Sub: Sampling Plan and SOPs for carrying out Sero Surveillance and Sero Monitoring under National Animal Disease Control Programme for Foot and Mouth Disease.

Under the National Animal Disease Control Programme for Foot and Mouth Disease all the States are required to carry out Sero Surveillance and Sero Monitoring. Accordingly, the State wise sampling plan for carrying out Sero Surveillance and Sero Monitoring under National Animal Disease Control Programme for Foot and Mouth Disease as developed by ICAR-NIVEDI and ICAR-DFMD is enclosed herewith for reference.

Also the SOPs for carrying out Sero Surveillance and Sero Monitoring under National Animal Disease Control Programme for Foot and Mouth Disease are also enclosed.

All the States are requested to strictly follow the above guidelines for Sero Surveillance and Sero Monitoring under National Animal Disease Control Programme for Foot and Mouth Disease.

With best regards,

Yours faithfully

(Mihir Kumar Singh)

To

Addl Chief Secretary/ Principal Secretary, Animal Husbandry all States and UTs.

Copy to

1. Commissioner/ Director, Animal Husbandry all States and UTs.
2. CEO, LDB
3. Director, ICAR, DFMD, Bhubaneshwar, with request to forward it to all FMD laboratories under DFMD for necessary action.
Protocol for carrying out for Sero-surveillance for Foot and Mouth Disease under National Animal Disease Control Programme

Introduction

Under the National Animal Disease Control Programme for Foot and Mouth Disease, Sero-Surveillance is one of the important activity which is aimed at determining the presence/ absence of disease or infection, or detecting as early as possible exotic or emerging diseases.

The Sero-Surveillance plan for FMD has been designed by ICAR-National Institute of Veterinary Epidemiology and Disease Informatics, Bengaluru and ICAR- Directorate of FMD. It is based on the two stages design stratified random sampling i.e one stage choosing the epidemiological units; second stage choosing the animals within the units. The Sample size calculation (number of epidemiological units and number of animals to be included in the survey) has been done taking into account the sensitivity and specificity of the test, aiming for a minimum design prevalence (that is the threshold detection level) of 1% between first stage level (i.e. village) and 5% within village.

Sampling Plan

The State wise, district wise and village wise number of animals to be sampled is provided in the sampling plan.

- **Animals to be covered:** Cattle and buffalo
- **Age Group:** 6 to 18 months of age
- **Frequency of sample collection:** 4 to 5 months after the previous vaccination.
- **Test employed:** The Sero-surveillance will be carried out by the 3 AB3-Non Structural protein based ELISA. (List enclosed at Annexure B). DFMD will provide the testing kits to their units and will coordinate the testing procedures.

Time line

<table>
<thead>
<tr>
<th>Timeline</th>
<th>Activities</th>
<th>Responsibility</th>
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<tbody>
<tr>
<td>D1*</td>
<td>Vaccination</td>
<td>Vaccinators</td>
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<tr>
<td>D1 + 2.5 months</td>
<td>Training of Veterinarians</td>
<td>SMU in coordination with State FMD Unit</td>
</tr>
<tr>
<td>D1 + 3.5 months</td>
<td>Procurement/ Arrangement of logistics</td>
<td>SMU and DVO</td>
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<td>D1 + 4 to 5 months</td>
<td>Sero surveillance (Sample collection)</td>
<td>DVO/ District Poly Clinic in coordination with BVO</td>
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<td>D1 + 5.5 months</td>
<td>Testing of samples #</td>
<td>State FMD Unit</td>
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<td>D1 + 6.5 months</td>
<td>Results</td>
<td>State FMD Unit to provide results to DFMD within 15 days and DFMD to submit the compiled results with interpretation to DAHD within 1 month.</td>
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</tbody>
</table>

*D1 = Day of Vaccination
# uniform SOPs of testing and training will be provided by DFMD to all laboratories one month before the testing starts.
Role of State Monitoring Unit/ State AH Director

a) Receive the Surveillance Plan for the State from DAHD and circulate it to the District level for implementation. The State wise, district wise and village wise number of samples to be collected has been detailed in the sampling plan.

