SAMPLING, SHIPPING AND TESTING OF FOOT AND MOUTH DISEASE **VIRUS INACTIVATED SAMPLES**

- Søren Alexandersen (1) & Manzoor Hussain(2)
 - (1) National Centre for Foreign Animal Disease, CFIA, Winnipeg, R3E 3M4 Manitoba, Canada
 - (2) Food and Agriculture Organization of the United Nations Pakistan, NARC, Islamabad, Pakistan

Introduction: FMD is endemic in many countries and causes significant economic losses despite efforts of control by vaccination. Efficient vaccination may be hampered by limited access to/excessive costs of quality vaccines and/or lack of an vaccination strategy based on epidemiological information. Excessive costs/limited access to quality vaccines may be mitigated by International support, but the methods for generating epidemiological information and strain characterisation need a leap forward. Previous experimental studies have used swab samples collected directly into lysis buffer followed by real-time RT-PCR analysis and the next step was to use this approach under field conditions facilitating easy shipping and downstream analyses.

Materials and Methods: The Landhi Cattle Colony (LCC) outside Karachi, the largest dairy colony in Pakistan and the biggest buffalo colony in the world, provided a suitable study population. Due to its size and fragmented vaccination coverage, together with the Pakistani husbandry tradition of frequent transportation of animals to and from dairy colonies, FMDV infection is prevalent in LCC. During the period April 2006 to April 2007, we collected mouth swab samples of randomly selected, non-clinically affected herds, from non-clinically affected animals in herds with clinical evidence of a prior FMD outbreak and from nonclinically affected animals in herds in which acute FMD was obvious in at least one animal. Swabs were immediately placed in Qiagen RLT buffer, which immediately stabilised the viral RNA and rendered the sample non-infectious. A questionnaire was used to gather health status and vaccination information and the location of the herd was determined by GPS. The laboratory-based analysis employed real-time RT-PCR as well as partial or full genome sequencing.

Results: Sampling of mouth swabs directly into lysis buffer followed by shipment of the samples either directly, or via the National Reference Laboratory for RNA extraction and cDNA preparation, to an International Laboratory successfully preserved FMDV nucleic acids. Such samples contain no infectious FMDV, can relatively easy be shipped as diagnostic samples and provided valuable information on prevalence of FMDV infection in LCC. Moreover, such samples could be sequenced provided they contained a reasonable amount of FMDV RNA. Discussion: As shown here, stabilised and inactivated swab samples can easily be collected and shipped internationally and may be used to determine the temporal-spatial distribution and prevalence of FMDV infection. Further analysis by sequencing can be used to characterise circulating strains of FMDV and thus, is a viable and valuable alternative to shipment of epithelial samples, containing infectious virus.





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