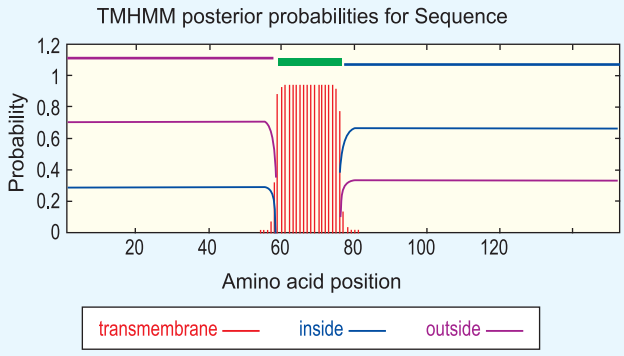


Fig. 8 Posterior probability plot showing transmembrane helix predicted at amino acid position 59-76 of 3A protein with TMHMM programme

replication complex. Based on sequence conservation, the aa regions from 11 to 35 and from 91 to 114, likely expressing T-cell epitopes and linear B-cell epitopes, would be most suitable for designing peptide vaccines and diagnostics, respectively.

Antigenic characterization

A total number of eight type A field isolate recovered from recent outbreaks during 2008-09 were subjected to two dimensional micro-neutralization test (2D-MNT) using bovine vaccinal serum against the current vaccine strain, IND40/00 to elucidate their antigenic relationship with the vaccine strain .

Seven out of eight isolates (87.5%) demonstrated good antigenic relationship with IND 40/00 as reflected from their respective 'r'–value of ≥0.3 (Fig.9). Only one isolate, IND17/09 from an outbreak in Kerala shared an 'r'–value

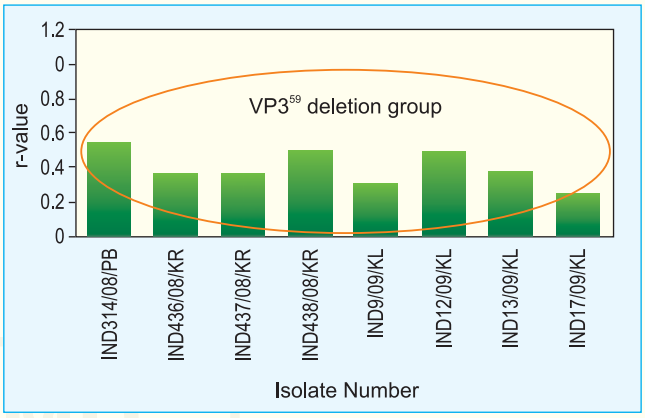


Fig. 9 Antigenic relationship (r-value) of type A isolates recovered during 2008-09 with in-use vaccine strain (IND40/00)

of 0.25. It is interesting to note that other isolates from the same outbreak which are supposedly epidemiologically related strains, shared varied 'r'–value even up to 0.50.

Hence in conclusion, the recent field isolate though belong to a single genetic cluster (VP3[59]-deletion group), show a fair amount of within group antigenic heterogenicity as their 'r'–value profile with the vaccine strain IND40/00 varied between 0.25 and 0.55 in compliance with earlier observations.

Inspite of these subtle variations in the antigenic makeup of field isolates, the new vaccine strain IND 40/00 offers optimum antigenic coverage as 87.5% of the recent isolates were found related to the vaccine strain in the 2D-MNT.

6.3.3 Type Asia 1

Molecular epidemiology

During 2008-09, FMD outbreaks due to serotype Asia1 are reported from the state of West Bengal, Assam, Madhya Pradesh, Rajasthan and Gujarat. The Asia1 filed isolates responsible for those outbreaks were grouped with lineage C (Fig. 10). The lineage C isolates dominated

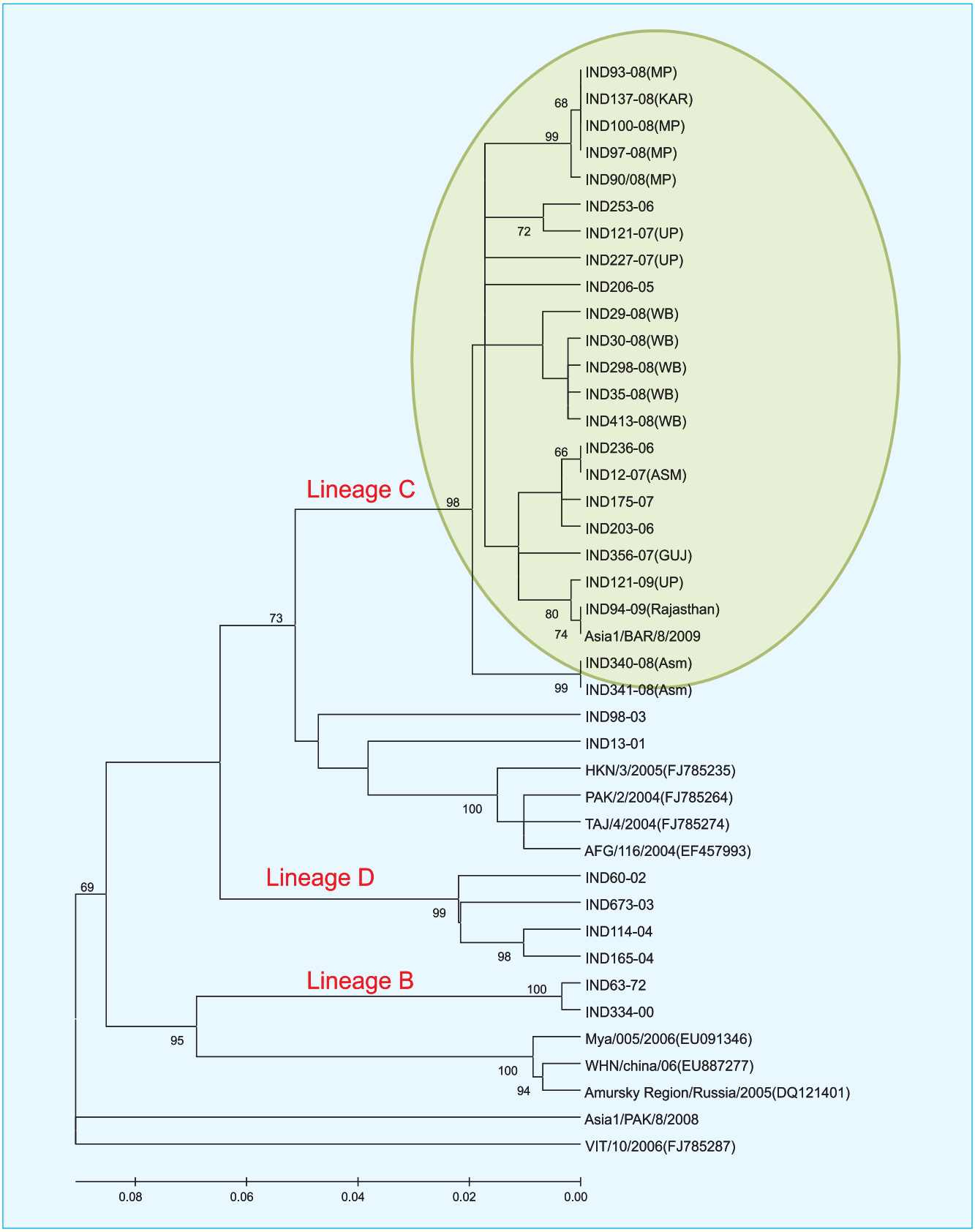


Fig. 10 Neighbour-joining tree depicting phylogenetic relationship among type Asia1 FMD virus isolates at 1D region

the (Asia 1) outbreaks between 1998 and 2002 and reemerged as dominating lineage since 2005. Comparison of Indian FMDV Asia1 isolates with other recent global FMDV Asia1 isolates revealed that one isolate from Bahrain in 2009 clustered with isolates of lineage C from India. Similarly, the isolates recovered from Bhutan in 2002 clustered with isolates of lineage D which dominated Asia1 outbreak between 2002 and 2004. These grouping pattern perhaps give us a hint towards circulation of similar viruses in both the countries at the same time. At nucleotide level, the isolates of lineage C were 9.4 to 14.5% divergent from the isolates of lineage D. The overall nucleotide divergence among the isolates of lineage C ranged from 0.0-12.2% while that with lineage D ranged from 0.0-11.2%. The isolates of lineage C and D were 13.9 to 17.2% divergent at nucleotide level from the in-use vaccine strain. At amino acid level, the isolates of lineage C and lineage D showed a maximum divergence of 10.7% and 7%, respectively among them. The isolates of lineage C and D were 10.2 to 16.5% and 8.6 to 11.6% divergent, respectively from the vaccine strain.

Comparison of deduced amino acid sequence across the VP1 region revealed the presence of lineage specific signature residues identified previously. The isolates of lineage D have unique amino acid substitutions in relation to the lineage C at residues $A_{86}\rightarrow V$, $T_{140}\rightarrow P$, $R/G_{155}\rightarrow E$, $Q_{156}\rightarrow R$, $M_{210}\rightarrow V$ and $M_{211}\rightarrow L$. The integrin receptor binding motif, RGD tri-peptide was found to be conserved in both lineages. Two leucine residues located at positions +1 and +4 downstream to RGD and alanine residue at position +2 known to be important in cell receptor recognition were analyzed. All the field isolates barring IND42/06 have methionine at +1 position instead of leucine (M→I). The leucine at +4 and alanine at +2 were totally conserved in all the isolates except a change from A to T at +2 position in IND 105/06 and IND 108/06. The lineage C and D isolates have alanine at amino acid position 44 whereas isolates of lineage B lack this amino acid residue. Out of the 211 amino acid positions of VP1, 50 positions were found to be variable which include single amino acid replacement at 31 positions and multiple amino acids replacement at 19 positions. Frequency of substitution was higher in βG-βH loop (140-160), βB-βC loop (40-60) and the C terminus of VP1 (200-213) which are known to be antigenically significant in other FMD viruses.

Antigenic analysis

FMDV Asia 1 field viruses isolated from outbreaks in Assam, Rajasthan, Uttar Pradesh and Gujarat were subjected to antigenic analysis using anti-146S bovine serum against the vaccine strain (IND 63/72). All of them showed an r value of more than 0.4 with in-use vaccine strain (Fig. 11) indicating its appropriate antigenic coverage.

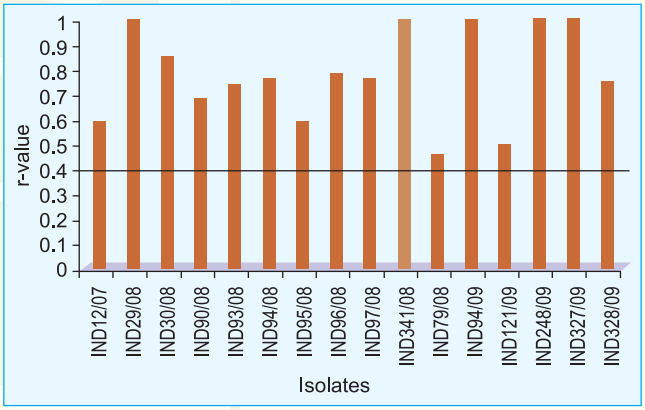


Fig. 11 Antigenic relationship (r-value) of type Asia1 isolates recovered during 2008-09 with in-use vaccine strain (IND 63/72)

6.4 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 53 virus isolates including 41 type O, 3 type Asia1 and 9 type A field isolates were added to the repository. At present this national repository contains a total of 1455 (934-O, 264-Asia 1, 242-A, 15-C) well characterized field isolates.

Table 12. FMDV isolates added in the virus repository during 2008-09

PDFMD ACCESSION NUMBER	STATE FROM WHICH SAMPLE CAME	SEROTYPE OF VIRUS ISOLATED
IND 284/08	BIHAR	Serotype O
IND 290/08	GUJARAT	Serotype O
IND 293/08	UTTAR PRADESH	Serotype O
IND 294/08	UTTAR PRADESH	Serotype O
IND 314/08	PUNJAB	Serotype A
IND 315/08	PUNJAB	Serotype O
IND 317/08	PUNJAB	Serotype O
IND 318/08	PUNJAB	Serotype O
IND 319/08	PUNJAB	Serotype O
IND 320/08	MAHARASHTRA	Serotype O
IND 324/08	MAHARASHTRA	Serotype O
IND 327/08	MAHARASHTRA	Serotype O
IND 328/08	MAHARASHTRA	Serotype O
IND 333/08	ASSAM	Serotype O
IND 341/08	ASSAM	Serotype Asia 1
IND 386/08	ASSAM	Serotype O
IND 387/08	ASSAM	Serotype O
IND 420/08	ORISSA	Serotype O
IND 433/08	NAGALAND	Serotype O
IND 434/08	MEGHALAYA	Serotype O
IND 435/08	ASSAM	Serotype O
IND 436/08	KARNATAKA	Serotype A
IND 437/08	KARNATAKA	Serotype A
IND 438/08	KARNATAKA	Serotype A
IND 450/08	KARNATAKA	Serotype O
IND 464/08	KERALA	Serotype O
IND 478/08	ORISSA	Serotype O
IND 489/08	KARNATAKA	Serotype O
IND 492/08	KARNATAKA	Serotype O
IND 502/08	KARNATAKA	Serotype O
IND 506/08	ANDHRA PRADESH	Serotype O
IND 507/08	ANDHRA PRADESH	Serotype O
IND 552/08	WEST BENGAL	Serotype O
IND 554/08	WEST BENGAL	Serotype O
IND 575/08	WEST BENGAL	Serotype O
IND 9/09	KERALA	Serotype A
IND 11/09	KERALA	Serotype A
IND 12/09	KERALA	Serotype A
IND 13/09	KERALA	Serotype A
IND 03/09	ANDHRA PRADESH	Serotype O
IND 17/09	KERALA	Serotype A
IND 33/09	GUJARAT	Serotype O
IND 35/09	GUJARAT	Serotype O
IND 46/09	UTTAR PRADESH	Serotype O
IND 47/09	UTTAR PRADESH	Serotype O
IND 79/09	GUJARAT	Serotype Asia 1
IND 86/09	RAJASTHAN	Serotype O
IND 87/09	RAJASTHAN	Serotype O
IND 88/09	RAJASTHAN	Serotype O
IND 91/09	RAJASTHAN	Serotype O
IND 92/09	RAJASTHAN	Serotype O
IND 93/09	RAJASTHAN	Serotype O
IND 94/09	RAJASTHAN	Serotype Asia 1

7.0 Applications of DIVA test (rDIVA-FMD Kit) in estimation of FMD prevalence

A recombinant nonstructural protein (3AB3) based ELISA test for differentiation of FMD infected from vaccinated animals (rDIVA-FMD) was developed at Central FMD laboratory, Project Directorate on FMD, Mukteswar. During active viral replication due to FMD virus infection, an array of nonstructural proteins (NSPs) are produced which elicit anti-NSP antibodies, which is not the case in animals which are vaccinated against FMD with an inactivated vaccine. This test is based on the principle of detecting anti-3AB3 NSP antibodies in an indirect ELISA format as a diagnostic marker for FMD infection. This test was conceived during June 2007 and was completed within a year's time during April 2008. This was the need of the hour as no discriminatory test was available in the country to differentiate between vaccinated and infected categories of animals. The indigenously developed rDIVA-FMD kit, amenable to mass scale comprehensive sero-surveillance, is first of its kind in the country and has been designed as per the OIE approved guidelines. This kit has got immense value in detecting evidence of infection and circulation of FMD virus in a herd practicing intensive vaccination in disease endemic regions, to diagnose carrier status postinfection and for import/export serology. The diagnostic sensitivity and specificity of this kit was estimated to be 90% and 95%, respectively. This kit is at least four-fold cheaper than the commercial DIVA kits available in foreign countries. An rDIVA-FMD Kit to test 450 serum samples costs Rs 2500/- only.

A total number of 18,326 random serum samples collected at the rate of 100 per district from 234 districts covering 20 different states of the country were tested in DIVA ELISA in an exercise to estimate FMD prevalence in the country (Table 13). This revealed 27.94 % of the bovine population in the country to be FMD infected during 2008-2009, which might fluctuate consequent upon inclusion of data from bigger states like Uttar Pradesh. State-wise FMD prevalence in bovines varied from 5% in Himachal Pradesh to 46% in Karnataka (Figure 12 & 13). Only 5% infected figure was estimated for the state of Himachal Pradesh complying with the disease outbreak scenario in the state, where no outbreak has been recorded during the last 3 years. In the state of Haryana, where the entire state is covered under FMD vaccination campaign, this figure was found to be only 15 % as against 46 % in Karnataka. This also commensurates the disease picture in Haryana, where in last 5 years only a couple of sporadic FMD cases have been recorded. The per cent infected figure in Haryana is almost half of that of the country's average indicating the effectiveness of the FMD control programmes running in the state and underscores importance of regular vaccination in disease containment. A trend of less than 20% FMDV infected figure was seen in the North Eastern states and Himachal Pradesh which might be ascribed to natural impediments to virus circulation through restricted animal movement due to hilly terrain, closed animal rearing practices in many pockets, geographic isolation etc. rather than intensive vaccination. The all inclusive prevalence figure for India can only be obtained after completion of testing of samples from the remaining states.

