Antigenic variation of Foot and Mouth Disease Virus - An Overview

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Abstract

Foot and mouth disease (FMD) is a highly contagious viral diseases of cloven-hoofed animals, caused by FMD virus, a single molecule of linear positive sense, with single stranded RNA of size 7.2– 8.4 kb. Antigenic variation is one of the striking characters of FMD virus. It is a process by which an infectious organism alters its surface proteins in order to evade a host immune response and is associated with mutation leading to amino acid replacement. These changes may result either in the field or in the laboratory leading to the development of a new strain of virus which totally differs from the circulating field strain challenging the vaccine strains use for controlling the diseases. The high sequence variability found in VP1 region of this virus accounts for the low cross-reactivity observed among different serotypes of FMDV and also due to high degree of antigenic variation may be attributed to different reasons like high rate of mutation, genetic recombination, quasispecies nature of the virus and continuous circulation of the virus in the field, which is a main loophole for severe economic loss in livestock productions.
Key words: antigenic variation, quasispecies, FMD.

Introduction

Foot and mouth disease is group under list-A of OIE classification of diseases, initially describe in the 16th century and recognized by Loeffler and Frosch (1898), is the first animal pathogen identified as a virus. Since, then it has been most contagious viral disease of cloven-hoofed animal.

Still being major global animal health problem and with its recent outbreak in developed countries of their significant economic impact have increased the concern of Government worldwide. About 2500 outbreaks are reported annually in India. The direct loss due to FMD in India is roughly estimated as 2250 crores of rupees, and indirect loss is much more, L. Mathew and D. G Menon (2008). So, knowing every detail of the virus and the disease is a necessity to go for controlling as early as possible. The main controlling method adopted is either slaughter or vaccination but in country like India and many other developing countries which vaccination is the only option.

However, in this method regular follow up of vaccination is not the crucial point but monitoring whether the vaccine in used have the same strain as compare to the circulating field strain or whether it is giving appropriate immunity, which it should give. All these problems are the consequences of antigenic variation of the FMD virus, which avail even within the serotype.

The Foot and Mouth Disease Virus

Classification :
Genome - Single stranded positive sense RNA
Order - Nidovirales
Family - Picornaviridae
Genus - Aphthovirus
Species - FMDV

Virus have seven distinct serotype. They are serotype O, A, C, Asia 1, SAT 1, SAT 2 and SAT 3.

Virion structure: The structure of the FMDV particle was first resolved for serotype O1BFS 1860 by x-ray diffraction analysis. Since then, other serotype of FMDV have been crystallized and analysed. They are non-enveloped, 27 nm in diameter approximately spherical and relatively smooth with capsid consisting of a densely packed icosahedral arrangement of 60 copies each consisting of four polypeptides VP 1, 2, 3 and 4.

Genomic RNA is a single molecule of linear positive sense, single stranded RNA with 7.2 – 8.4 kb in size. It is infectious in nature and has polycytidylic acid (poly C tract) and is polyadexylated at its 3' end. Have VPg (virus protein genome linked) protein, linked covalently to its 5' end.

Structural Proteins of FMDV

The four main structural proteins of FMDV are VP1, VP2, VP3 and VP4. VP1-3 have a molecular weight approximately 24 KDa each and VP4 approximately 8.5 KDa. VP1-3 have surface component and

VP4 is internal, buried within the capsid and has a myristyl group covalently attached to its N terminus.

VP1, VP2 and VP3 are structurally rather similar to one another, each being composed of a wedge-shaped, eight stranded Beta–barrel and differing primarily in the size and conformation of the loops that occur between the strands and also in the extensions of their amino and carboxy termini. VP1 proteins are located around the 5-fold axes of icosohedral symmetry, and VP2 and VP3 alternate around the two- and three fold axes, the amino terminal extension of these three proteins form an intricate network on the inner surface of the protein shell. The small, myristylated protein VP4, is located entirely at the inner surface of the capsid, probably in contact with the RNA.

Non-Structural Proteins of FMDV

There are about seven to nine non-structural (non-capsid) proteins, viz., L (Lab and Lb) and 2A proteins; 2B, 2C and their precursor (2BC); 3A, 3AB, and 3D (VPg), 3ABC. Both Lab and Lb of FMDV are proteases with multiple activities. 3^{Cpro} is a protease and an RNA-binding protein, whereas 3^{Dpro} is an RNA polymerase and is involved in proteolysis in the form of 3CD. 2A pro and B^{Cpro} not only participate in the virus replication but also appear to inhibit essential host function (RNA transcription and protein synthesis) and VPg (3B) plays a role in the initiation of viral RNA synthesis (Lawson, M.A.A, and B.L. Semler, 1991).

