ORIGINAL ARTICLE

The emergence of foot-and-mouth disease virus serotype O PanAsia-02 sub-lineage of Middle East–South Asian topotype in Bangladesh

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ABSTRACT

Objective: This research work was conducted for the molecular characterization of the circulating foot-and-mouth disease (FMD) virus in Bangladesh and revealed out their serotype.

Materials and methods: The *VP1* gene of six field isolates of FMD virus (FMDV) serotypes (two serotypes O, two serotypes A, and two serotypes Asia 1) was subjected for sequencing and phylogenetic analysis. Neighbor-joining trees were constructed by using the Molecular Evolutionary Genetics Analysis 6, having the field nucleotide sequences of FMDV and related sequences available in the GenBank.

Results: The nucleotide sequences of the *VP1* genes of serotypes O, A, and Asia-1 of the isolates revealed that overall isolates were 91%–100% similar to the isolates reported from Bangladesh and other neighboring countries. Among the isolates reported from Bangladesh, serotype O had 98%–100% identity, serotype A had 91%–100% identity, and serotype Asia-1 had 94%–100% identity. A phylogenetic analysis revealed that the FMDV serotype O PanAsia-02 sub-lineage was confirmed in Bangladesh under the Middle East–South Asian (ME-SA) topotype. On the other hand, we identified genotype VII (18) of Asia topotype (serotype A) and lineage C (serotype Asia-1). **Conclusion:** The FMDV serotype O PanAsia-02 sub-lineage was confirmed in Bangladesh under the ME-SA topotype for the first time. The extensive cross-border animal movement from neighbor.

boring countries may act as the source of diversified FMDV serotypes in Bangladesh.

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KEYWORDS

PanAsia-02; serotype; FMDV; Bangladesh



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Introduction

Foot-and-mouth disease (FMD) is an economically important and highly contagious viral disease of cloven-hoofed animals, including cattle, sheep, goats, and swine. [1]. The disease is caused by the genus *Apthovirus* belonging to the family *Picornaviridae* [2]. This RNA virus has seven serotypes, namely, A, O, C, Asia-1, and South African Territories 1, 2, and 3. FMD virus (FMDV) is subdivided into several serotypes, and each serotype may contain several subtypes [3]. The *VP1* gene is the

dominant epitope of capsid protein *VP1*, which is used for the detection of FMDV serotypes, and targeted for vaccine development. Within each serotype of FMDV, there is considerable antigenic diversity, and anti-sera against one strain of a serotype may not recognize other strains of the same serotype [2].

Rahman et al. [4] and Ali et al. [5] reported that FMDV serotypes prevailed in Bangladesh were A, O, C, Asia-1, and sub-type A22. From 1996, serotype C has not been reported in Bangladesh. Later, Chowdhury et al. [6], Hossen et al.

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[7], Nazneen et al. [8], Nandi et al. [9], Loth et al. [10], and Zinnah et al. [11] reported that the circulating FMDV serotypes in Bangladesh were A, O, and Asia-1. Among three FMDV serotypes, O is the most prevalent and the maximum outbreaks occurred with this serotype, and almost similar findings were reported in the other nine Southeast Asian countries [9–13]. The objective of this study was to characterize the circulating FMDV in Bangladesh sequence and phylogenetic analysis.

Materials and Methods

Six field isolates of FMDV serotypes (two serotypes 0, two serotypes A, and two serotypes Asia-1), which were previously confirmed by the previous work [7], were subjected for sequencing and phylogenetic analysis. Polymerase chain reaction amplicons of those six genes (*VP1* gene) were sequenced commercially. The sequence identity and multiple sequence alignment of nucleotide sequences were performed with the Clustal W algorithm as per the method of Thompson et al. [14]. The sequences were deposited in the GenBank and were aligned, and the phylogenetic analysis was performed by the neighbor-joining method [15] using the Molecular Evolutionary Genetics Analysis 6 software [16].

Results and Discussion

The VP1 genes from the six isolates were amplified and sequenced, and the accession numbers were received from the GenBank (Figs. 1–3 and Table 1). In this study, VP1 gene sequences of the FMDV serotypes O, A, and Asia-1 were compared with the corresponding serotypes that were previously reported from Bangladesh, India, Pakistan, Bhutan, and Nepal, revealing that these new Bangladeshi local isolates had 91%-100% identity (Table 1). The FMDV serotype O isolates (KT960948 and (KT982203) were 98%-99% similar to IND23/2012, IND172/2011, IND56/2011, IND35/2011, and IND52/2011, and 92% identical to NEP/6/2003, NEP/4/2003, PAK/L288/2005, PAK/1/2008, PAK/61/2006, BHU/24/2003, BHU/30/2004 BHU/31/2004, BHU/39/2004, BHU/40/2004, and BHU/49/2003 (Fig. 1).

The FMDV serotype A was first reported in Bangladesh in 2012 (accession KJ754939), having only

91.69% identity with the isolate of this study (Table 1). In 2016, the serotype had 91.38% identity (accession MK088171), whereas, in 2018, the serotype was 100% (accession MN968767) similar to our isolate (Table 1). This observed diversity in different locations of Bangladesh might be due to the known high rate of mutation capability of FMDV. On the other hand, the FMDV serotypes showed considerable similarities with Indian isolates (Fig. 2). This might be due to the cross-bordering of the serotype.