Table 13. State-wise FMD prevalence during 2008-2009 in terms of % infected bovines as estimated in DIVA test applied to random serum samples

Sl. No	States	No. of districts covered	Total no. of samples tested	Total no. tested positive	% FMD infected
1	Tripura	04	391	44	11.25
2	Gujarat	06	612	202	33.00
3	Mizoram	08	799	56	7.00
4	Himachal Pradesh	11	957	49	5.12
5	Nagaland	06	195	39	20
6	Bihar	08	558	162	29.03
7	Madhya Pradesh	39	3330	1135	34.08
8	West Bengal	08	657	203	30.89
9	Manipur	05	501	88	17.56
10	Maharashtra	14	1252	466	37.22
11	Punjab	05	155	8	5.16
12	Kerala	09	900	83	9.22
13	AndhraPradesh	17	1700	363	21.35
14	Arunachal Pradesh	11	493	118	23.93
15	Orissa	30	2780	1099	39.53
16	Haryana	18	888	128	14.41
17	Jammu & Kashmir	03	74	23	31.08
18	Rajasthan	09	247	91	36.84
19	Karnataka	19	1477	677	45.84
20	Tamil Nadu	04	360	88	24.44
Grand total for India		234	18326	5122	27.94

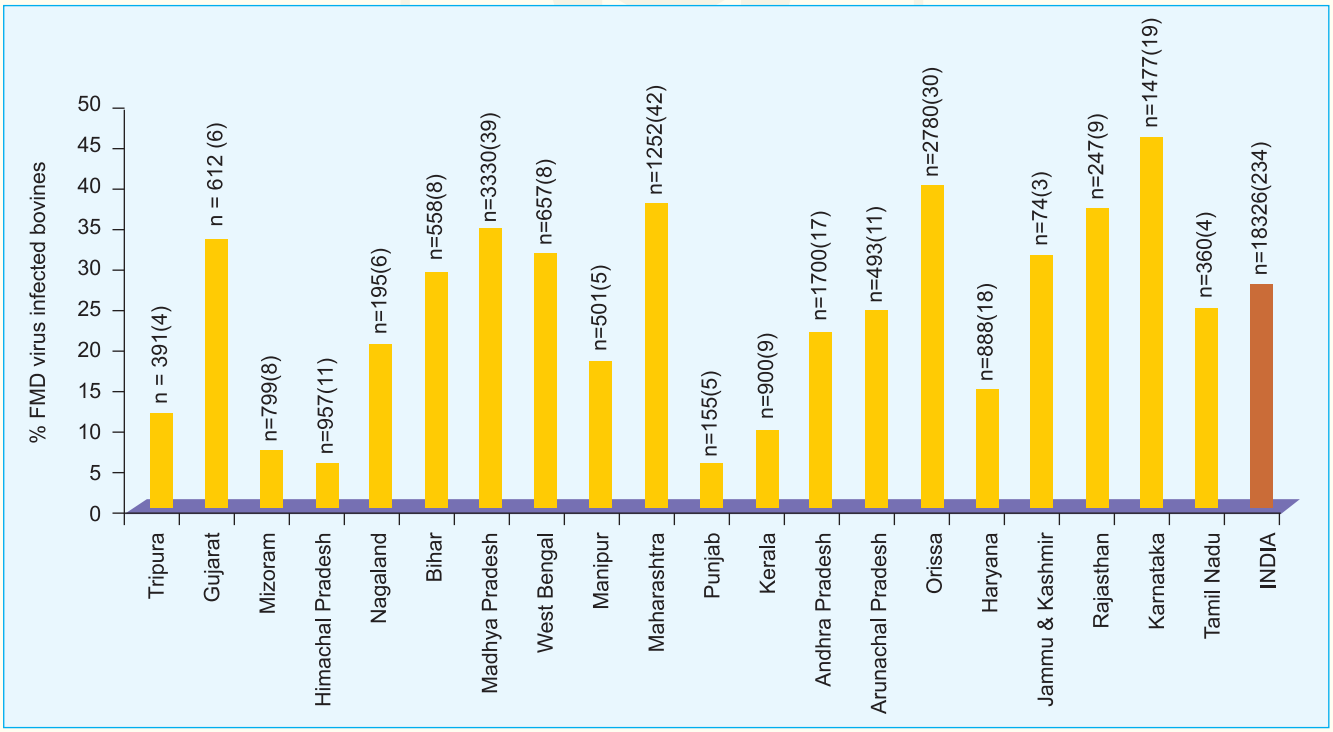


Fig. 12 Prevalence of Foot and Mouth Disease in bovine species during 2008-2009 as estimated in DIVA assay. 'n' indicates number of serum samples screened and within parentheses total number of districts screened are given

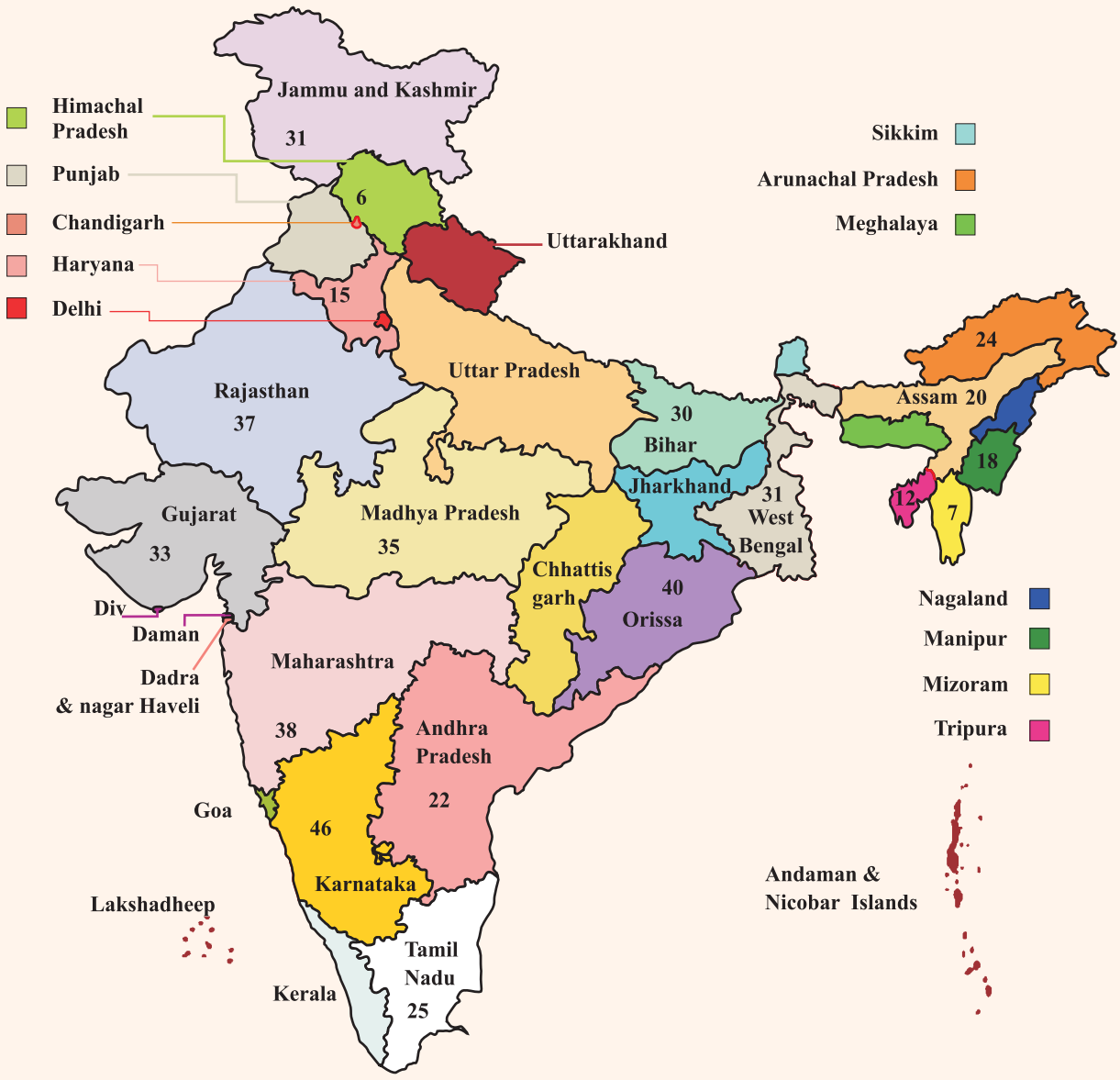


Fig. 13 Per cent FMD virus infected bovines during 2008-2009 depicted on individual state maps as estimated applying DIVA test on random serum samples

8.0 Production, Standardization and Supply of Diagnostic Reagents

FOR production of reagents, the virus {O (IND R2/75), Asia1 (IND 63/72) and A (IND 40/00)} was 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. 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. Kits for testing 7000 clinical materials were supplied to 14 centers/ network units. Kits for testing 80,000 serum samples by LPB-ELISA were produced and supplied to the 15 centers/ network units and other government agencies and industry to ensure uniformity in test results.

DIVA Kit to test a total number of 50,000 serum samples was produced and reagents for a total of 30,000 samples have been supplied so far to the network units and vaccine manufacturing companies for testing random serum samples for final estimation of FMD prevalence, for capacity build up and internal cross validation of the kit.

9.0 Post vaccinal seroconversion studies

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

Foot and Mouth Disease Control Programme (FMD CP) was initiated in 2003 by the Government (DAHD&F) in 54 districts of the country governing 9 States and 1Union Territory (Andaman & Nicobar islands). Vaccination was 100% and done twice a year with trivalent (O, A and Asia1) FMD vaccine. Serum samples of 10 cattle and 10 buffalo 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 (LPB ELISA) developed at central FMD laboratory before launch of the FMD CP in the country. All the 08 regional FMD centers and 1 network unit (Jalandhar) of the Project Directorate participated in the post vaccinal sero-conversion study. All 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 ≥ $\log_{10}$ 1.8 was indicative of protection against FMD. It is noteworthy to mention here that in all FMD outbreaks investigated in detail, the affected had LPB antibody level below $\log_{10}$ 1.8 at 2 to 4 days post clinical sickness.

9.1.1 Andaman & Nicobar

- Under FMDCP eight districts are covered namely, Junglighat, Rangachang, Portmout, Garacharama, Wimberligunj, Monglutan, Elephant Point, Dollygunj.
- Serum samples from A & N were submitted to Kolkata FMD center for testing.
- 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'.

Table 14. Result of seroconversion in Andaman & Nicobar Islands

Phase	Species	Number and % animals showing titres ≥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
I	Cattle+Buff	Serum samples not available					
II	Cattle+Buff	Serum samples not available					
III	Cattle+Buff	40(25.9)	97(60.0)	5(2.8)	37(20.3)	52(34.0)	118(73.6)
IV	Cattle+Buff	50(33.5)	94(64.6)	50(33.5)	96(65.9)	35(23.4)	101(67.6)
V	Cattle+Buff	72(57.2)	68(55.8)	62(50.8)	64(52.5)	54(44.3)	62(50.8)
VI	Cattle+Buff	50 (18.5)	80 (29.6)	66 (24.4)	104 (38.4)	28 (10.2)	36 (13.2)
VII	Cattle+Buff	Serum testing is in progress					

- 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'.
- Testing for phase VII is in progress.

9.1.2 Andhra Pradesh

- Under FMDCP four districts are covered namely, Ananthapur, Chitoor, Medak, Rangareddy.
- Serum samples were submitted to Hyderabad FMD center for testing.
- In phase I, 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 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

Table 15. Result of seroconversion in Andhra Pradesh

Phase	Species	Number and % animals showing titres ≥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
I	Cattle+Buff	83 (10.3)	340 (42.5)	43 (5.3)	244 (30.5)	92 (11.5)	340 (42.5)
II	Cattle+Buff	N.A.	434 (54.2)	N.A.	498 (62.3)	N.A.	438 (54.7)
III	Cattle+Buff	210 (26.2)	286 (35.7)	395 (49.3)	532 (66.5)	306 (38.2)	422 (52.7)
IV	Cattle+Buff	281 (35.1)	374 (46.8)	465 (58.1)	617 (77.1)	329 (41.1)	518 (64.8)
V	Cattle+Buff	247 (30.8)	440 (55)	466 (58.2)	574 (71.8)	343 (42.8)	450 (56.3)
VI	Cattle+Buff	275 (34.3)	490 (61.3)	554 (69.2)	690 (86.3)	446 (55.7)	634 (79.3)
VII	Cattle+Buff	Serum testing is in progress					

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'.

- Testing for phase VII is in progress.

9.1.3 Delhi

- Serum samples were submitted to Hissar FMD center for testing.
- For phase I & II, serum samples from buffalo were available; for phase III, IV, V and VI the serum samples from both buffalo and cattle were tested.
- In phase I, 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 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 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

Table 16. Result of seroconversion in Delhi

Phase	Species	Number and % animals showing titres ≥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
I	Cattle+Buff	26 (53)	50 (100)	13 (26)	47 (94)	17 (34)	48 (96)
II	Cattle+Buff	22 (91)	23 (96)	12 (40)	15 (62)	23 (95)	22 (86)
III	Cattle+Buff	47 (94)	49 (98)	30 (60)	40 (80)	43 (86)	46 (92)
IV	Cattle+Buff	38 (76)	38 (82.6)	14 (28)	40 (86.9)	27 (54)	41 (89.1)
V	Cattle+Buff	26 (59)	47 (88.6)	23 (52.2)	37 (69.8)	32 (72.7)	41 (77.3)
VI	Cattle+Buff	76 (77.5)	97 (98.9)	60 (61.2)	93 (94.9)	71 (72.4)	97 (98.9)
VII	Cattle+Buff	Serum testing is in progress					

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'.

- During last three phases, persistence of antibodies in pre-vaccination serum samples was more than 50% against all the three serotypes barring against type A in phase IV.
- Vaccination programme in Delhi region achieved and crossed 80% herd immunity level after six phases of vaccination.
- Testing for phase VII is in progress.

9.1.4 Gujarat

- Under FMDCP four districts are covered namely, Banaskantha, Sabarkantha, Mehsana and Patan.
- Serum samples were submitted to Pune FMD center for testing.
- 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 27.8for 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'.
- From VIth phase onwards it was decided that samples will be tested by Ahmedabad center. 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'.
- Testing for phase VII is in progress.

Table 17. Result of seroconversion in Gujarat

Phase	Species	Number and % animals showing titres ≥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
I	Cattle+Buff	50 (19.1)	116 (44.7)	59 (24.5)	128 (48.7)	42 (16.1)	114 (43.5)
II	Cattle+Buff	Serum samples not available					
III	Cattle+Buff	123 (27.8)	171 (47.9)	171 (39.2)	268 (58.3)	51 (12.4)	149 (35.4)
IV	Cattle+Buff	113 (22.7)	277 (60.7)	184 (40.7)	355 (81.2)	73 (14.6)	218 (46.8)
V	Cattle+Buff	46 (23.6)	99 (49.0)	126 (66.1)	179 (91.6)	44 (26.5)	92 (51.3)
VI	Cattle+Buff	119 (30.1)	223 (56.4)	249 (63.0)	317(80.2)	195 (49.3)	240 (60.7)
VII	Cattle+Buff	Serum testing is in progress					

9.1.5 Haryana

- Under FMDCP eight districts are covered namely, Bhiwani, Fatehabad, Hisar, Jhajjar, Jind, Rohtak, Sirsa, Sonipat
- Serum samples were submitted to Hissar FMD center for testing.
- Serum samples were not available for Phase I.

Table 18. Result of seroconversion in Haryana

Phase	Species	Number and % animals showing titres ≥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
I	Cattle+Buff	Serum samples not available					
II	Cattle+Buff	NA	1065(68.3)	NA	859 (55.1)	NA	831 (53.3)
III	Cattle+Buff	NA	1146(72.3)	NA	1007(63.6)	NA	1005(63.4)
IV	Cattle+Buff	953 (60.1)	1222(78.7)	668 (42.1)	887 (57.1)	844(53.2)	1170(75.3)
V	Cattle+Buff	955 (59.7)	1352(84.5)	813 (50.8)	1274(79.6)	941(58.8)	1353(84.5)
VI	Cattle+Buff	995 (66.5)	1306(87.1)	895 (59.8)	1229(82.0)	844(56.4)	1118(74.6)
VII	Cattle+Buff	Serum testing is in progress					

- 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 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 58.8for 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'.
- Testing for phase VII is in progress.

9.1.6 Kerala

- Three districts namely, Trivandrum, Kollam and Pathanamthitta are covered under FMDCP.
- Serum samples are tested by Ranipet Centre.
- 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.

Table 19. Result of seroconversion in Kerala

Phase	Species	Number and % animals showing titres ≥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
I	Cattle+Buff	158(32.7)	255(51.4)	140(29.0)	236(47.5)	165(34.2)	280(56.4)
II	Cattle+Buff	158(32.7)	255(51.4)	140(29.0)	236(47.5)	165(34.2)	280(56.4)
III	Cattle+Buff	Serum samples not available					
IV	Cattle+Buff	158(32.7)	255(51.4)	140(29.0)	236(47.5)	165(34.2)	280(56.4)
V	Cattle+Buff	67(23.1)	197(67.9)	52(17.9)	171(58.9)	61(21.0)	211(72.7)
VI	Cattle+Buff	49 (20.4)	185(77.1)	41(17.1)	169(70.4)	38(15.8)	171(71.3)
VII	Cattle+Buff	Serum testing is in progress					

- 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'.
- Testing for phase VII is in progress.

9.1.7 Maharashtra

- Under FMDCP six districts are covered namely, Ahmadnagar, Aurangabad, Pune, Satara, Mumbai and Thane.
- 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, 334 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 22.4 for type 'O', 71.2 for type 'A' and 40 for type 'Asia-1'. The same for post-vac samples was 51.5 for type 'O', 93.3 for type 'A' and 61.8 for type 'Asia-1'.

- For phase VII, serum testing is in progress.
- 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'.
- 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 40.27 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'.
- Phase IV, serum samples are being tested at Bangalore & Guwahati FMD centre.
- 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'.

Table 20. Result of seroconversion in Maharashtra

Phase	Species	Number and % animals showing titres ≥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
I	Cattle+Buff	173 (20.5)	456 (59.9)	151(17.9)	437 (57.4)	192 (22.8)	466 (61.2)
II	Cattle+Buff	N.A.	508 (60.9)	N.A.	490 (58.6)	N.A.	553 (66.2)
III	Cattle+Buff	184 (24.4)	438 (54.8)	351 (46.8)	580 (72.7)	262 (34.7)	534 (66.9)
IV	Cattle+Buff	191 (24.2)	417 (52.3)	517 (65.6)	679 (85.3)	278 (35.2)	509 (63.9)
V	Cattle+Buff	142 (17.7)	271 (35.1)	353 (44.2)	477 (62.3)	121 (15.0)	245 (31.8)
VI	Cattle+Buff	75 (22.4)	170 (51.5)	238 (71.2)	308 (93.3)	134 (40.0)	204 (61.8)
VII	Cattle+Buff	Serum testing is under progress					
VIII	Cattle+Buff	646(64.6)	904(90.4)	574(57.4)	848(84.8)	198(19.8)	452(45.2)

9.1.8 Punjab

- Under FMDCP the districts covered are namely, Amritsar, Bhatinda, Fatehgarh Sahib, Ferozpur, Mansa, Sangrur, Jalandhar, Patiala and Gurdaspur.
- Phase I and II serum samples tested at Kolkata FMD center.
- Phase III and IV serum samples are being tested at Bangalore FMD Centre.
- Phase IV and V serum samples are being tested at Guwahati FMD Center.
- Phase VI samples tested at Jalandhar network unit.

- In phase VI, 1578 pre and 1504 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 44.72 for type 'O', 45.13 for type 'A' and 35.8 for type 'Asia-1'. The same for post-vac samples was 61.12 for type 'O', 58.7 for type 'A' and 50.8 for type 'Asia-1'.

- 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'.

Table 21. Result of seroconversion in Punjab

Phase	Species	Number and % animals showing titres ≥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
I	Cattle+Buff	N.A.	187(25.2)	N.A.	90(11.5)	N.A.	273(49.5)
II	Cattle+Buff	N.A.	219(43.8)	N.A.	113(20.9)	N.A.	279(58.1)
III	Cattle+Buff	915(84.4)	1175(86.1)	816(75.3)	1007(73.8)	437(40.27)	573(42.0)
IV	Cattle+Buff	Serum testing is in progress					
V	Cattle+Buff	477(34.8)	621(54.5)	445(32.8)	630(53.7)	513(38.5)	690(60.1)
VI	Cattle+Buff	706(44.7)	919(61.1)	712(45.1)	883(58.7)	565(35.8)	764(50.8)
VII	Cattle+Buff	165(35.9)	116(65.9)	129(28.1)	110(62.5)	159(34.6)	99(56.3)
VIII	Cattle+Buff	Serum testing is in progress					

- For VII out of 3084 samples 635 have been tested. In tested samples the percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 35.9 for type 'O', 28.1 for type 'A' and 34.6 for type 'Asia-1'. The same for post-vac samples was 65.9 for type 'O', 62.5for type 'A' and 56.3 for type 'Asia-1'.
- Testing for phase VIII is in progress.