Antigenic Structure of FMDV

Antigenic site on the surface of the FMD virion have been identified for five of the seven serotypes of the virus (SAT1 and SAT 3 being the only exception). Serological studies showed that different serotypes of FMDV shared a highly variable region of VP1, comprising residue 135 to 155, as one of the major antigenic sites of the virus. Several overlapping Beta-cell epitopes are located within this region and are able to induce both neutralizing and non-neutralizing antibody responses. (Strohmaier *et al.*, 1982). The high sequence variability found in this region accounts for the low cross-reactivity observed among different serotypes (Cheung *et al*., 1983).

This immunodominant region was seen to correspond to the loop which connects Beta-sheet G and H of the VP1 Beta-barrel, named the GH loop (Acharya *et al*., 1989). Within this loop antigenic site A is present and very likely to face substitution because of high mutation rates during RNA replication resulting in antigenic variants. This site also contains the RGD (Arg-Gly-Asp) motif receptor binding recognition sequence and this is the major antigenic site.

Other important antigenic site which have been identified in several FMDV serotypes are the C-terminal stretch of VP1 (which together with the GH-loop, defines the main antigenic site 1 in serotype O), or site involving different loops from the three accessible viral protein. (McCahon and Crowther, 1989). Another is the site D which involves at least part of the C-terminal region of VP1, the B-B knob of VP3 and the B-C loop of VP2 located near the capsid threefold axis.

Antigenic Variation

Antigenic variation is the process by which an infectious organism alters its surface proteins in order to evade a host immune response and is associated with mutation leading to amino acid replacement. This change in antigenic profile may occur as the pathogen passes through a host population (also called antigenic diversity) or may take place in the originally infected host. The strategy is particularly important for organism that target long-lived host, repeatedly infect a single host, and are easily transmitted. Pathogens that express these characteristics and undergo antigenic variation have a selective advantage over their more genetically stable counterparts.

RNA viruses are characterized by an error-prone RNA replication, which gives them great potential for variation (Domingo et al., 1990 and 1992). Antigenic variants result from the high mutation rates during RNA replication which allow FMD viruses to continuously evolve and adapt to new environments. Although most mutations will be detrimental and eliminated by natural selection (negative selection), others can be of value under the particular conditions where the virus is replicating and are therefore selected (positive selection) (Domingo *et al.*,1990 ; Fry et al., 1999; Novella et al., 1996; Baranowski et al., 1998). Antigenic variants have been isolated under variable conditions, such as in partially immune animals, persistently infected cattle4 and in cell culture (Diez *et al.*, 1990; Piatti *et al.*, 1995), in the latter case both in the presence or the absence of immune pressure (Borrego *et al.*, 1993 and Holguin *et al.*,1997). This antigenic diversity has serious implications in vaccine design since synthetic vaccines should include multiple independent epitopes in order to decrease the probability of selection of FMD viruses resistant to the immune response.

Types of Antigenic Variation

1. Antigenic variation in the field

Antigenic variations in the field increases with time and most probably result from immunogenic pressure placed on the virus by either the infected or vaccinated host species and to the selective pressure exerted in the field by virus replication in the animal having inadequate level of neutralizing antibodies (Domingo *et al.*, 2003). Genetic variants also arise during persistent infection of animal (Gebauer *et al.*, 1988; Salt, 1993; Malirat *et al*., Woodbury, 1995). Which give rise to virus population different from those that initiated the infection. This process cause changes in the sequence of the nucleotides of the amino acids and among other aspect changing the antigenic and immunogenic properties of the viral antigenic determinants.

2. Antigenic variation during propagation in cell culture

As virus is propagated in cell culture and as replication increase it increased every possibility of producing antigenic variant. Say, for example, in production of vaccine where a number of tissue culture passages are required, antigenic variant do developed which becomes a constraint in vaccine production (Sobrins *et al*., 1983; Bolwell *et al.*, 1989).

Causes of Antigenic Variation

1. High rate of mutation

FMD virus have very high mutation rate in the range of 10-3 to 10-5 per nucleotide site per genome replication, due to the lack of error correction mechanisms i.e. proof reading during RNA replication (Domingo and Holland. 1988; Drake and Holland. 1999). This high error rate leads to differences of FMD virus replicated genome from the original parental genome and thus resulting in antigenic variation.