b) SMU shall ensure that the required logistics for sample collection are available at district / block level well i.e 15 days before start of surveillance plan.

c) SMU shall ensure availability of trained manpower for carrying out sero surveillance and also ensure extension activities, training to field staff etc.

d) SMU shall ensure diagnostic facilities to all the laboratories engaged in the sero-surveillance as per annexure B.

e) SMU shall establish liaison with concerned ICAR laboratories / ICAR-DFMD / ICAR-NIVEDI, etc. for sero-surveillance. SMU shall also ensure that the result of sero-surveillance is conveyed to DAHD within 1 months of sample collection.

Role of District Veterinary Officer/ PolyClinic

a) DVO shall be responsible for training of staff engaged in sero-surveillance programme 2.5 months after the date of vaccination programme and should prepare calendar of operation with the help of District Animal Husbandry officer and Block Officers.

b) DVO shall be responsible in ensuring that required logistics for sero-surveillance including man-power for carrying sero-surveillance are in place at least 3.5 months after the start of the vaccination programme. Necessary transportation facility must be provided for the sample collection team.

c) DVO shall supervise sero-surveillance programme and provide all necessary required infrastructure facilities, transportation arrangement, sample collection vials and vaccutainers, etc.

d) DVO shall provide publicity at local level at least 1 month before actual sample collection using leaflets, pamphlets, posters etc., to village panchayat, cattle herds etc.

e) Collection of samples is the primary responsibility of the DVO with the help of the District Diagnostic Laboratory and District PolyClinic in coordination with the block veterinary officer

f) The blood is usually collected from jugular vein. After collection the sample should be allowed to stand in a cool area out of direct sunlight for at least 15 minutes to allow clot formation.

g) At least 2 ml of serum should be submitted to the laboratory and for that, 4-8 ml of whole blood must be collected without anticoagulant.

h) The sample should be centrifuged or if not available, stand the sample in an upright position overnight in a cool box or refrigerator. DVO has to ensure that no hemolysis or RBC are present in the serum samples.
i) Place the serum in a sterile tube with proper labelling and send to the laboratory under cool condition.

j) DVO shall ensure maintenance of cold chain, as prescribed, in ice boxes at the time of transport of serum samples.

k) If despatch to a laboratory is delayed, serum samples should be frozen and stored at -20 C.

l) Serum should be accompanied by proper epidemiological data (Annexure A)

m) DVO shall supervise proper disposal of bio-medical waste generated during sero-surveillance

n) The District Veterinary Officer is responsible for facilitating the transportation of samples from the District Laboratory to FMD Testing Laboratory at the State level within 7 days after collection of samples.

Role of District Diagnostic Laboratory/ State Laboratory/ FMD Network Unit

a) The DDL and FMD Unit at the State level should finalise the requirement of logistics 1 month before the sample collection and submit the same to the District as well as the State Monitoring Unit.

b) The DDL and FMD Unit will prepare a check list for collection of samples for sero-surveillance

c) The DDL and FMD Unit should train the personnel on sample collection involved in sero-surveillance sample collection at least 2.5 months after the date of vaccination.

d) The District Lab will keep a record of all the samples collected and dispatch the samples to State FMD Unit for testing within 7 days.

e) The State FMD Unit will follow the test procedure as designed by the DFMD for testing of samples.

f) The State FMD Unit will compile the test results and submit it to the DFMD and State Monitoring Unit within 1 month of receiving the samples.

Role of ICAR-DFMD

a) Develop/ Modify the Standard Protocol for testing of samples for sero-surveillance as per OIE guidelines.

b) Training of the State Laboratories/ FMD Units for testing of samples one month before the start of surveillance.

c) Analysis of the test results from different State Units and submission to DAHD for suitable action within 1 month.
d) DFMD will investigate all NSP positive samples. However, in the States proposed for FMD Disease Free Zones all the NSP positive samples will be investigated for detection of any virus circulation.