9.1.9 Tamil Nadu

- Kanyakumari district is covered under FMDCP.
- Serum samples are 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'.
- 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'.
- In phase VII, 300 pre and 300 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and

Table 22. Result of seroconversion in Tamil Nadu

Phase	Species	Number and % animals showing titres ≥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
I	Cattle+Buff	28(28)	51(51)	29(29)	57(57)	24(24)	54(54)
II	Cattle+Buff	23(23.0)	63(63.0)	24(24.0)	40(40.0)	18(18.0)	61(61.0)
III	Cattle+Buff	59(32.7)	246(74.5)	61(33.8)	201(60.9)	45(25.0)	216(65.4)
IV	Cattle+Buff	59(32.7)	246(74.5)	61(33.8)	201(60.9)	45(25.0)	216(65.4)
V	Cattle+Buff	Serum samples not available					
VI	Cattle+Buff	30(18.7)	99(76.1)	31(23.8)	109(83.8)	28(21.5)	103(79.2)
VII	Cattle+Buff	35(11.7)	210(70)	34(11.3)	231(77)	36(12)	226(75.3)

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'.

9.1.10 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 during phases I to VI.
- 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 1st phase.
- In phase II, 139 and 402 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 100 for type 'O', 100 for type 'A' and 100 for type 'Asia-1'. The same for post-vac samples was 92 for type 'O', 94.2 for type 'A' and 97.1 for type 'Asia-1'.
- In phase III, 1137 and 1577 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 79.1 for type 'O', 85.4 for type 'A' and 82.7 for type 'Asia-1'. The same for post-vac samples was 85.4 for type 'O', 87.3 for type 'A' and 88.5 for type 'Asia-1'.
- In phase IV, 1730 and 1517 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 66.2 for type 'O', 76.9 for type 'A' and 80.5 for type 'Asia-1'. The same for post-vac samples was 63.2 for type 'O', 78.8 for type 'A' and 72.8 for type 'Asia-1'.
- In phase V, 1459 pre and 1207 post-vac serum samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 50.3 for type 'O', 50.5 for type 'A' and 55.9 for type 'Asia-1'. The same for post-vac samples was 62.2 for type 'O', 66.9 for type 'A' and 65.7 for type 'Asia-1'.
- Serum testing is in progress for sixth phase.

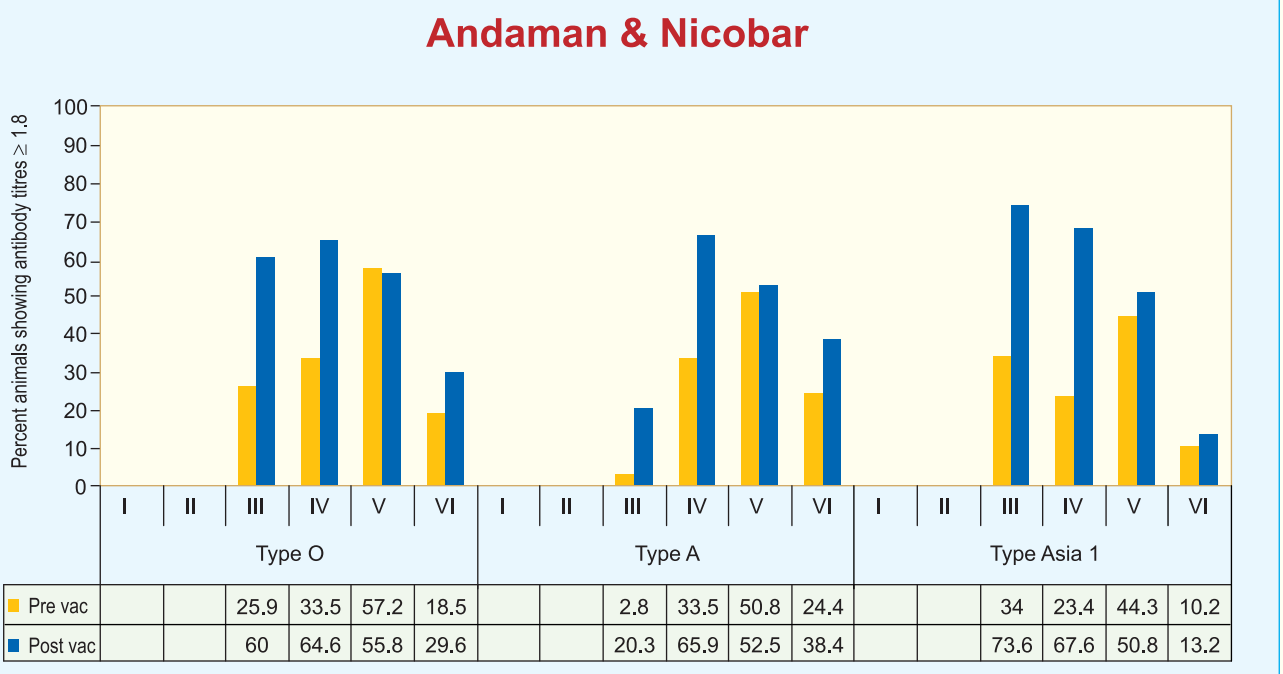
In phase VII at present 1085 pre and post vac serum samples out of total 2095 collected

Table 23. Result of seroconversion in Uttar Pradesh

Phase	Species	Number and % animals showing titres ≥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
I	Cattle+Buff	Serum samples not available					
II	Cattle+Buff	139(100)	370(92.0)	139(100)	384(94.2)	139(100)	399(97.1)
III	Cattle+Buff	900(79.1)	1347(85.4)	980(85.4)	1377(87.3)	942(82.7)	1398(88.5)
IV	Cattle+Buff	1145(66.2)	959(63.2)	1395(76.9)	1299(78.8)	1451(80.5)	1167(72.8)
V	Cattle+Buff	734(50.3)	751(62.2)	810(50.5)	835(66.9)	821(55.9)	816(65.7)
VI	Cattle+Buff	Serum testing is in progress					
VII	Cattle+Buff	350(32.3)	400(55.3)	270(24.9)	366(50.6)	233(21.5)	367(50.8)
VIII	Cattle+Buff	Serum testing is in progress					

samples were tested. Percent serum sample having protective antibody titer of 1.8 and above for pre-vac samples was 32.3 for type 'O', 24.9 for type 'A' and 21.5 for type 'Asia-1'. The same for post-vac samples was 55.3 for type 'O', 50.6 for type 'A' and 50.8 for type 'Asia-1'.

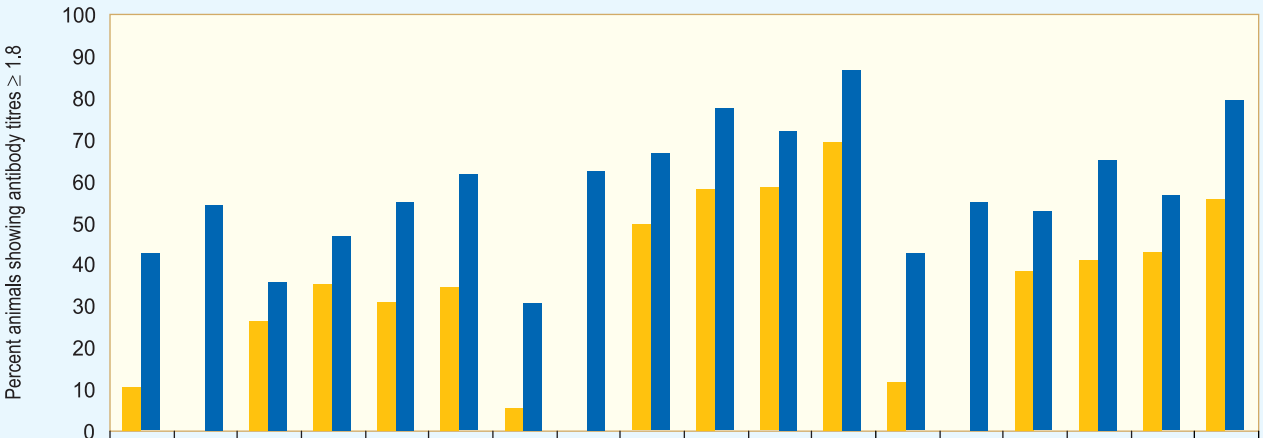
9.1.11 FMDCP Summary (State-wise). Percent animals showing antibody titer ≥1.8 log_{10} against FMD virus (post vaccination) in different states



*Serum samples were not available for Phases I & II

Fig. 14 Seroconversion in Andaman & Nicobar

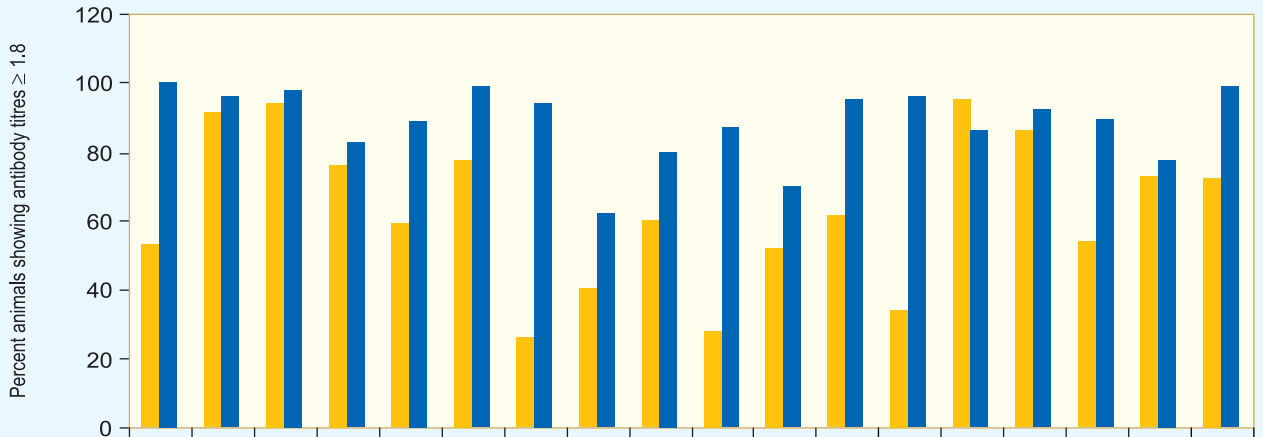
Andhra Pradesh



	Type O						Type A						Type Asia 1					
	I	II	III	IV	V	VI	I	II	III	IV	V	VI	I	II	III	IV	V	VI
Pre vac	10.3		26.2	35.1	30.8	34.3	5.3		49.3	58.1	58.2	69.2	11.5		38.2	41.1	42.8	55.7
Post vac	42.5	54.2	35.7	46.8	55	61.3	30.5	62.3	66.5	77.1	71.8	86.3	42.5	54.7	52.7	64.8	56.3	79.3

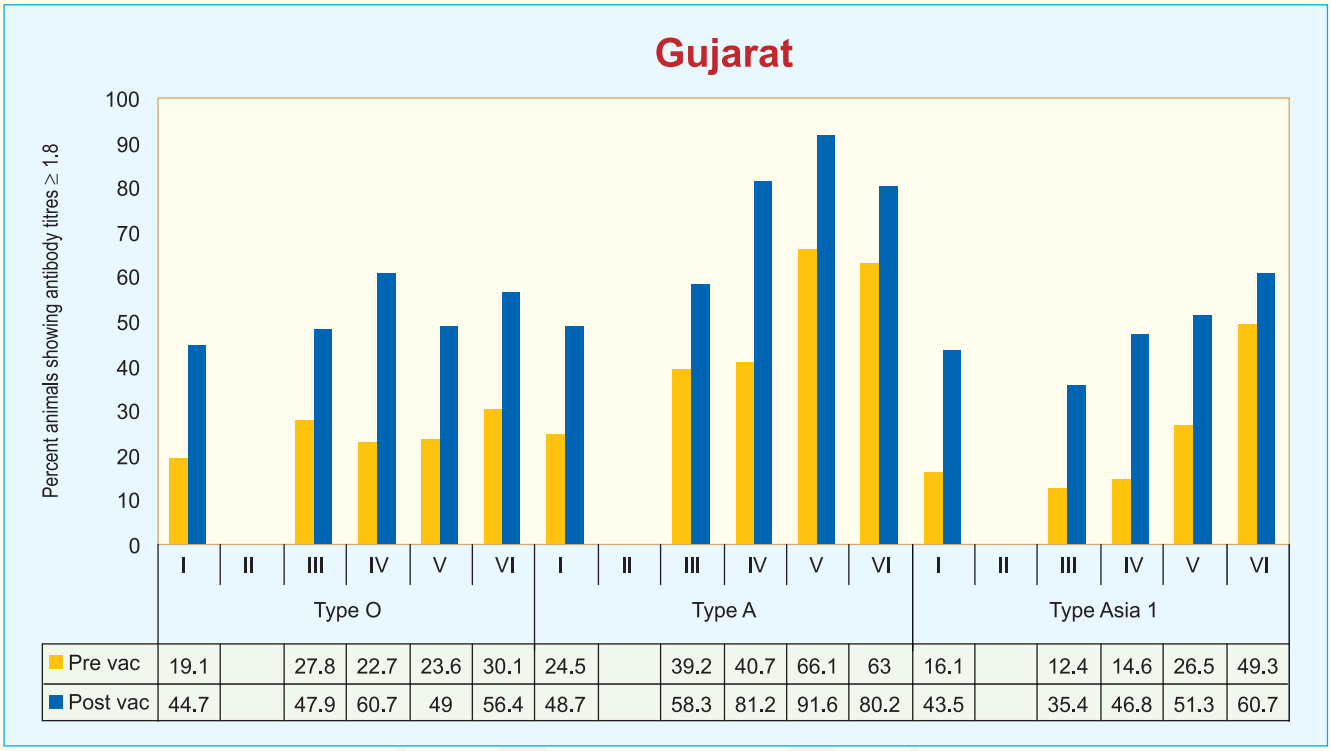
Fig. 15 Seroconversion in Andhra Pradesh

Delhi



	Type O						Type A						Type Asia 1					
	I	II	III	IV	V	VI	I	II	III	IV	V	VI	I	II	III	IV	V	VI
Pre vac	53	91	94	76	59	77.5	26	40	60	28	52.2	61.2	34	95	86	54	72.7	72.4
Post vac	100	96	98	82.6	88.6	98.9	94	62	80	86.9	69.8	94.9	96	86	92	89.1	77.3	98.9

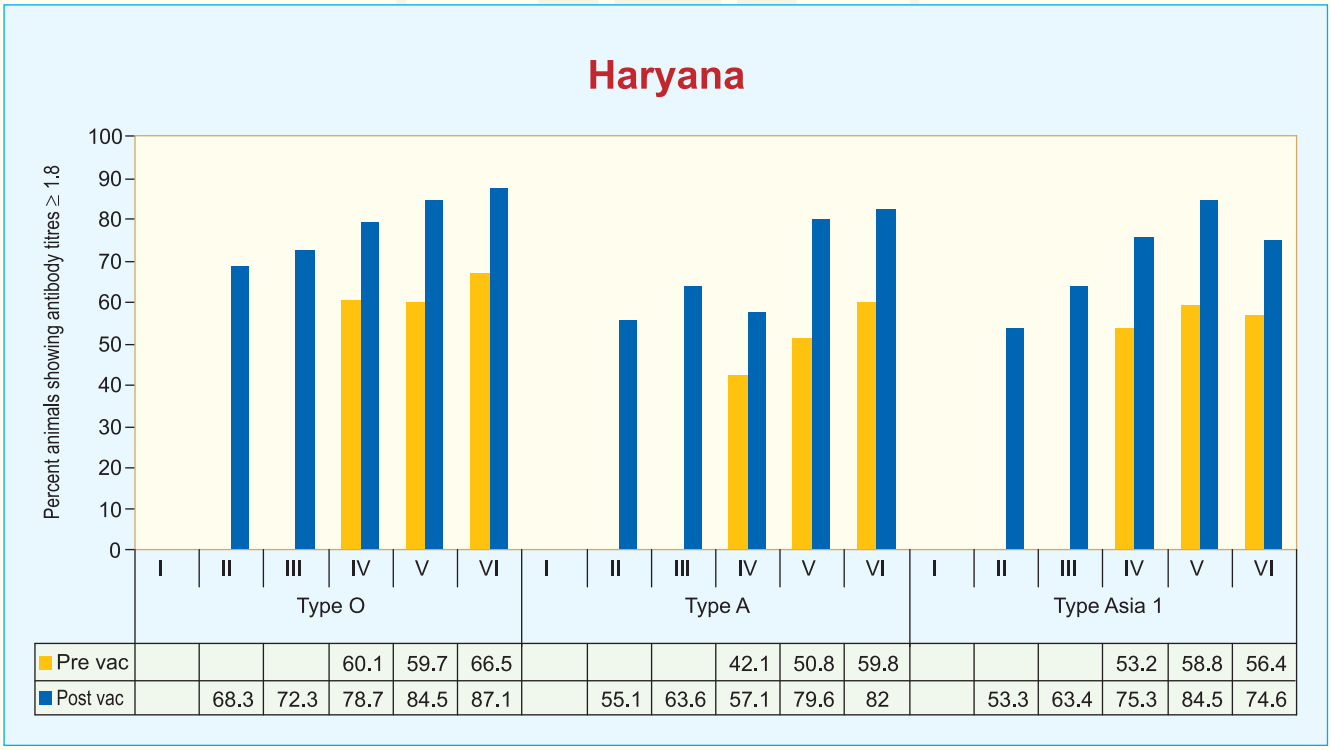
Fig. 16 Seroconversion in Delhi



	Type O						Type A						Type Asia 1					
	I	II	III	IV	V	VI	I	II	III	IV	V	VI	I	II	III	IV	V	VI
Pre vac	19.1		27.8	22.7	23.6	30.1	24.5		39.2	40.7	66.1	63	16.1		12.4	14.6	26.5	49.3
Post vac	44.7		47.9	60.7	49	56.4	48.7		58.3	81.2	91.6	80.2	43.5		35.4	46.8	51.3	60.7