2. Genetic recombination

Compare to DNA viruses, the RNA viruses are uncommon to recombination (there are probably no host enzymes for RNA recombination). But, picorna viruses show a form of very low efficiency recombination and study has shown that recombination was more likely to occur within the 3 half region of the genome coding for the NSP, and has not been demonstrated in the capsid-coding genes of FMDV (King *et al.*, 1985, King, 1988). However, a more recent study has suggested, that RNA recombination within the capsid-coding PI region of the genome may contribute to the genetic diversity in FMDV isolated from the field. Wilson *et al*., 1988 also demonstrated that the recombination events increased much more steeply with increasing secondary structure in the region encoding non-structural proteins compared with the structural protein-coding region of the genome.

3. Quasispecies nature of the virus

FMD virus being quasispecies in nature, high variability of FMDV population is manifested and when mutation lead to codon change it result in a change in the viral phenotype. And when, variant population are selected from the quasispecies pool of genome, either in the presence (Borrego *et al.*, 1993) or absence of immune pressure (Doming *et al.*, 1993; Holguin *et al.*, 1997) a new antigenic variant developed which is different from its origin.

4. Continuous circulation of the virus in the field

The rise of new variants is inevitably caused by continued circulation of the virus in the field and the quasispecies nature of the RNA genome (Domingo *et al.*, 2003; Haydon *et al*., 2001). As the virus continues to circulate in the field, continues to cause infection of susceptible host and remained persistence in their host, then with due course of time or probably due to the immunological pressure placed on the virus either by the infected or vaccinated host or either by the challenge in the environment and also to the selective pressure exerted in the field by virus replication in animals having inadequate level of neutralizing antibodies had somehow lead to the antigenic variation giving rise to virus population different from those that initiated the infection.

Antigenic Variation in absence of any detectable immune selection

Antigenic variation of viruses may occur upon virus replication in the absence of any detectable immune selection. The observation has been explained by two possible models.

First, residue at antigenic site may be involved in viral function unrelated to the evoking of an immune response. Such residues may be replaced as a result of selective forces unconnected to immune pressure, but the replacement may fortuitously contribute to antigenic variation. The second model termed the random change model, suggest that fluctuation in the genomic distribution of a quasispecies-triggered by any selective force other than immune selection, or by random sampling event may mediate the hitch-hiking of mutation occurring at those genomic site where mutation are more tolerated.

Study of Antigenic variation is important

- For virus classification

r• For selection of suitable vaccine strain.
- For epidemiological studies of outbreaks.
- For analysis of virus within countries where the disease is enzootic and
- In case of a possible deliberate introduction of vius, it will also have forensic value in tracking the source.

The great variability of the FMD virus continually produces new strains that must be classified according to pre-established standards to avoid confusion. And so, the knowledge of the antigenic and immunogenic characteristics of the vaccine strains and field strains, and the importance of the latter, had been the fundamental elements for classifying the new strains. To study any new strain availing in the field and to classify it, become an important element in knowing and controlling the disease. In country, like India were the disease is controlled by means of vaccination only, selection of suitable vaccine become the prime most important to achieve effective control. A vaccine strain, is said to be good enough when it covered up the circulating field isolate and this is so, when vaccine strains are antigenically related with majority of field isolates. Therefore, study of of antigenic relation between the field isolate and vaccine strain become a must in developing an ideal vaccine for controlling the disease. According to OIE, World Organization for Animal Health, Paris, guidelines, the serological relationship between field isolate and a vaccine virus (r value) can be determined by CFT (complement fixation test), ELISA (Enzyme Linked Immunosorbent Assay) and VNT (Virus Neutralisation Test). ELISA and VNT are recommended to be used as screening method whereas VNT methods, more definitive result.

Antigenic Variation - A Problem

'Antigenic variation represents an important adaptive strategy for this virus and may contribute to decrease vaccine efficacy in the field." (Feigelstock *et al.*, 1996). As a result of decrease vaccine efficacy in the field ultimately means decreased in prevention and control of the disease. Thus, antigenic variation is a major obstacle in controlling the disease (Peneina, 1977; Domingo *et al.*, 1980, 1990).

Conclusion

The high contagiousness of foot and mouth disease (FMD) and the great variability of the virus are factors that negatively affect the control of the disease and aggravate the socio-economic problems of the affected countries compelling them to adopt strong measures to control and eradicate the disease. In order for those measure to be affective, they must originate from the united determination of all those who, in one way or another, are dedicated to control the disease.

The control programs must unite the efforts of the livestock producers, the veterinary field services and the diagnosis, production and control laboratories, and must likewise maintain among them active, continuous information regarding the characteristics of the viruses active in the field, the performance of the vaccine and the epidemiological field situation. Additionally, FMD control programs must put forth every effort to implement new techniques to study the antigenic and immunogenic characteristics of the virus and the changes in its nucleic acid for its better identification. Thus will the process of eradicating the disease be accelerated.

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