Role of ICAR-NIVEDI

a) For every two round of vaccination the sampling plan to be prepared in consultation with DFMD and submitted to DAHD 1 month before the start of vaccination round in the State.

Role of CMU( DAHD)

a) Monitoring of analysed results received from DFMD

b) Advise concerned State in case of any improvement or change are required for effective implementation.

c) Preservation of data for sharing with OIE for endorsement of programme whenever required.
Protocol for carrying out for Pre and post vaccination Sero-monitoring for Foot and Mouth Disease under National Animal Disease Control Programme

Introduction

Vaccination against FMD is the most important tool to combat the disease. Since most factors can influence the effectiveness of vaccination against FMD, the programmes must be monitored continuously to identify any findings and to ensure sustained control programme.

Estimation of immunity of the population targeted for protection by vaccination is the core of post vaccination monitoring as it is the key indicator of how well vaccination has been carried out and whether or not protection against infection is likely. Serological tests that detect antibodies towards the virus structural proteins are suited to measuring protective antibody responses induced by vaccination.

Serum samples at the time of vaccination (0 day of vaccination) and 21 to 28 days post vaccination will be collected by the respective state AH Department and tested by ICAR-DFMD laboratories for estimation of level of serotype specific antibodies.

The sampling plan for FMD has been designed by ICAR-National Institute of Veterinary Epidemiology and Disease Informatics, Bengaluru and ICAR- Directorate of FMD. It is based on the two stages design stratified random sampling i.e one stage choosing the epidemiological units; second stage choosing the animals within the units.

Sampling Plan

The State wise, district wise and village wise number of animals to be samples is provided in the sampling plan.

1. Animals to be covered: Cattle and Buffalo
2. Age Group:
   - In the new stratified sampling frame, population immunity at individual animals is followed. T
   - The district/village wise number of animals to be samples should be as per the sampling plan designed by ICAR-NIVEDI.
   - However the State has to distribute the samples to be collected from each village among three age categories i.e 6-12 months, 12-24 months and more than 24 months at the ratio of 5:4:1.
3. Frequency of sample collection: Animals are sampled at two different points in time to assess the changes in population immunity. Sampling at time 0 (day of vaccination) and again after 21-28 days post vaccination to assess increase in the level of population immunity.
   It should be noted that, in particular round, pre and post vaccination samples should be collected from same animal. However in subsequent rounds it is not essential that the same animals are as sampled as sampled in previous round.
4. Test employed: Samples will tested by Solid Phase competitive ELISA by State Units of ICAR-DFMD (List enclosed at Annexure B). DFMD will provide the testing kits to their units and will coordinate the testing procedures.
## Time Line

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<tr>
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<tbody>
<tr>
<td>1.5 months before vaccination</td>
<td>Training of Veterinarians</td>
<td>SMU in coordination with State FMD Unit</td>
</tr>
<tr>
<td>15 days before vaccination</td>
<td>Procurement/Arrangement of logistics</td>
<td>SMU and DVO</td>
</tr>
<tr>
<td>D1 *</td>
<td>Initial Sample Collection</td>
<td>DVO/ District Poly Clinic in coordination with BVO</td>
</tr>
<tr>
<td>D1</td>
<td>Vaccination</td>
<td>Vaccinators</td>
</tr>
<tr>
<td>D1 + 21 to 28 days</td>
<td>2nd round sample collection from same animal</td>
<td>DVO/ District Poly Clinic in coordination with BVO</td>
</tr>
<tr>
<td>D1 + 2 months</td>
<td>Testing of samples #</td>
<td>State FMD Unit</td>
</tr>
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<td>D1 + 2.5 months</td>
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*D1 = Day of Vaccination

# uniform SOPs of testing and training will be provided by DFMD to all laboratories one month before the testing starts.

### Role of State Monitoring Unit/State AH Director

a) Receive the sampling plan for the State from DAHD and circulate it to the District level for implementation. The State wise district wise and village wise number of samples to be collected has been detailed in the sampling plan.