*Serum samples were not available for Phase II

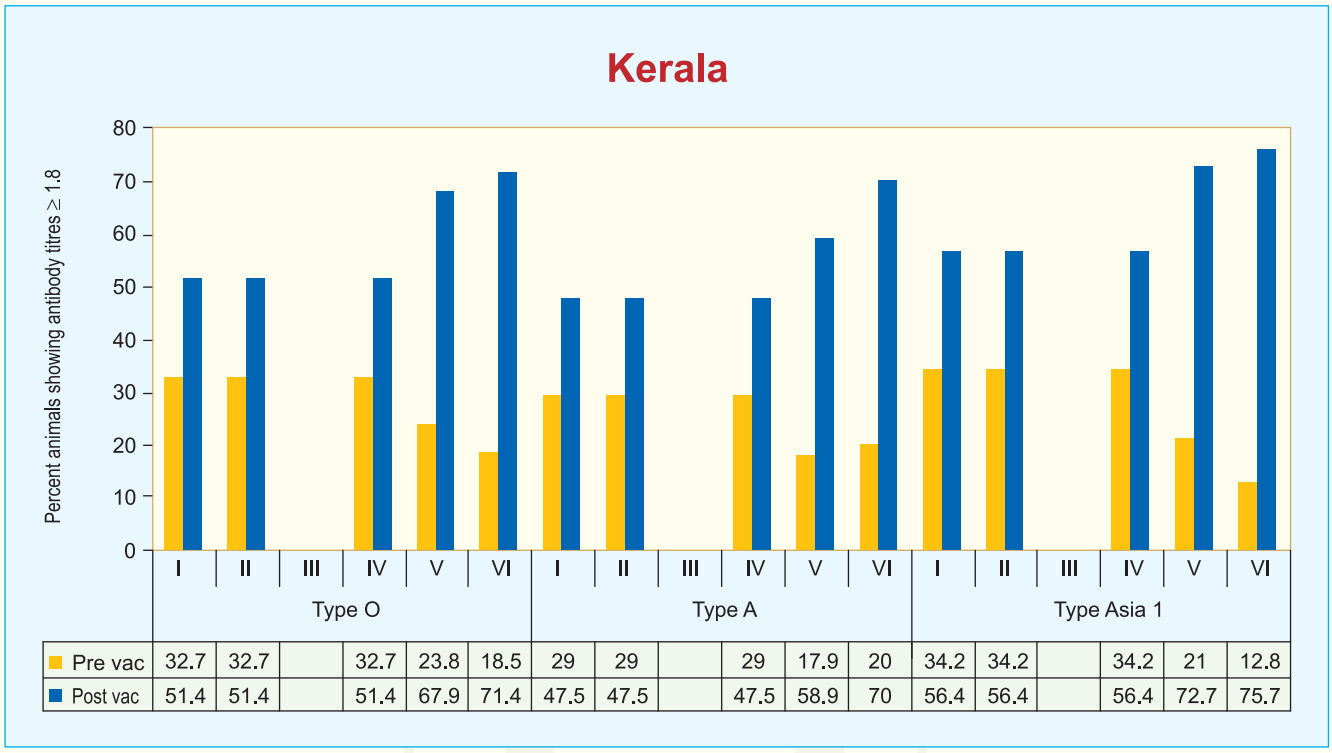
Fig. 17 Seroconversion in Gujarat



	Type O						Type A						Type Asia 1					
	I	II	III	IV	V	VI	I	II	III	IV	V	VI	I	II	III	IV	V	VI
Pre vac				60.1	59.7	66.5				42.1	50.8	59.8				53.2	58.8	56.4
Post vac		68.3	72.3	78.7	84.5	87.1		55.1	63.6	57.1	79.6	82		53.3	63.4	75.3	84.5	74.6

*Serum samples were not available for Phase I

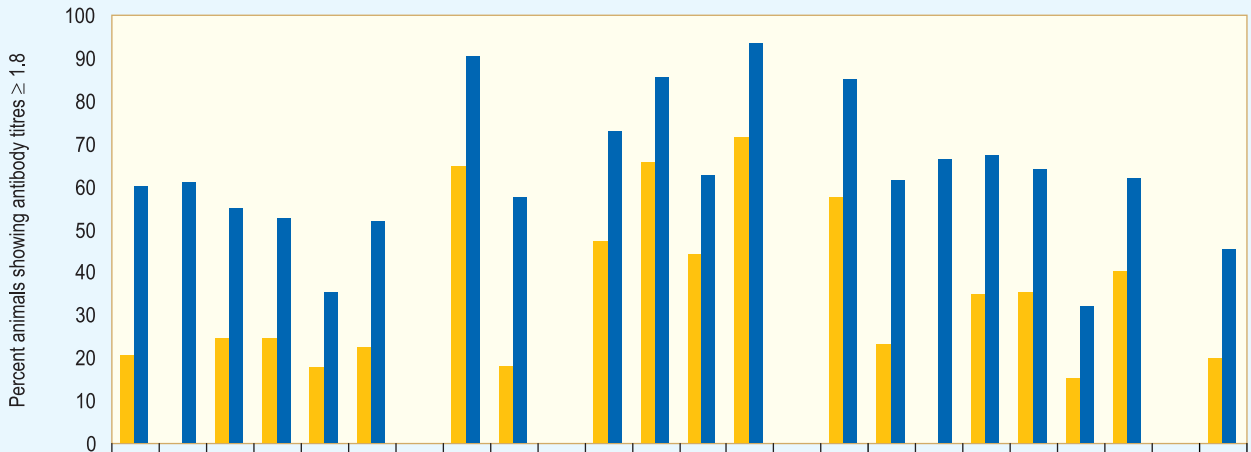
Fig. 18 Seroconversion in Haryana



	I	II	III	IV	V	VI	I	II	III	IV	V	VI	I	II	III	IV	V	VI
	Type O						Type A						Type Asia 1					
Pre vac	32.7	32.7		32.7	23.8	18.5	29	29		29	17.9	20	34.2	34.2		34.2	21	12.8
Post vac	51.4	51.4		51.4	67.9	71.4	47.5	47.5		47.5	58.9	70	56.4	56.4		56.4	72.7	75.7

*Serum samples were not available for Phase III

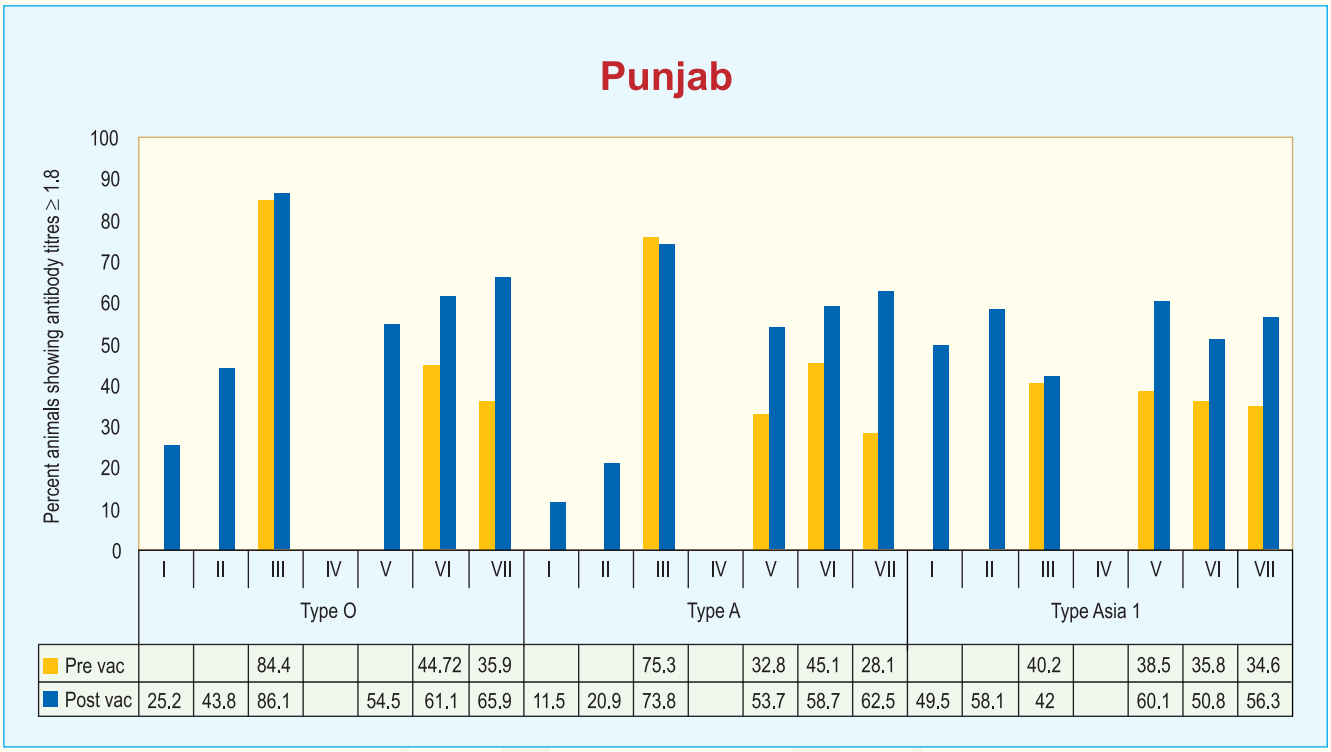
Fig. 19 Seroconversion in Kerala



	I	II	III	IV	V	VI	VII	VIII	I	II	III	IV	V	VI	VII	VIII	I	II	III	IV	V	VI	VII	VIII
	Type O								Type A								Type Asia 1							
Pre vac	20.5		24.4	24.2	17.7	22.4		64.6	17.9		46.8	65.6	44.2	71.2		57.4	22.8		34.7	35.2	15	40		19.8
Post vac	59.9	60.9	54.8	52.3	35.1	51.5		90.4	57.4	58.6	72.7	85.3	62.3	93.3		84.8	61.2	66.2	66.9	63.9	31.8	61.8		45.2

*Serum samples for Phase VII under testing

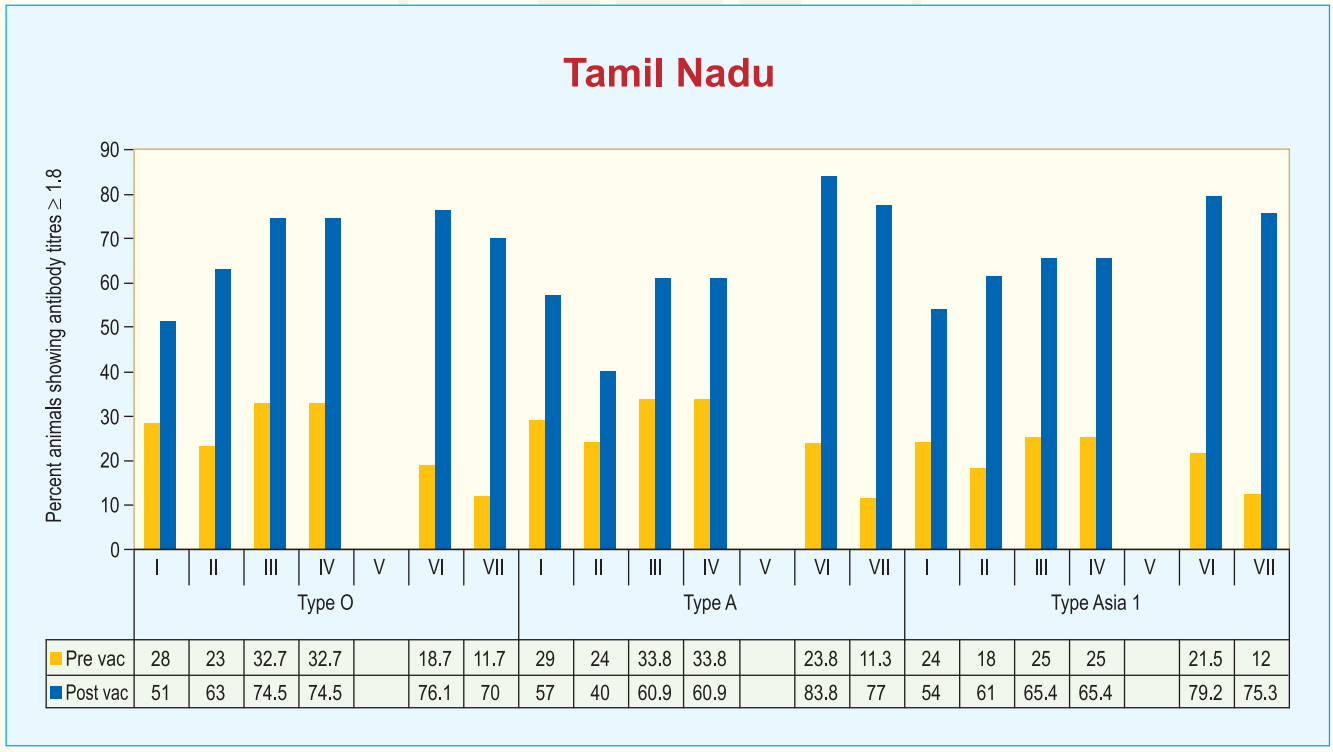
Fig. 20 Seroconversion in Maharashtra



	Type O							Type A							Type Asia 1						
	I	II	III	IV	V	VI	VII	I	II	III	IV	V	VI	VII	I	II	III	IV	V	VI	VII
Pre vac			84.4			44.72	35.9			75.3		32.8	45.1	28.1			40.2		38.5	35.8	34.6
Post vac	25.2	43.8	86.1		54.5	61.1	65.9	11.5	20.9	73.8		53.7	58.7	62.5	49.5	58.1	42		60.1	50.8	56.3

*Sample testing is in progress for Phase IV at Bangalore and Guwahati regional centres

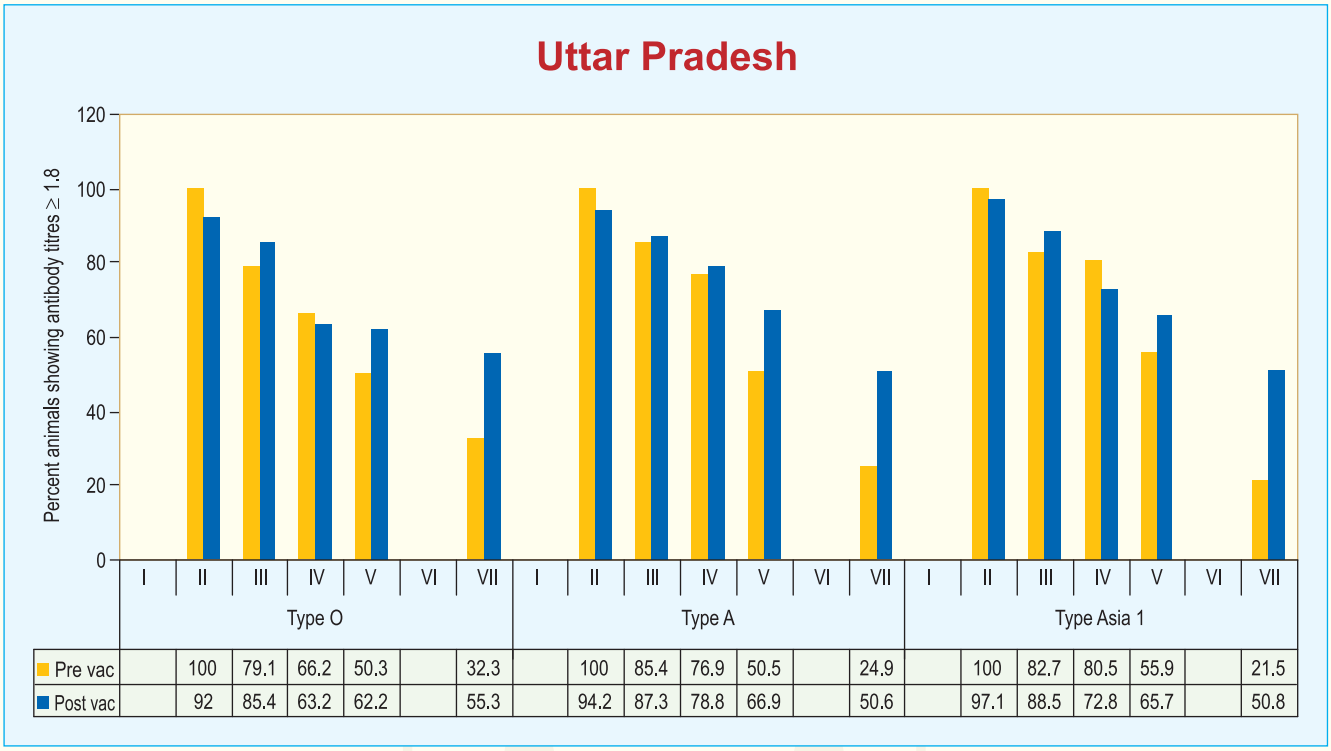
Fig. 21 Seroconversion in Punjab



	Type O							Type A							Type Asia 1						
	I	II	III	IV	V	VI	VII	I	II	III	IV	V	VI	VII	I	II	III	IV	V	VI	VII
Pre vac	28	23	32.7	32.7		18.7	11.7	29	24	33.8	33.8		23.8	11.3	24	18	25	25		21.5	12
Post vac	51	63	74.5	74.5		76.1	70	57	40	60.9	60.9		83.8	77	54	61	65.4	65.4		79.2	75.3

*Serum samples were not available for Phase V

Fig. 22 Seroconversion in Tamil Nadu



*Serum samples were not available for Phase I
Sample testing in progress for Phase VI

Fig. 23 Seroconversion in Uttar Pradesh

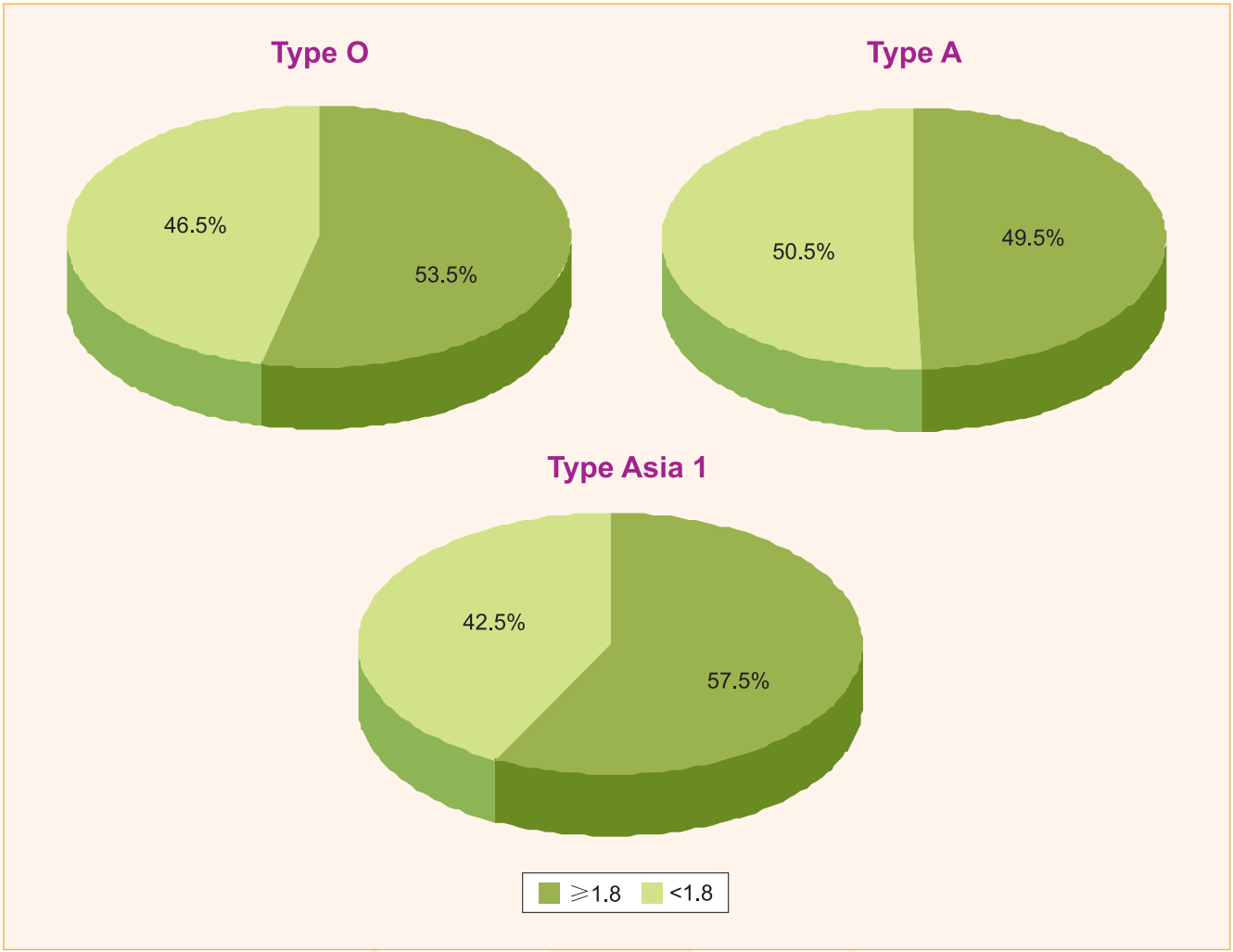
1.12 FMDCP Summary (Phase-wise). Number and percent animals showing antibody titer ≥1.8 log_{10} against FMD virus from Phase I to VIII