Similarly, the serotype Asia-1 (accessions KU159763 and KU159762) was 94%–99% identical to other Bangladeshi isolates reported between 2009 and 2013. Similar to serotype A, the isolates were closely related to Indian isolates (Fig. 3).

The FMDV serotype O reported in this paper has been placed under the sub-lineage PanAsia-02 under the Middle East-South Asian (ME-SA) topotype. This is a new addition along with the previous reports of FMDV under sub-lineage Ind-2001 [9,10,17]. In this study, the observed genetic variation of FMDV serotypes might be due to the difference in sample size, geographic locations, etc. According to Biswal et al. [18], currently, three sub-lineages of serotype 0, namely, Ind-2001, PanAsia-01, and PanAsia-02, are circulating in India and other South Asian countries. The PanAsia-02 lineage emerged in 2003, and since then, it causes outbreaks along with parent PanAsia-01 viruses. The lineage Ind-2001 was first identified in the year 2001, re-emerged in 2008, and is co-circulating along with PanAsia lineages since then. However, this report is the first report of the lineage PanAsia-02 in Bangladesh.

Available literature supported that, in South Asian countries, including Bangladesh, the circulating FMDV serotype A was placed under genotype VII (18) of Asia topotype [18,19]. Similarly, both the isolates of this study belonged to the genotype VII (18) with other Bangladeshi, Indian, and Bhutani isolates (Fig. 2). Similar results were described by Nandi et al. [9], Biswal et al. [18], Mohapatra et al. [19], and Subramanian et al. [20]. They have reported that the FMDV serotype Asia-1 placed under lineage C is circulating in India and other South Asian countries as we are reporting in this paper (Fig. 3). This indicates the sustainability of the serotype A and Asia-1 in Bangladesh, but serotype O lineage PanAsia-02 has newly emerged in Bangladesh.



Figure 1. Sequence similarity tree showing the relationship of FMDV serotype 0 of this study and previously reported isolates constructed using the neighbor-joining method with MEGA 6 software. A number of the nodes indicate bootstrap values calculated using 1,000 replicates. The red marking indicates the isolates of this study.



Figure 2. Sequence similarity tree showing the relationship of FMDV serotype A of this study and previously reported isolates constructed using the neighbor-joining method with MEGA 6 software. A number of the nodes indicate bootstrap values calculated using 1,000 replicates. The red marking indicates the isolates of this study.



Figure 3. Sequence similarity tree showing the relationship of FMDV serotype Asia-1 of this study and previously reported isolates constructed using the neighbor-joining method with MEGA 6 software. A number of the nodes indicate bootstrap values calculated using 1,000 replicates. The red marking indicates the isolates of this study.

FMDV serotype	GenBank accession number	Submission year	% of sequence identity
Serotype O	КТ960948	2013	100 (This study)
	КТ982203	2013	99.53 (This study)
	KT037118	2013	98.59
Serotype A	KY421678	2012	98.84
	KC795955	2012	92.96
	KC795954	2012	92.96
	KC795950	2012	93.49
	KJ754939	2012	91.69
	KC795949	2012	92.93
	KC795952	2012	93.16
	KC795951	2012	93.29
	КТ982204	2013	100 (This study)
	KR869773	2013	100
	KC795953	2013	92.99
	KY421679	2013	92.72
	КТ982205	2014	100 (This study)
	MK088171	2016	91.38
	MN968767	2018	100
Serotype Asia-1	MG603264	2009	94.79
	KY421683	2011	98.26
	KY421682	2011	98.42
	KY421687	2011	98.42
	KY421686	2012	98.10
	KY421685	2012	94.00
	KJ175173	2012	98.26
	KJ175172	2012	98.26
	KJ175170	2012	98.42
	KY421680	2012	94.00
	KR869774	2013	96.37
	KU159762	2013	100 (This study)
	KU159763	2013	99.53 (This study)
	KJ175186	2013	95.58
	MF782478	2013	98.10
	KY421684	2013	95.89
	KY421681	2013	95.89

Table 1. Percentage of identity of the VP1 gene among the sequences of this study and other studies of Bangladesh.

Conclusion

The phylogenetic analysis revealed that PanAsia-02 sub-lineage was confirmed in Bangladesh under the ME-SA topotype for the first time. On the other hand, we identified genotype VII (18) of Asia topotype (serotype A) and lineage C (serotype Asia-1). The extensive cross-border animal movement from neighboring countries may act as the source of diversified FMDV serotypes in Bangladesh.

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Conflict of interests

The author declares that they have no conflict of interests.

Authors' contributions

MLH conceptualized and designed the study and conducted the experiments. SA, MFRK, and KHMNHN took part in sequence analysis and bioinformatics analysis. SS, MAI, and MTR coordinated the research works. SMS took part in data analysis and checking the draft of the manuscript. MBR supervised the work. All the authors read the final version of this manuscript and approved it for publication.

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