b) SMU shall ensure in advance that the required logistics for sample collection are available at district / block level 15 days before start of sero-monitoring plan

c) SMU shall ensure availability of trained manpower for carrying out sero monitoring and also ensure extension activities, training to field staff etc

d) SMU shall ensure diagnostic facilities to all the laboratories engaged in the sero-monitoring as per annexure B.

e) SMU shall establish liaison with concerned ICAR laboratories / ICAR-DFMD / ICAR-NIVEDI, etc. for sero-monitoring. SMU shall also ensure that the result of sero-monitoring is conveyed to DAHD within 1 month of sample collection.

### Role of District Veterinary Officer/ PolyClinic

a) DVO shall be responsible for training of staff engaged in sero-monitoring programme 1.5 months before the date of vaccination and should prepare calendar of operation with the help of District Animal Husbandry officer and Block Officers.

b) DVO shall be responsible in ensuring that required logistics for sero-surveillance including man-power for carrying sero-monitoring are in place at least 15 days before the start of the...
vaccination programme. Necessary transportation facility must be provided for the sample collection team.

c) DVO shall supervise sero-monitoring programme and provide all necessary required infrastructure facilities, transportation arrangement, sample collection vials and vaccutainers, etc.

d) DVO shall provide publicity at local level at least 1 month before actual sample collection using leaflets, pamphlets, posters etc., to village panchayat, cattle herds etc.

e) Collection of samples is the primary responsibility of the DVO with the help of the District Diagnostic Laboratory and District PolyClinic in coordination with the block veterinary officer.

f) The blood is usually collected from jugular vein. After collection the sample should be allowed to stand in a cool area out of direct sunlight for at least 15 minutes to allow clot formation.

g) At least 2 ml of serum should be submitted to the laboratory and for that, 4-8 ml of whole blood must be collected without anticoagulant.

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j) DVO shall ensure maintenance of cold chain, as prescribed, in ice boxes at the time of transport of serum samples.

k) If despatch to a laboratory is delayed, serum samples should be frozen and stored at -20° C.

l) Serum should be accompanied by proper epidemiological data (Annexure A)

m) DVO shall supervise proper disposal of bio-medical waste generated during sero-monitoring.

n) The District Veterinary Officer is responsible for facilitating the transportation of samples from the District Laboratory to FMD Testing Laboratory at the State level within 7 days of sample collection.

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a) The DDL and FMD Unit at the State level should finalise the requirement of logistics 1 month before sample collection and submit the same to the District as well as the State Monitoring Unit.

b) The DDL and FMD Unit will prepare a check list for collection of samples for sero-monitoring.
c) The DDL and FMD Unit should train the personnel on sample collection involved in sero-monitoring sample collection at least 1.5 months after the date of vaccination.

d) The District Lab will keep a record of all the samples collected and dispatch the samples to State FMD Unit for testing within 7 days.
e) The State FMD Unit will follow the test procedure as designed by the DFMD for testing of samples.

f) The State FMD Unit will compile the test results and submit it to the DFMD and State Monitoring Unit within 1 month of submission of samples.

**Role of ICAR-DFMD**

a) Develop/ Modify the Standard Protocol for testing of samples for sero-monitoring as per OIE guidelines.

b) Training of the State Laboratories/ FMD Units for testing of samples one month before the start of vaccination programme.

c) Analysis of the test results from different State Units and submission to DAHD for suitable action within 1 month.

**Role of ICAR-NIVEDI**

a) For every two round of vaccination the sampling plan to be prepared in consultation with DFMD and submitted to DAHD 1 month before the start of vaccination round in the State.

**Role of CMU( DAHD)**

a) Monitoring of analysed results received from DFMD

b) Advise concerned State in case of any improvement or change are required for effective implementation.

c) Preservation of data for sharing with OIE for endorsement of programme whenever required.