1. Phase I

State	Species	Number and % animals showing titres ≥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						
Percent overall sero-conversion	Cattle+Buff	27.3	53.5	35.0	49.5	23.8	57.5

* Kerala Phase I, II & IV data is combined

Per cent Post vaccinal antibody titers against different serotypes in First phase

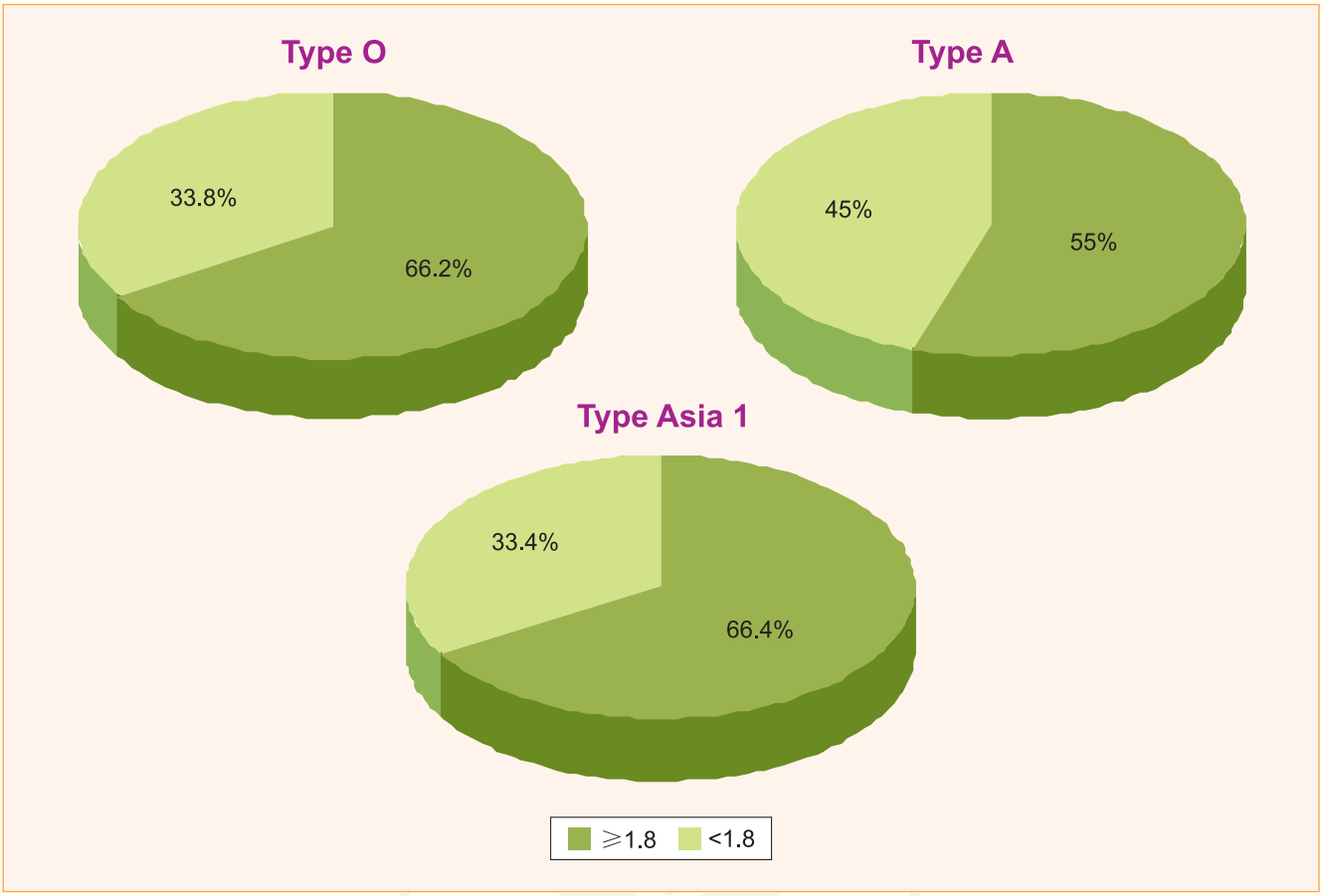


2. Phase II

State	Species	Number and % animals showing titres ≥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)

* Kerala Phase I, II & IV data is combined

Per cent Post vaccinal antibody titers against different serotypes in Second phase

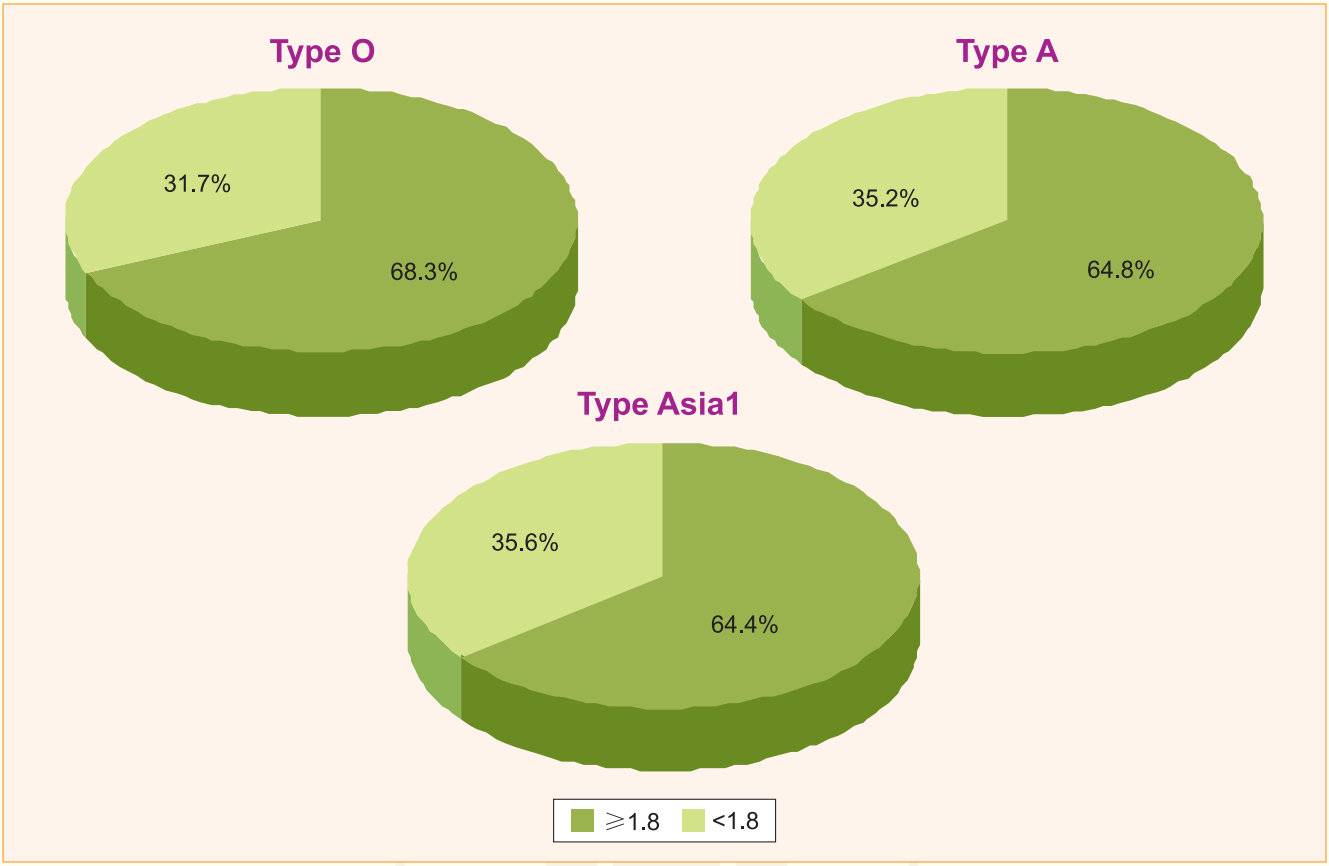


3. Phase III

State	Species	Number and % animals showing titres ≥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	900(79.1)	1347(85.4)	980(85.4)	1377(87.3)	942(82.7)	1398(88.5)
Percent overall sero-conversion	Cattle+Buff	49.3	68.3	49.0	64.8	44.1	64.4

** Tamil Nadu Phase III & IV data is combined

Per cent Post vaccinal antibody titers against different serotypes inThird phase



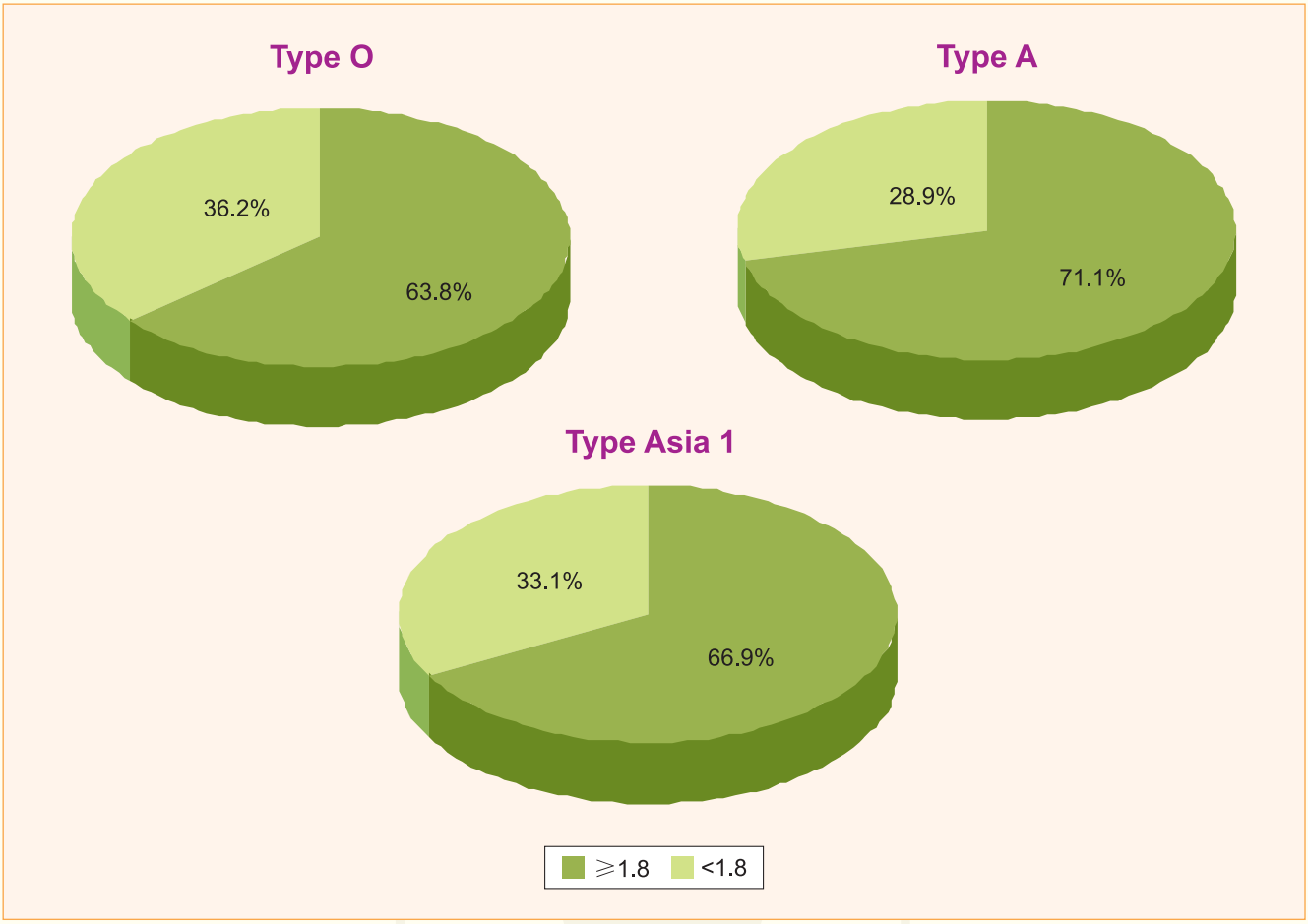
4. Phase IV

State	Species	Number and % animals showing titres ≥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	Serum samples submitted to Bangalore & Guwahati centre for testing						
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	1145(66.2)	959(63.2)	1395(76.9)	1299(78.8)	1451(80.5)	1167(72.8)
Percent overall sero-conversion	Cattle+Buff	42.6	63.8	45.3	71.1	40.1	66.9

* Kerala Phase I, II & IV data is combined

* *Tamil Nadu Phase III & IV data is combined

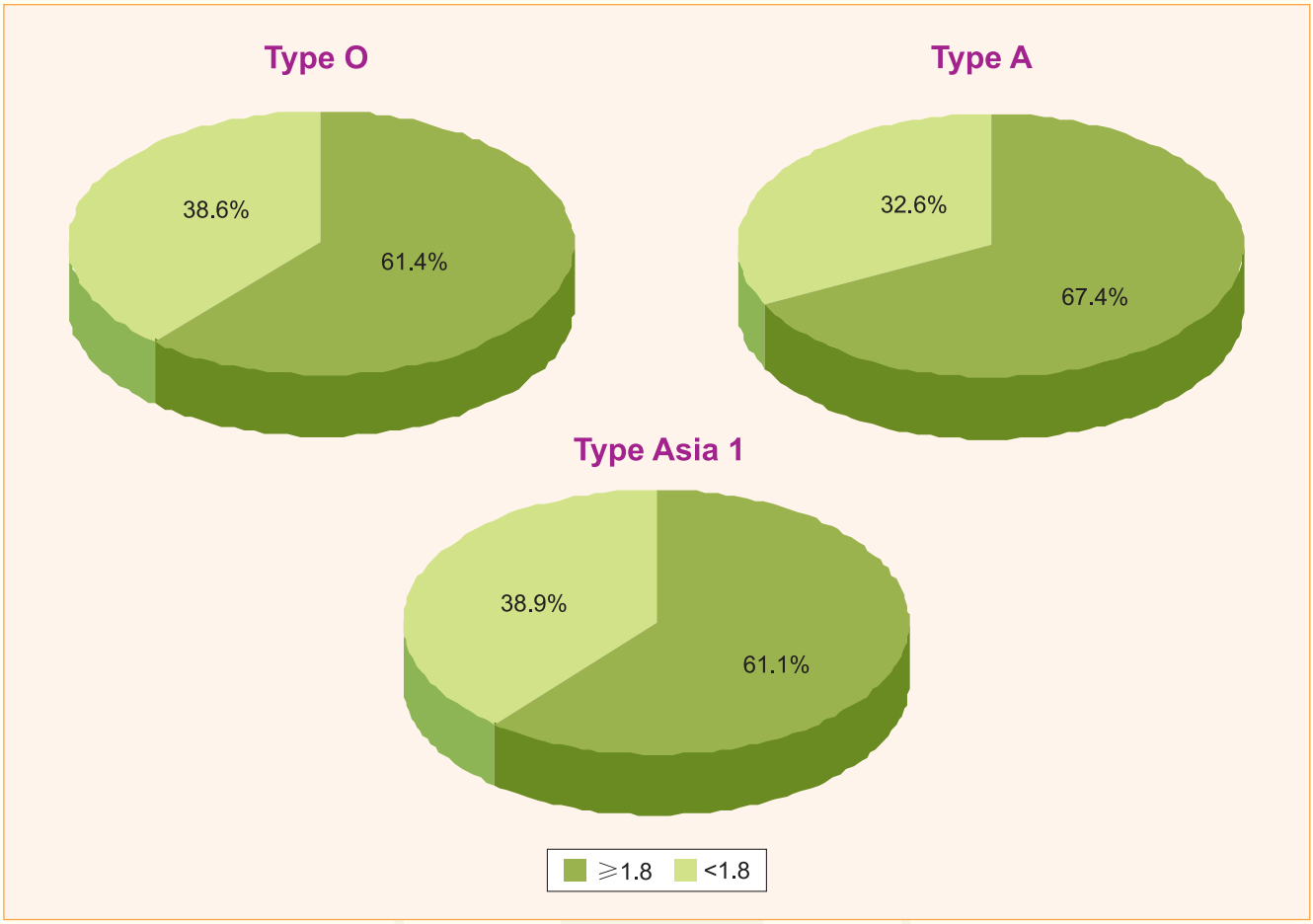
Per cent Post vaccinal antibody titers against different serotypes in Fourth phase



5. Phase V

State	Species	Number and % animals showing titres ≥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	734(50.3)	751(62.2)	810(50.5)	835(66.9)	821(55.9)	816(65.7)
Percent overall sero-conversion	Cattle+Buff	39.6	61.4	47.1	67.4	41.7	61.1

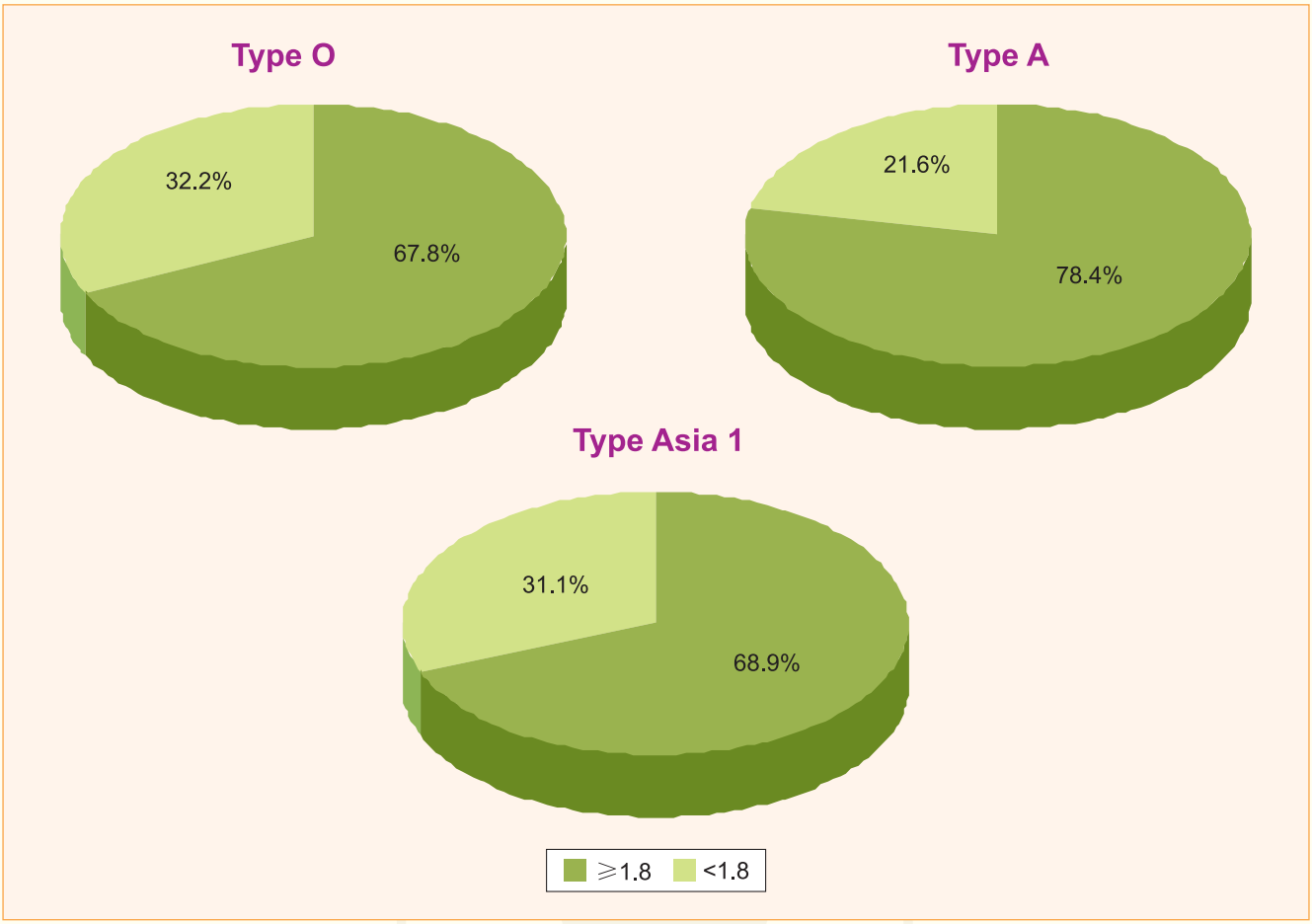
Per cent Post vaccinal antibody titers against different serotypes in Fifth phase



6. Phase VI

State	Species	Number and % animals showing titres ≥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(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	13(18.5)	50(71.4)	14(20.0)	49(70.0)	9(12.8)	53(75.7)
Maharashtra	Cattle+Buff	75 (22.4)	170 (51.5)	238(71.2)	308 (93.3)	134(40.0)	204 (61.8)
Punjab	Cattle+Buff	357(48.5)	526 (78.1)	393(53.4)	519 (77.1)	267(36.3)	517 (76.8)
Tamil Nadu	Cattle+Buff	30(18.7)	99(76.1)	31(23.8)	109(83.8)	28(21.5)	103(79.2)
Uttar Pradesh	Serum testing is in progress						
Percent overall sero-conversion	Cattle+Buff	37.2	67.8	49.5	78.4	39.4	68.9

Per cent Post vaccinal antibody titers against different serotypes in Sixth phase



7. Phase VII

State	Species	Number and % animals showing titres ≥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 testing is in progress						
Andhra Pradesh	Serum testing is in progress						
Delhi	Serum testing is in progress						
Gujarat	Serum testing is in progress						
Haryana	Serum testing is in progress						
Kerala	Serum testing is in progress						
Maharashtra	Serum testing is in progress						
Punjab	Serum testing is in progress						
Tamil Nadu	Cattle+Buff	35(11.7)	210(70.0)	34(11.3)	231(77.0)	36(12.0)	226(75.3)
Uttar Pradesh*	Cattle+Buff	350(32.3)	400(55.3)	270(24.9)	366(50.6)	233(21.5)	367(50.8)

* Sample testing in progress. Results represent partial of total samples tested

8. Phase VIII

State	Species	Number and % animals showing titres ≥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 testing is in progress						
Andhra Pradesh	Serum testing is in progress						
Delhi	Serum testing is in progress						
Gujarat	Serum testing is in progress						
Haryana	Serum testing is in progress						
Kerala	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	Serum testing is in progress						
Tamil Nadu	Serum testing is in progress						
Uttar Pradesh	Serum testing is in progress						

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

Table 24. Percent animals showing post vaccinal antibody titers of ≥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	35.0	49.5	23.8	57.5
II	61.7	66.2	48.3	55.0	61.8	66.6
III	49.3	68.3	49.0	64.8	44.1	64.4
IV	42.6	63.8	45.3	71.1	40.1	66.9
V	39.6	61.4	47.1	67.4	41.7	61.1
VI	37.2	67.8	49.5	78.4	39.4	68.9
Overall seroconversion	43.0	63.5	45.7	64.4	41.8	64.2

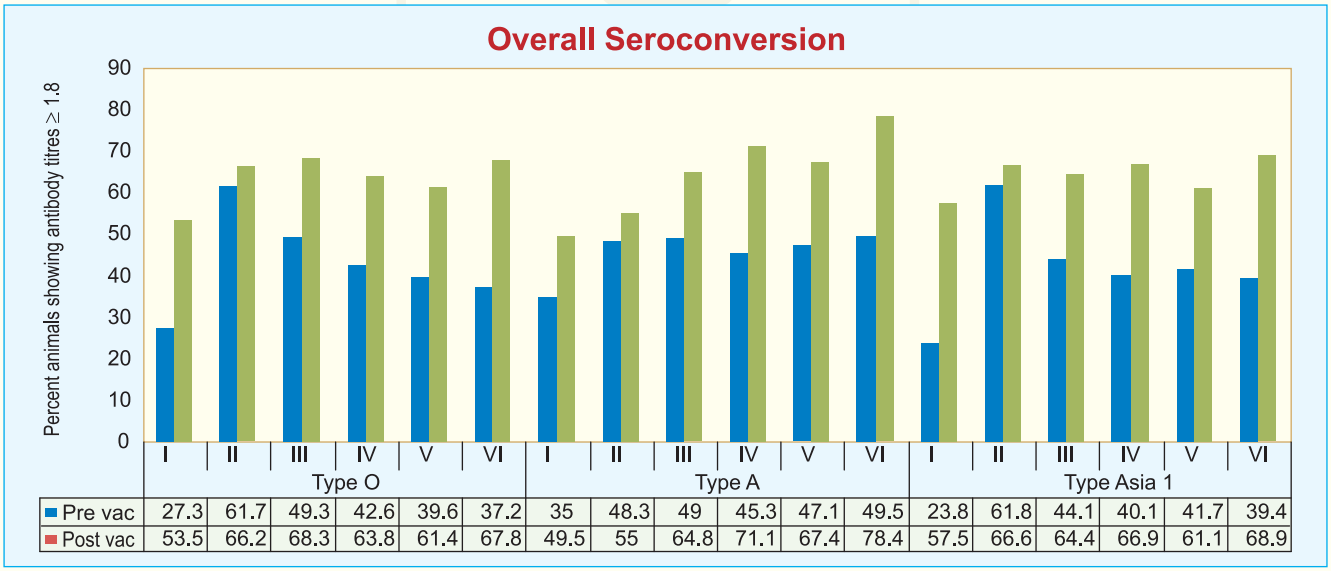


Fig. 24 Overall seroconversion from Phase I to VI

9.2 Sero-Epidemiology

9.2.1 Random serum samples tested at Central FMD laboratory, Mukteswar

During the period under report, a total of 1888 random serum samples were subjected to LPB ELISA for determination of antibody level against serotypes O, A and Asia1 at Central FMD laboratory, Mukteswar. The results of LPB ELISA are shown in Fig 25.

Table 25. Number of animals showing titres ≥1.8 $\log_{10}$ against FMD virus Type O

State	No. of random samples	Number of animals showing titres ≥1.8 $\log_{10}$ against type O				
		< 1.5	1.5-1.79	1.8-2.09	2.1-2.4	>2.4
Himachal Pradesh	25	0	0	0	4	21
Jammu & Kashmir	160	73	10	7	20	50
Karnataka	275	18	36	16	52	153
Kerala	3	0	0	1	0	2
Orissa	854	473	111	45	71	154
Punjab	23	0	0	4	1	18
Rajasthan	417	175	93	63	62	24
Tamil Nadu	32	20	9	1	1	1
Tripura	99	23	16	9	20	31
% Seroconversion		41.4	14.5	7.7	12.3	24.0

Table 26. Number of animals showing titres ≥1.8 $\log_{10}$ against FMD virus Type A

State	No. of random samples	Number of animals showing titres ≥1.8 $\log_{10}$ against type A				
		< 1.5	1.5-1.79	1.8-2.09	2.1-2.4	>2.4
Himachal Pradesh	25	0	0	1	4	20
Jammu & Kashmir	160	80	25	11	18	26
Karnataka	275	37	27	20	41	150
Kerala	3	0	0	0	0	3
Orissa	854	484	130	56	90	94
Punjab	23	0	0	0	4	19
Rajasthan	417	192	90	68	51	16
Tamil Nadu	32	11	8	7	3	3
Tripura	99	29	14	7	25	24
% Seroconversion		44.1	15.6	9.0	12.5	18.8

Table 27. Number of animals showing titres ≥1.8 $\log_{10}$ against FMD virus Type Asia1

State	No. of random samples	Number of animals showing titres ≥1.8 $\log_{10}$ against type Asia1				
		< 1.5	1.5-1.79	1.8-2.09	2.1-2.4	>2.4
Himachal Pradesh	25	0	0	4	4	17
Jammu & Kashmir	160	117	9	6	6	22
Karnataka	275	74	70	52	53	26
Kerala	3	0	0	0	0	3
Orissa	854	579	98	48	65	64
Punjab	23	0	2	3	5	13
Rajasthan	417	226	85	52	25	24
Tamil Nadu	32	19	8	3	1	1
Tripura	99	23	24	9	18	25
% Seroconversion		55.0	15.7	9.5	9.5	10.3

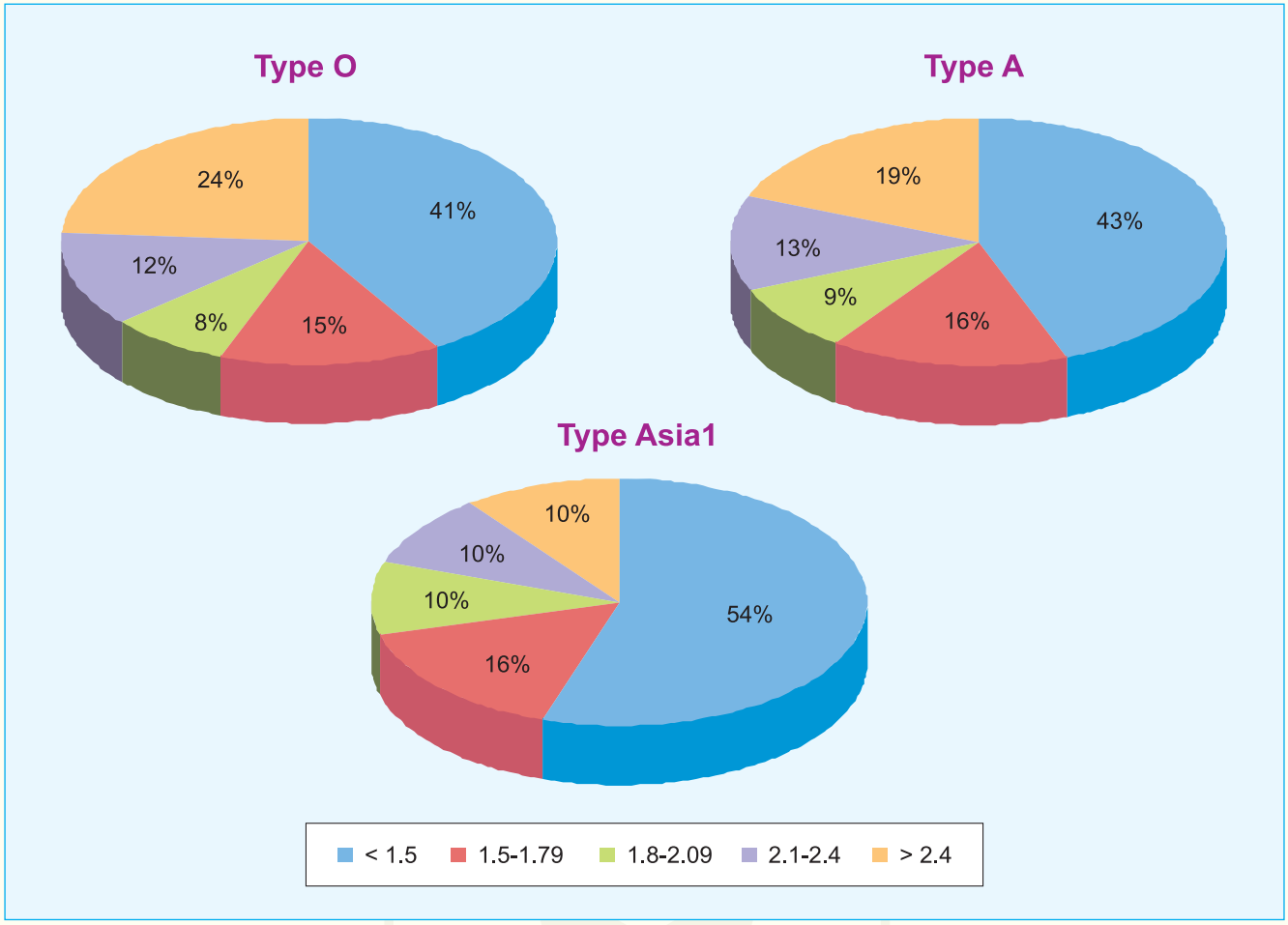


Fig. 25 Serum antibody titre against different serotypes serum samples collected at random

9.2.2 Random serum samples tested at Regional Centers and Network Units

Along with the Central Laboratory, Mukteswar the testing of random serum samples was carried out by selected Regional Centers and Network Units. The results obtained are given in Table 28. Although a small number of serum samples were tested from Himachal Pradesh, the protective antibody present in the population can very well corrected with no incidence of FMD in the state. Inspite of vaccination under FMDCP, a low level of protective antibody was observed in Andaman and Nicobar Islands.

Table 28. Random serum samples showing protective antibodies against different serotypes

State	No. of random samples	Number and % of animals showing titres ≥1.8 $\log_{10}$		
		Type O	Type A	Type Asia1
Andaman & Nicobar	657	127(19.3)	138(21)	69(10.5)
Haryana	2634	1482(56.2)	1739(66.0)	1518(57.6)
Himachal Pradesh	25	25(100)	25(100)	25(100)
Madhya Pradesh	1418	533(37.6)	587(41.4)	380(26.8)
Uttar Pradesh	164	64(39.0)	53(32.3)	61(37.2)

10.0 International Collaboration

10.1 OIE/FAO Global FMD vaccine matching exercise

The Project Directorate on FMD, a regional reference laboratory within the OIE/FAO Network of FMD Reference Laboratories, has participated in 2008 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 Laboratories 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 harmonisation of such approaches.

Objectives

1. To gather, generate, analyse 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 harmonise approaches to the characterisation 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 intergrated 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.

Materials

A historical Eurasian FMD virus Serotype A Iraq 24/64 was chosen to be used as the vaccine strain. Five cattle have been vaccinated with this virus in Onderstepoort to generate the BVS, and five FMD Serotype A field isolates have been selected in WRL-FMD, Pirbright to be matched against the A Iraq 24/64 vaccine for this study.

On mutual agreement, we have received 15ml of each of five BVS (heat inactivated), 8 ml of BEI inactivated vaccine virus A Iraq 24/64 and 5 ml of each of 5 coded BEI inactivated FMD serotype A field isolates.

Methodology

1. To find out capture and detector antibodies for use in LPB-ELISA, the closest virus IND 17/77 was tested in two-dimensional-micro-neutralization test using BVS against Iraq 24/64.
2. Serial dilution of rabbit and guinea-pig anti-146S serum against IND 17/77 as capture and detector antibodies to be used was determined by checker board titration.
3. Optimized dilution of coating (1:5000) and tracing (1:4000) serum was used to determine the one way antigenic relationship of type A field isolates with Iraq 24/64. Antigens of field isolates were also titrated by making dilutions ranging from 1:2 to 1:14. Antigens were used at dilutions (A Iraq 24/64 and A62 at 1:5; A66 at 1:4; A61 at 1:3 and A63 and A65 at 1:2) as determined by checker board titration. Indian vaccine strains of type A were also included in this study.
4. The dilution of anti-guinea-pig conjugate (1:2000) was also determined by checker board titration.
5. LPB-ELISA was carried out using serial two fold dilutions of bovine vaccinate serum (BVS 06104, BVS 0610, BVS 0637, BVS 0645 and BVS 0679) ranging from 1:16 to 1:1024.
6. Titer of serum samples was calculated graphically as the reciprocal of the serum dilution giving 50% Optical Density (OD) as compared to the antigen controls. Or, in other words, reciprocal of the serum dilution which inhibits 50% of the guinea pig serum binding to the homologous virus.
7. The one way antigenic relationship (r value) was calculated as follows.

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

The r value was calculated as an average of two different tests conducted independently on separate days to understand intra-laboratory variation between tests using paired students *t*-test.

Antigenic analysis of the field isolates in relation to the vaccine strain(s) is significant for testing the appropriateness of the existing vaccine strain as well as for selection of new vaccine strain(s), if required. The ratio between the antibody titers against the heterologous virus *i.e.* the field isolates and the homologous virus *i.e.* the reference/vaccine virus was used to calculate the one-way antigenic relationship (r-value) between them.

Interpretation of r1 value in LPB ELISA

r1 = 0.4-1.0. Suggests that there is a close relationship between field isolate and vaccine strain. A potent vaccine containing the vaccine strain is likely to confer protection.

r1 = 0.2-0.39, Suggests that the field isolate is antigenically related to the vaccine strain. The vaccine strain might be suitable for use if no closer match can be found provided that a potent vaccine is used and animals are preferably immunized more than once.

r1 = <0.2. Suggests that the field isolate is so different from the vaccine strain that the vaccine is unlikely to protect.

None of the field isolates reacted with BVS 0610 collected from cattle vaccinated with Iraq 24/64 in Onderstepoort, indicating poor antibody response. The result is also similar for the BVS 0679 where a low level (r<0.30) of reactivity was observed with all the field isolates.

Table 29. Determination of day-to-day variation of one-way antigenic relationship value of serotype A FMD virus field isolates in relation to Iraq 24/64 using bovine vaccinates serum

		BVS06104		
	DAY 1	DAY 2	AVERAGE	p-value
IND 17/77	2.56	2.56	2.56	0.13
IND 40/00	1.9	1.72	1.81	
IRAQ 24/64	2.6	2.6	2.6	
A61	1.98	1.8	1.89	
A62	2.01	2	2.005	
A63	1.56	1.5	1.53	
A65	2.44	2.46	2.45	
A66	2.16	2.24	2.2	
		BVS0610		
	DAY 1	DAY 2	AVERAGE	p-value
IND 17/77	<1.5	<1.5	<1.5	-
IND 40/00	<1.5	<1.5	<1.5	
IRAQ 24/64	<1.5	<1.5	<1.5	
A61	<1.5	<1.5	<1.5	
A62	<1.5	<1.5	<1.5	
A63	<1.5	<1.5	<1.5	
A65	<1.5	<1.5	<1.5	
A66	<1.5	<1.5	<1.5	
		BVS0637		
	DAY 1	DAY 2	AVERAGE	p-value
IND 17/77	2.36	2.35	2.355	0.05
IND 40/00	2.04	1.76	1.9	
IRAQ 24/64	2.64	2.64	2.64	
A61	2	1.86	1.93	
A62	2.1	1.98	2.04	
A63	<1.5	<1.5	1.5	
A65	2.4	2.48	2.44	
A66	2.1	1.98	2.04	
		BVS0645		
	DAY 1	DAY 2	AVERAGE	p-value
IND 17/77	2.46	2.56	2.51	0.04
IND 40/00	2.16	1.9	2.03	
IRAQ 24/64	2.87	2.8	2.835	
A61	1.96	1.88	1.92	
A62	2.18	2.08	2.13	
A63	1.7	1.5	1.6	
A65	2.62	2.5	2.56	
A66	2.12	2.18	2.15	

	BVS0679			
	DAY 1	DAY 2	AVERAGE	p-value
IND 17/77	2.62	2.7	2.66	0.25
IND 40/00	2.15	2.2	2.175	
IRAQ 24/64	3.12	3.12	3.12	
A61	2.16	2.17	2.165	
A62	2.1	1.95	2.025	
A63	1.74	1.72	1.73	
A65	2.56	2.6	2.58	
A66	2.31	2.1	2.205	

Table 30. One-way antigenic relationship (average of two different tests) of serotype A FMD virus field isolates in relation to Iraq 24/64 using bovine vaccinates serum

	BVS06104	BVS0610	BVS0637	BVS0645	BVS0679
	r1	r1	r1	r1	r1
IND 17/77	0.91	0	0.52	0.47	0.35
IND 40/00	0.16	0	0.18	0.15	0.11
IRAQ 24/64	1.00	0	1.00	1.00	1.00
A61	0.19	0	0.19	0.12	0.11
A62	0.25	0	0.25	0.19	0.08
A63	0.09	0	0.07	0.06	0.04
A65	0.71	0	0.63	0.52	0.29
A66	0.40	0	0.25	0.20	0.12

Field isolates A61, A62 and A63 behaved poorly with all the serum and in no case an r value of >0.40 was observed. These isolates are antigenically divergent from Iraq 24/64. The virus A66 was also found to be antigenically divergent from Iraq 24/64, except with serum BVS 06104 where a close antigenic relationship (r>0.40) was observed. With all the four BVS (BVS0610, BVS0637, BVS0679, BVS0645), the virus A65 was found to be antigenically similar except a low level of relationship with serum BVS 0679 (r1=0.29).

To conclude, A65 virus is antigenically related to A Iraq 24/64, A66 is intermediate and the viruses A61, A62 and A63 are antigenically divergent from A Iraq 24/64. As IND 40/00 is antigenically divergent from A Iraq 24/64, the currently circulating genotype VII type A viruses in India may not be covered by Iraq 24/64. This is also reflected by a close antigenic relationship of IND 17/77 with Iraq 24/64 and IND 17/77 has poor antigenic coverage to genotype VII viruses circulating in the country.

10.2 Collaboration with USDA-ARS (Under GFRA)

Collaboration on "Antigenic and genetic characterization of Foot and Mouth Disease viruses in India: Application to effective molecular vaccines" has been initiated. IVRI, Bangalore campus also participates in this programme. The overall objective of this project is to identify molecular determinants responsible for antigenic variation of Indian FMDV field strains. Objectives include:

1. Apply bioinformatics tools using currently

available serological and genetic data to identify molecular determinants responsible for antigenic variation.

2. Utilize the hAd5-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.
3. Test in pilot proof of concept experiment the immunological response of animals vaccinated with the novel hAd5-FMD vaccines compared to the current vaccine.
4. Test candidate Ad5 vaccine efficacy in buffalo.
5. Determine duration of immunity of Ad5 vaccine candidates and study FMDV persistence in vaccinated animals.
6. Determine effect of multiple Ad5 immunizations on FMD vaccine response.
7. Characterize immune response of cattle immunized with Ad5 and/or current vaccine.

The outcome of the study is expected to be a thermostable viral vectored molecular vaccine with longer duration of immunity.

11.0 Reports, Recommendations and Publications

11.1 Institute Management Committee

The Institute management Committee comprises the following officials:

1.	Rule 66 a(1) Director of the Institute	Dr. B. Pattnaik	Chairman
2.	Rule No. 66 a(2) A representative of State Govt. in which the Instt. Is located, nominated by President, ICAR	Director of Animal Husbandry & Veterinary Services Govt. of Uttrarakhand, Dehradun	Member
3.	Rule No. 66 a(3) A representative of any other State Govt. concerned with the Research in the Institute nominated by President, ICAR	Director of Animal Husbandry & Veterinary Services Govt. of Uttar Pradesh, Lucknow	Member
4.	Rule No. 66 a (4) A representative of Agril. University having jurisdiction over the area nominated by President, ICAR	Dean, College of Veterinary Science G.B. Pant University of Agril. & Tech. Pantnagar, Uttarakhand	Member
6.	Rule No. 66 a(6)4 Scientists of Council's Institutes to be nominated by the Director General	Dr. R.K.Singh, Head, Division of Virology IVRI, Mukteswar Dr. Satish Kumar Principal Scientist, IVRI, Izatnagar Dr. A. Sanyal, Sr. Scientist PD on FMD Dr. D. Hemadri, Sr. Scientist PD on ADMAS	Member Member Member Member Member
7.	Rule No. 66 a (7) A representative from the Council nominated by the Director General	Dr. Lal Krishna Asstt. Director General (AH), ICAR Krishi Bhavan, New Delhi	Member
8.	Rule 66 a(8) The financial adviser of the council or department of Agril. Research and Education or the Accounts Officer of the same or another Institute, nominated by the President.	Finance & Accounts Officer, IVRI Izatnagar	Member

9.	Rule 66(a)5 Non official members nominated by President ICAR	Ms Chanda Nimbkar, Director Animal Husbandry Division Nimbkar Agricultural Research Institute Phaltan, MH Mr Suryakant Dhasmana President, NCP, Dehradun, UK	Member Member
10.	Rule No. 66 a(9) Asstt. Admn. Officer P.D. on FMD	Shri D N Joshi, Asstt. Admn. Officer, P.D. on FMD, IVRI Campus Mukteswar	Member Secretary

5th meeting of Institute Management Committee held at Mukteswar

6th meeting of Institute Management Committee held at New Delhi

The 5th Meeting of the IMC was held on 10.06.2008 at Mukteswar and the 6th Institute Management Committee meeting was held on 06-02-2009 at ICAR Committee room, Krishi Bhawan, New Delhi. The Project Director presented scientific achievements and targets of the Directorate before the members. He informed that EFC for 11th Plan has already been sanctioned and apprised the honourable members that suitable land has already been acquired for establishment of International FMD Center with containment laboratory and experimental animal facility. The ADG (AH) informed the house that this center will function as FAO Reference center for FMD in the region. The IMC has reviewed the different activities, programmes, manpower requirement and expenditure etc. The IMC made recommendations on various agenda as discussed.

11.2 Annual Scientists Meeting

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. There were special invitees and also representatives of all the four FMD vaccine industry of the country.

Dr. B. C. Bist, Director, Animal Husbandry, HP gave the inaugural address and highlighted that concerted efforts and vaccination has resulted in no FMD outbreak in the state since 2007-08. Dr. Lal Krishna, ADG(AH), ICAR, the Guest of Honour 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

19th Annual Scientists Meet of the PD FMD held at Shimla

diagnosis. He 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 further stressed that as the incidence has come down to near zero in the Northern states of HP, Punjab, Haryana and Delhi, creation and maintenance of disease free zone (DFZ) can be thought of to boost export. He informed the house that ICAR is going to host the 3rd Annual meeting of the "OIE/FAO Global Network of FMD Reference Laboratories" during November 2009 in Delhi, and scientists of the regional centers/ network units must participate in this Global Meet and present the scenario of FMD in their state. He further said that there is immediate necessity of developing a DIVA Kit for use in the country.

The first session was chaired by Dr. Lal Krishna, ADG (AH), ICAR, and co-chaired by Dr. R. Venkataramanan, Joint Director, IVRI, Bangalore.

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, 73.4, 80.4 and 75.8 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. 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. Having this assay system in place well in time will be of immense help as the FMD Control Programme expands gradually and the incidence of the disease drops across the country. This DIVA kit that contains freeze dried 3AB3 antigen and

all other components is being distributed throughout the country (both public and private sector) to detect FMD negative/ positive bovine and estimate prevalence of FMD in the country at the district level. 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, whereas Dr. J. K. Mohapatra, Scientist, elaborated the DIVA test kit developed and its evaluation and application.

Summary of the progress made during 2007-08

- A total of 1211 outbreaks were recorded/ reported as against 1467 and 2962 outbreaks during the years 2006-07 and 2005-06, respectively. There is visible positive effect of regular vaccination under FMD Control Programme and other programmes of the country.
- Maximum numbers of outbreaks were reported in Kerala (237), Tamil Nadu (219), West Bengal (178), Bihar (138) followed by Karnataka (113). The four states of South India contributed to 50% of the FMD outbreaks; almost same as that of 2006-07.
- There was reduction in the number of outbreaks in Eastern region. But Northern, Western, Central and North Eastern regions of the country experienced more number of outbreaks compared to last year (2006-07).
- 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 outbreak was reported in Manipur and Himachal Pradesh, whereas Punjab and Haryana recorded five and two sporadic cases, respectively.
- A total of 2258 clinical samples were collected from 1211 FMD outbreaks through the network of laboratory and subjected to virus typing. Virus could be identified in 1269 samples viz. type O 1042, type A 136, type Asia1 91 and the remaining samples were negative in ELISA. Using sandwich ELISA, 842 (69.5%) FMD outbreaks could be diagnosed at center/unit level.
- Multiplex PCR (mPCR) was applied on ELISA negative samples and by which another 71 outbreaks could be diagnosed. Overall 66.4% of FMD outbreaks referred could be diagnosed using both the tests at the Central laboratory.
- There was no incidence of type C FMD virus during this year also. Type O dominated the outbreaks scenario followed by types A and Asia 1. The activity of Type O virus was diagnosed in all the states except Haryana where only type A was diagnosed. In all the geographical regions, other than Central India, serotype O was most prevalent.
- In contrast to the previous year, Northern region had outbreaks due to serotypes A 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, but the incidence of type Asia 1 decreased compared to the previous year.
- In Eastern region, increase in incidence of type O and decrease in incidence of Asia1

was noticed. Type O activity was considerably increased in North Eastern region compared to last year.

- In the Central region, though there was absence of serotype A during previous year, this year circulation of type A increased and even dominated marginally over serotype O.
- All the three serotypes were involved in FMD outbreaks of West Bengal, Maharashtra, Kerala, Tamil Nadu, Madhya Pradesh, Uttar Pradesh and Assam,
- Only serotypes O and A were responsible for FMD outbreaks in Bihar, Karnataka, Orissa and Tripura.
- In Andhra Pradesh, Gujarat and Arunachal Pradesh, there was prevalence of only O and Asia-1 serotypes.
- Outbreaks in Rajasthan, Punjab, Jammu & Kashmir, Meghalaya, Mizoram and Nagaland were exclusively due to serotype O FMD virus.
- Though majority of the outbreaks involved cattle, disease was also reported in buffaloes, pigs, goat and sheep. Outbreaks in wild animals were reported from four states. Outbreak in Mithun was recorded in Arunachal Pradesh, Kerala and Karnataka. Wild animals like Black buck, Wild Boar, Mithun and Nilghai were affected in Thrissur and Thiruvananthapuram zoo and these animals developed typical FMD clinical signs. Outbreak was reported/suspected in wild Indian gaur and wild elephant calf in Tamil Nadu.
- Though there is seasonal variation in occurrence of FMD in different parts of the country during the years 2007 and 2008, FMD outbreaks occurred round the year also during 2008 with maximum incidence in the month of March.
- Molecular epidemiological analysis of type O isolates based on 1D sequence analysis resembled that of the previous year. During this year also, PanAsia II strains continued to dominate followed by its parent lineage PanAsia I. Besides, the "Ind 2001 strain" was also detected in a few outbreaks in the state of UP.
- In serotype A, within the currently circulating genotype VII, a divergent and unique sub-lineage emerged in late part of 2002, which showed an amino acid deletion at 59th position of VP3 (VIIb-VP3^{59} deletion group) and dominated the field outbreak scenario in 2002-03. In 2007-08, there is once again an upsurge in incidence of outbreaks due to this sub lineage.
- In case of serotype Asia1, Lineage CI continued to dominate also during 2007-08. This lineage was in circulation during 1998-2000, and has reappeared since 2005. During 2001-2004, lineage CII was in circulation.
- Majority of field isolates of serotypes O and Asia 1 demonstrated close antigenic relationship with respective vaccine strains indicating good antigenic coverage by them.
- In case of serotype A, some isolates of VP3 deletion group displayed closer antigenic relatedness with 17/82 (old vaccine strain), whereas a few others displayed more relatedness with IND 40/00 (new vaccine strain). No antigenic superiority of IND 40/00 above 17/82 was observed in relation to this deletion mutant sub-lineage, as similar number of isolates revealed r-value of more than 0.4 with both these vaccine strains. Sudden upsurge of this type A deletion group is being monitored.
- 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. This DIVA kit that contains freeze dried 3AB3 antigen and all other components is being distributed throughout the country (both public and private sector) to detect FMD negative/ positive bovine and estimate prevalence of FMD in the country at the district level.

- 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, 73.4, 80.4 and 75.8 percent of animals vaccinated/ tested were having protective antibody level (log_{10} 1.8 and above) against serotypes O, A and Asia-1, respectively. This result is quite encouraging.
- The National Repository of FMDV was upgraded during this year also and now it comprises a total of 1402 (893-O, 261-Asia 1, 233-A, 15-C) well characterized field isolates.

Subsequent technical sessions on 19th and 20th were also chaired by ADG (AH), and Project Director, ADMAS co chaired. The scientists of the regional centers and network units presented the report for the year. The report of the Bangalore Regional Center and Jammu Network Unit has to be resubmitted after suggested inclusions. As the FMDCP result in Uttar Pradesh appeared complicated, it was decided that Dr. Sharad Yadav, I/C, Mathura regional center will submit a report for inclusion in the recommendations. It was also 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. In Arunachal Pradesh, there was no outbreak during 2005-07, but during 2007-08 eighteen outbreaks were recorded in cattle and mithun. The scientist in charge, Itanagar network unit and Scientist in charge, Guwahati Regional Center were advised to submit a detailed report on the cause of this. Good work done on quantitative epidemiology by the Manipur, Nagaland and Orissa network units was appreciated. The situation of FMD in Kerala in spite of vaccination was viewed very seriously and it was observed that this matter required to be taken up by DAHD&F.

The plenary session, on 20th, was chaired by ADG (AH) and co-chaired by Director, AH, Himachal Pradesh 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.

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 programme 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]

11.3 Research Advisory Committee

The first RAC meeting of the PD on FMD was held on 09.06.2008 at IVRI campus, Mukteswar under the chairmanship of Prof. M. P. Yadav, Vice-chancellor, S.V.B.P. University of Agricultural and Technology, Meerut. The members who attended the meeting include Dr. Lal Krishna, ADG (A.H), ICAR; Dr. B. Pattnaik, Project Director, PD on FMD, Mukteswar; Dr. P.K.Dutta, Director of Research (Vety), AAU, Guwahati; Dr. V.A. Srinivasan, Research Director, IIL, Hyderabad; Dr. Chanda Nimbkar, Director, Nimbkar.Agri.Res.Inst, Phaltan, MH; . Dr. V.D.P. Rao and Dr. D.V. Rai as special invitee and scientists of the IVRI Campus, Mukteswar and Central FMD Laboratory of PDFMD. The Project Director welcomed the chairman and members of RAC including special invitees to the first RAC meeting and requested the chairman to conduct the proceedings. The chairman in his opening remarks complimented the scientists for the good work done and explained the importance of FMD in adversely affecting the Dairy and meat Industry, Livestock health and productivity.

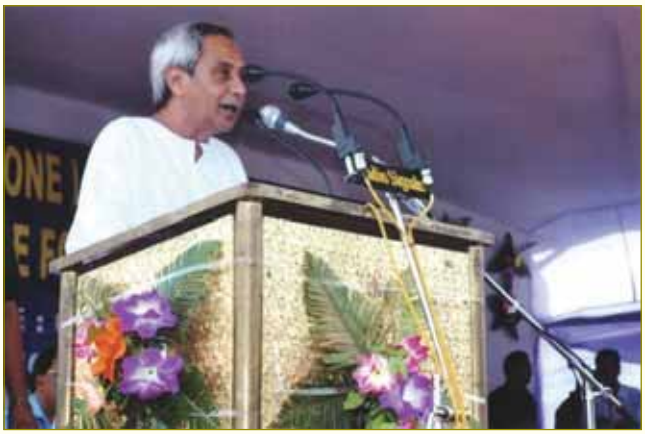
1st RAC meeting of PD on FMD held at Mukteswar

Dr. A. Sanyal, Member Secretary and Scientist-in-charge, Central FMD Laboratory presented the research programmes and achievements of the Project Directorate including sero-monitoring under FMD-CP during the last five years, development of diagnostic tests for rapid and precise diagnosis of FMD, and identification of candidate vaccine strains for FMD virus types O, A and Asia1. The recommendations made by the QRT (1998-2003) and already approved by the Governing body of ICAR were also discussed at length with regard to their implementation and it was observed that most of the recommendations are being implemented. Dr. Lal Krishna, ADG, Animal Health informed the committee about different factors/attributes discussed at the government level at length before finalizing the FMD control program being undertaken in 54 districts in 7 states of the country. Honourable members of RAC offered a number of suggestions and recommendations for improving the research programmes of the Institute. The RAC members expressed their happiness and satisfaction on the progress of the work. Dr. B. Pattnaik, Director proposed vote of thanks.

11.4 Foundation Stone Ceremony of International Center for FMD

Foot and mouth disease (FMD) is the major disease constraint to international trade in livestock and animal products. THE Project Directorate on Foot and Mouth Disease (FMD), which is the premier Institute for FMD in the country, was established as an All India Coordinated Research Project (AICRP) for FMD in 1968 at IVRI Campus, Mukteswar. During the last four decades of its existence the scope of the project was expanded considerably and

Foundation Stone Ceremony of International Center for FMD at Aragul (Bhubaneswar, Orissa)



several milestones were achieved to reach the current status of a Project Directorate 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, second to none in the world and the leader in South Asia, in the field of FMD diagnosis, epidemiology and research.

During the 11th Five-Year Plan period (2007-12) it is envisaged to establish a National FMD Laboratory with BSL-3+ facility in the name of National Center for FMD. FMD is a OIE list A disease (risk group 4), and with the expansion of FMD control programme in 11th Five-Year Plan period the number of disease outbreaks is bound to come down. In such a scenario it is essentially required to handle field strains of FMD virus in a laboratory with containment (BSL-3+) facility. In addition, six regional containment laboratories are required to be built to handle the virus.

International Center for FMD for SAARC/ South Asia (South Asia Regional Reference Laboratory for FMD) to cater to the demand of the South Asian (SAARC) countries is also to be established during this period (2007-2012) which will help in generating unified epidemiological data and formulation of Regional FMD Control Programme with Indian experience. There has been constant demand from the international agencies (FAO, OIE) to ICAR for establishment of an International Center for FMD for South Asia in India. Creation of this international laboratory with state-of-the-art features of bio-safety and bio-containment (BSL 3+) will facilitate participation in GFTAD (Global Frame work on Transboundary Animal Diseases initiated by FAO. This state-of-the-art bio-containment laboratory facility will be first of its kind in Orissa in any branch of life science. Further, this international FMD center will be the 11th in the world.

INDIAN COUNCIL OF AGRICULTURAL RESEARCH
Department of Agricultural Research & Education

FOUNDATION STONE

for the

International Center for Foot and Mouth Disease

laid by

Shri Sharad Pawar Ji
Hon'ble Union Minister of Agriculture, consumer Affairs, Food and Public Distribution, Govt. of India

Shri Naveen Patnaik Ji
Hon'ble Chief Minister, Orissa as Chief Guest

in the auspicious presence of

Prof. K.V. Thomas Ji
Minister of State for Agriculture, Consumer Affairs, Food and Public Distribution, Govt. of India

Dr. Prasanna Kumar Patasani
Hon'ble Member, Lok Sabha

Dr. Damodar Rout
Minister of Agriculture, Cooperation and Fisheries & Animal Resources Development, Orissa

Dr. Mangala Rai
Secretary, DARE and Director General, ICAR

Dr. K.M. Bujarbaruah
Dy. Director General (AS), ICAR

Dr. Lal Krishna
Asst. Director General (AH), ICAR

Dr. B. Pattnaik
Project Director, PD-FMD, ICAR

at Aragul, Dist. Khurda, Orissa on 5th July 2009.

ADDRESS OF

SHRI SHARAD PAWAR

UNION MINISTER OF AGRICULTURE &
CONSUMER AFFAIRS, FOOD & PUBLIC DISTRIBUTION

ON THE OCCASION OF

FOUNDATION LAYING CEREMONY OF
INTERNATIONAL LABORATORY
ON FOOT AND MOUTH DISEASE

It is a matter of pride that the Indian economy is growing between 6 and 8 per cent due to the contribution from industry, service, agriculture and other sectors. However, the sustainability of agricultural growth is a matter of concern in the face of challenges facing the sector today. Within agriculture , the livestock sub sector fortunately has positioned itself very well as far as its production is concerned. We are first in cattle, buffalo and goat numbers in the world housing 16.10, 56.50 and 16.50 per cent respectively of the world's population. In sheep and poultry, we are 5th in the world while the position with regard to pig is 17th. On the production front, our milk production has been growing at the rate of 3.3 million tonnes per annum since 2000-2001 with current production of 101 million tones, maintaining thereby a steady growth of around 3.72 per cent. With 7.94% growth in poultry meat and 2.22% in non-poultry sector, an overall growth rate of 5.08 per cent has been recorded in meat sub-sector besides a phenomenal 27 times increase in egg production. Since 1950-51, when we produced only 1.8 billion eggs. Per capita availability of milk and egg has also increased from 220 g and 36 in 2000-2001 to 245 g and 46 in 2006-2007. The share of export of livestock products has also increased from 3.3% to 6.9% in the period between 1981-85 and 1999-2004 besides the increase in the contribution of livestock sector to agriculture from 19.8% to 25% in the same period. This is also the sector whose strength has been amply recognized today in so far as providing economic and social security to 52 per cent of India's population engaged in the farm sector is concerned. All these developments have been possible due to timely generation and provisioning of needed technologies-be it in health or production aspects, ranging from the development of advanced disease diagnostics / vaccine production capabilities to improved strains and feeding packages. Standardization of cloning technology (buffalo cloning), completion of buffalo genome project and application of marker assisted selection both for production and disease resistance are the future technological options to sustain growth in the livestock sub-sector. Mass multiplication of quality animal germplasm produced in research institutions shall facilitate production enhancement with limited number of livestock. I am happy to note that our scientists have already taken initiatives on these lines, including the designing suitable animal shelters for adaptation to impending climate change.

2. We are aware that rapid urbanization, higher per capita income (Rs. 9,450 in 1983 to Rs. 23,313 in 2004-2005), increase in human population (2% p.a.) has fuelled the demand for livestock and poultry products which is indicated by the increase in household food expenditure from 21% to 25% in urban and 16.1% to 21% in rural households, within the period from 1983 to 2004-05. On the employment front, this sector employs 11.4 million people in primary status and

another 11 million in secondary sector related to it and most of this population belongs to small, marginal and landless category who holds around 71% of our cattle, 63% of its buffaloes, 72% of small ruminants and 74% of poultry. The contribution of this sector to agricultural economy has also been impressive even during the Green Revolution period when major focus was on crop sub-Sector. The dairy sector has remained the maximum contributor with 62-68% of total livestock economy. The contribution could have been much more if we could have effectively controlled some of the major animal diseases like Foot and Mouth Disease (FMD) on the lines of Rinderpest, which has been totally eradicated from the country.

3. Although FMD is not very much a killer disease, it causes direct economic losses to the livestock growers by adversely affecting the performance of the animals and such losses has been estimated to the tune of US$4.45 billion per year (Rs. 20,000 crores). Apart from this, the prevalence of the disease also affects export potential of the livestock industry as a result of trade barrier that prevents export of livestock and their products to countries free from this disease. FMD is also a trans-boundary disease for which it is important for countries like India with diverse, vast and unique animal genetic resource to take a lead and intensively participate in controlling this disease in the SAARC Region. Considering the urgency to manage this disease in the country, FMD-Control Program was jointly launched by DAH&DF and ICAR during Xth plan period and I am happy to note that there has been a considerable reduction in the incidence of this disease, particularly in those states covered under the program. Since India could manage the disease through the above program, it has become important to test the efficacy of such a program in the neighboring SAARC countries owing to the trans-boundary nature of the disease. I am happy to learn that a step has already been taken in this direction by offering diagnostic services to the neighboring countries for detection of FMD virus in suspected clinical samples.

4. At present, knowledge regarding the epidemiology of FMD in the South Asian region in general and India in particular is known to a major extent by the research conducted at the Project Directorate on FMD. In order to meet the Sanitary & Phyto-sanitary requirements and also to control FMD, all laboratories, irrespective of the FMD situation in the country (free or endemic), manipulation of FMD virus must be carried out under high containment conditions and the safety precautions must preclude escape of even a single infectious FMD virus from the laboratory. It is again a matter of pride to inform you that PD-FMD could earn the referral status of OIE (Office International Epizootics) for FMD, obviously due to the adherence of international protocols in developing diagnostic capabilities.

5. The prevalence of the disease, as monitored by the scientists, indicates that this disease is almost a mirror image of the world-wide global economic –high-income, industrialised countries being generally free from FMD, while the low –income countries suffering from food deficits are reporting persistent endemicity. As per study conducted by OIE, FMD has been ranked as one of the most important diseases that affects livestock productivity in small holders especially in Asia. Consultative document of FAO has also identified FMD as one of the key impediments for the poor livestock farmers to formal market access.

6. It is thus clear that unless we make sincere efforts to contain and manage this disease, we will be far away from achieving the targeted production of 160-170 million tones of milk by 2020. Besides this, the poverty alleviation program of the UPA Govt. through livestock centric employment agenda, among others, will also be difficult to be achieved, unless the huge economic loss due to diseases like FMD is checked through scientific and development initiatives.

7. The Project Directorate on Foot and Mouth conducts surveillance and research pertaining to epidemiology of FMD in the different regions of the country through its regional units in almost all the states and are primarily involved in collection of epidemiological data, clinical samples and laboratory diagnosis besides advising on the control measures to be undertaken. Availability of such laboratories is a boon considering that any suspected outbreaks can be attended to for undertaking needed control measures as quickly as possible.

8. In order to strengthen regional capabilities and competiveness in achieving excellence in FMD diagnostic capabilities at a faster pace, Animal Production and health Commission for Asia and Pacific (APHCA) had recommended establishment of a Regional Reference Laboratory for FMD in its meeting held at Bangalore in 1995. Another objective of this recommendation was to produce trained manpower in the region in the field of FMD diagnosis, surveillance and epidemiology as well as to help in establishing FMD laboratories in the member countries. I am happy to tell you that today, we are laying the foundation of this laboratory with a major focus on achieving the intended intention of the recommendation mentioned above. We have, for this purpose, already sanctioned around Rs. 45 crores during XI Plan period to facilitate its establishment within a reasonable period of time.

9. I hope that this laboratory would be beneficial for both the country as well as other SAARC nations in developing required human resource, diagnostics and vaccines to control FMD and eventually help the region to be free from this disease.

Thank You

ADDRESS BY

PROF. K.V. THOMAS

MINISTER OF STATE FOR AGRICULTURE
CONSUMER AFFAIRS, FOOD AND PUBLIC DISTRIBUTION
GOVT. OF INDIA

Talking points on Foundation Stone Laying Ceremony of the Project Directorate on Foot and Mouth Disease

1. It is indeed a great privilege for me to associate myself in this Foundation stone laying ceremony of the International and National Centre for Foot and Mouth disease here at Aragul, Bhubaneswar.
2. The country is the home of around 200 million people living below poverty line, 80 million protein-energy malnourished children and a sizeable women population suffering from anemia.
3. All out efforts are required to boost up livestock sub sector where 70 million population is directly involved to produce quality food of livestock origin both for domestic consumption and export.
4. Department of Animal Husbandry, Dairying & Fisheries, Ministry of Agriculture, Government of India in their vision document calculated the need of 20 gram of protein per person per day in addition to protein sources from cereal and other sources.
5. Out of this requirement, 50% i.e. 10 gram is targeted from milk. 20% i.e. 4 gram from each of meat and fish and 10% i.e. 2 gram from eggs.
6. To achieve this, milk production (@ 250 ml per day for non-vegetarian and @ 500 ml for vegetarian population) has to go up around 160 million ton from the present level of 100 million ton by 2020.
7. The requirement of meat and eggs are estimated to be to the tune of 10.58 million ton and around 90-120 billion eggs respectively.
8. In order to achieve the target, suitable technologies to support livestock production optimization will have to be put in place.
9. We are also building up our capabilities and competitiveness to manage the import diseases specially after the complete eradication of disease like Rinderpest.
10. FMD is one such disease which is causing severe economic loss to the poor livestock grower.
11. In order to develop suitable diagnostic tools and vaccines, ICAR established a Project Directorate on FMD at Mukteswar which has been providing technology backstopping to the development departments to control this disease.
12. Considering the capacity that has been built in the country on surveillance, monitoring and control of FMD, International organization like OIE desired that India takes a lead in facilitating competence development on FMD in the SAARC region. Accordingly, ICAR approved the plan.
13. While complimenting the scientists on their achievement thus far, I look forward with interest the success emanating from this newly planned centre as controlling this disease will tantamount to saving around Rs. 20,000 crores annually.

This address was delivered on the occasion of Foundation Stone Laying ceremony of International Centre for Foot and Mouth Disease on 5th July, 2009 at Aragul, Near Bhubaneswar

11.5 Publications

1. J. K. Mohapatra, P. Priyadarshini, L. Pandey, S. Subramaniam, A. Sanyal, D. Hemadri and B. Pattnaik (2009). Analysis of the leader proteinase (L^{pro}) region of type A foot-and-mouth disease virus with due emphasis on phylogeny and evolution of the emerging $VP3^{59}$-deletion lineage from India. ***Virus Research*.** 141, 34-46.
2. J. K. Mohapatra, A. Sahu, L. Pandey, A. Sanyal, D. Hemadri and B. Pattnaik (2009). Genetic characterization of type A Foot-and-Mouth Disease virus 3A region in context of the reemergence of $VP3^{59}$-deletion lineage in India. ***Infection, Genetics and Evolution.*** 9, 483-492.

Abstracts

1. J. K. Mohapatra, P. Priyadarshini, L. Pandey, S. Subramaniam, A. Sanyal and B. Pattnaik (2008). Analysis of the leader proteinase (L^{pro}) region of type A foot-and-mouth disease virus with due emphasis on phylogeny and evolution of the emerging $VP3^{59}$-deletion lineage. **XXIII Annual Conference of IAVMI,** 25-26 Nov. 2008, IVRI, Izatnagar, U.P.
2. J. K. Mohapatra, P. Priyadarshini, L. Pandey, A. Sanyal and B. Pattnaik (2008). Comparative genomics of type A foot-and-mouth disease virus from India at 3C protease ($3C^{pro}$) *vis a vis* VP1 coding region. **XXIII Annual Conference of IAVMI,** 25-26 Nov. 2008, IVRI, Izatnagar, U.P.
3. S. Saravanan, D Hemadri, A sanyal, R.P.Tamilselvan, K. Muniswamy and B. Pattnaik. (2008).Detection of foot and mouth disease virus in ELISA negative clinical samples by multiplex PCR. XXIII annual conference of IAVMI and National symposium, November 25-26, IVRI, Izatnagar.
4. B. Pattnaik, D. Hemadri, S. Pawar, A. Sanyal. Emerging challenges in Vaccinology of FMD vaccine. National Seminar on Veterinary Biologicals (ILDEX), New Delhi, 23 August 2008, pp 16-18.
5. B. Pattnaik, S. Pawar, R. P. Tamilselvan, A. Sanyal. Economic Importance of FMD and its Control Strategy in India. Compendium of winter school on Economic Appraisal of livestock disease control projects. Dept of livestock economics, statistics and IT. IVRI Bareilly. pp 184-187. 12 Feb 09 to 4 March 09.

Participants of training cum workshop on DIVA at PDFMD, Mukteswar

Training on DIVA in progress

12.0 Education and Training

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.

12.1 Capacity building and Human resource development

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.

Two training *cum* workshop sessions of six days each on "recombinant 3AB3 nonstructural protein based indirect ELISA for differentiation of FMD infected from vaccinated animals (DIVA)" for 37 researchers from network units and private FMD vaccine manufacturing companies were organized.

A total of seventeen scientist form regional centers and network unit has been trained to carryout LPB ELISA at PDFMD.

12.2 Participation in Training

1. Dr. J.K. Mohapatra 21 days DBT, New Delhi sponsored training from 10-6-2008 to 1-7-2008 at Division of Virology, IVRI, Mukteswar on "Modern methods and their application in the diagnosis of animal viral diseases"
2. Dr. R.P. Tamil Sevan and Dr.S. Sararvanan attented ICAR Training-cum-Workshop on IP and Technology Management CCS Haryana Agricultural University, Hisar 19-21. May 2008 Theme: procedures of patenting
3. Dr. S. Sararvanan and Dr. Sachin S. Pawar attended 21 days CAS training programme on development of contents for online e-learning systems from 10th September to 30th September at IASRI, New Delhi
4. Dr. P. Rameshkumar attented 3 days workshop on "Genetic monitoring techniques for Laboratory rodents"- organized by ACTREC Navi Mumbai on Nov 12-14, 2008
5. Dr. P. Rameshkumar attended winter School on "Molecular diagnostic techniques for Zoonotic and foodborne diseases"- Feb 07-27, 2009 at IVRI, Izatnagar (UP).

12.3 Participation in Conference/ symposium

Scientists of PDFMD participated in following meetings

1. National Symposium on "Newer Concept & Strategies for Disease Diagnostics & Immunoprophylactics for Enhancing Livestock Health & Production" held at Indian Veterinary Research Institute, Izatnagar from 25 to 26 Nov 2008.
2. National workshop on integrated livestock farming systems in the foot-Hills of Himalayas (HILF-2008), May 17-18, 2008.Mukteswar
3. Second meeting of sero-monitoring under FMD-Control Programme (FMD-CP) held at Mathura on 19th July 2008.
4. 19th Annual Scientists Meet on FMD, Shimla (HP) from18-05-09 to 20-05-09.

13.0 Personnel Milestones

13.1 Award

Dr. J.K. Mohapatra received Indian National Science Academy Medal for Young Scientist (2009) by INSA, New Delhi.

13.2 Joining

Dr. Sachin S. Pawar (Animal Biotechnology), Dr. P. Rameshkumar (Veterinary Pathology) and Dr. K Muniswamy (Animal Biotechnology) joined PDFMD as scientist consequent upon their completion of FOCARS training at NAARM.

14.0 Acknowledgements

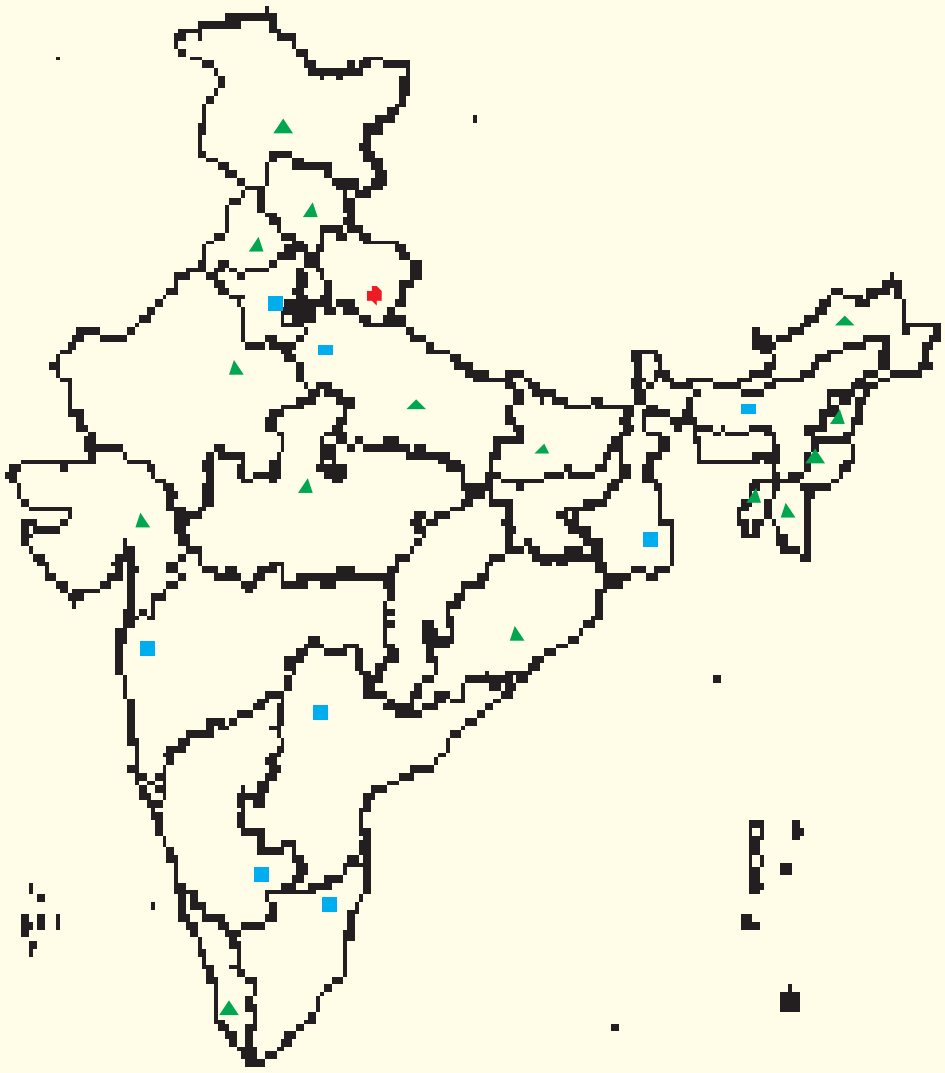
WE express our deep sense of gratitude to Deputy Director General (Animal Sciences) ICAR, and ADG (Animal Health), ICAR for providing all the necessary financial and infra-structural facilities and providing the guidance. We also express our sincere gratitude to Station-In-charge, Assistant Administrative Officer, Assistant Finance & Accounts Officer and other staffs of IVRI, Mukteswar for their help and cooperation in the smooth functioning of the PD on FMD. We also wish to express our appreciation to the technical supporting and office staff of the Project Directorate for their excellent assistance.

Project Directorate on FMD

■ Regional centers (8)

Central Laboratory Mukteswar

▲ Network units (15)



PDFMD
ICAR