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Genetic diversity of foot-and-mouth disease virus serotype O in Pakistan and Afghanistan, 1997–2009

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ABSTRACT

Foot-and-mouth disease (FMD) is endemic in Pakistan and Afghanistan; serotypes O, A and Asia-1 of the virus are responsible for the outbreaks in these countries with FMDV type O usually being the most common. In the present study, the nucleotide sequences encoding the FMDV capsid protein VP1 from virus samples were determined. Phylogenetic analysis of the serotype O FMD viruses circulating in Pakistan and Afghanistan between 1997 and 2009 revealed the presence of at least three different lineages within the ME-SA (Middle East South Asia) topotype. The three lineages detected in this study are Pak98, Iran2001 and PanAsia. The PanAsia lineage is currently dominant in the area and is evolving with time as revealed by the appearance of distinct variants e.g. PanAsia-II and a new variant designated here as PanAsia-III. The rates of evolution of the O-PanAsia-II and III sublineages prevalent in the region were found to be 6.65×10^{-3} (95% CI = 5.49–7.80 $\times 10^{-3}$) and 7.80×10^{-3} (95% CI = 6.72–8.89 $\times 10^{-3}$) substitutions per nucleotide per year, respectively. The present study reveals the presence of multiple (sub-)lineages of FMDV serotype O co-circulating in the region and that significant new variants are frequently emerging.

1. Introduction

Foot-and-mouth disease (FMD) is an infectious, contagious, viral disease of cloven-hoofed mammals (Alexandersen and Mowat, 2005). The disease is economically very important as it causes heavy losses to the livestock industry in terms of high morbidity in adult animals (especially cattle and pigs), reduced production efficiency and also mortality in young stock. Furthermore, the disease is a severe restraint to international trade of livestock and livestock products due to sanitary control measures. The causative agent, foot-and-mouth disease virus (FMDV), belongs to the genus *Aphthovirus* within the family *Picornaviridae* (Belsham, 2005). The virus is non-enveloped and has a positive-sense single stranded RNA genome, with a length of about 8.3 kb. The genome is enclosed within a nearly spherical protein capsid of about 28 nm, which consists of 60 copies of four different structural polypeptides, i.e. VP1, VP2, VP3 and VP4. The VP1 is the most variable of the capsid polypeptides and is considered to be highly immunogenic.

The virus exists in seven distinct serotypes, i.e. O, A, C, Asia-1, SAT 1, SAT 2 and SAT 3, that do not confer protection against each other after either infection or vaccination (Bachrach, 1968; Domingo et al., 2003). Within the serotypes, many subtypes can also be identified that sometimes do not induce total cross protection against other viruses of the same serotype. In addition, the control of this disease is constantly challenged by the emergence of new strains. The distribution of strains varies by location and this has been used to define virus topotypes. These describe the range of genetically distinct strains, within a serotype, present within a particular geographical area (Knowles and Samuel, 2003). In some areas, several serotypes and subtypes can occur at the same time. Moreover, the distribution of serotypes and subtypes around the world is not static.

FMDV is present in large areas of Africa, Asia and South America and can readily cross international boundaries and causes epidemics in previously free areas. FMD serotype O virus is predominant among the serotypes worldwide (Rweyemamu et al., 2008) and has been found responsible for important outbreaks in normally disease-free countries (Knowles et al., 2001; Brückner et al., 2002; Sakamoto et al., 2002; Shin et al., 2003; Tsutsui and Yamamoto, 2010).

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Table 1
Details of the samples from Pakistan and Afghanistan used in the present study.

Reference number	Species	Date of collection	Area	Country	Accession no.	Reference
O/PAK/1/1997	Cattle	24.3.1997	Karachi	Pakistan	AJ303526	Samuel and Knowles (2001)
O/PAK1/1998	Bovine	13.2.1998	NK	Pakistan	AJ318848	Knowles et al. (unpublished)
O/PAK/15/2002	Buffalo	15.3.2002	Sheikhopura	Pakistan	DQ165062	Knowles et al. (2005)
O/PAK/16/2002	Bovine	15.3.2002	NK	Pakistan	DQ165063	Knowles et al. (2005)
O/PAK/18/2002	Cattle	27.7.2002	Karachi	Pakistan	DQ115064	Knowles et al. (2005)
O/PAK/1/2003	Cow	6.1.2003	Karachi	Pakistan	DQ155065	Knowles et al. (2005)
O/PAK/12/2003	NK	2003	Lahore	Pakistan	DQ155066	Knowles et al. (2005)
O/PAK/14/2003	NK	2003	Lahore	Pakistan	DQ155067	Knowles et al. (2005)
O/PAK/16/2003	NK	2003	Lahore	Pakistan	DQ155068	Knowles et al. (2005)
O/PAK/17/2003	Vaccine strain			Pakistan	DQ155069	Knowles et al. (2005)
O/PAK/45/2003	Cow	19.3.2005	CBF, Harichand	Pakistan	DQ164942	Knowles et al. (2005)
O/PAK/53/2003	Buffalo	27.5.2003	Karachi	Pakistan	DQ164943	Knowles et al. (2005)
O/PAK/73/2003	NK	27.10.2003	Lahore	Pakistan	DQ165070	Knowles et al. (2005)
O/PAK/31/2005	Cattle	22.1.2005	Hafizabad	Pakistan	FJ798162	Waheed et al. (2010)
O/PAK/32/2005	Cattle	22.1.2005	Hafizabad	Pakistan	FJ798163	Waheed et al. (2010)
O/PAK/33/2005	Cattle	00.3.2005	Arifwala	Pakistan	FJ798164	Waheed et al. (2010)
O/PAK/34/2005	Cattle	00.3.2005	Arifwala	Pakistan	FJ798165	Waheed et al. (2010)
O/PAK/35/2005	Buffalo	28.2.2005	Lahore	Pakistan	FJ798166	Waheed et al. (2010)
O/PAK/38/2005	Cattle	12.2.2005	Karak	Pakistan	FJ798167	Waheed et al. (2010)
O/PUN/PAK/L133/2005	Blue bull	26.1.2005	Lahore	Pakistan	HQ439221	This study
O/PUN/PAK/L149/2005	Buffalo	4.2.2005	Kharian, Gujrat	Pakistan	HQ439222	This study
O/PUN/PAK/L150/2005	Buffalo	4.2.2005	Kharian, Gujrat	Pakistan	HQ439223	This study
O/PUN/PAK/L181/2005	Cattle	1.2.2005	Sargodha	Pakistan	HQ439224	This study
O/PUN/PAK/L282/2005	Cattle	30.3.2005	Layyah	Pakistan	HQ439225	This study
O/ISL/PAK/L285/2005	Buffalo	30.3.2005	Islamabad	Pakistan	HQ439226	This study
O/ISL/PAK/L286/2005	Buffalo	30.3.2005	Islamabad	Pakistan	HQ439227	This study
O/ISL/PAK/L287/2005	Buffalo	30.3.2005	Islamabad	Pakistan	HQ439228	This study
O/ISL/PAK/L288/2005	Cattle	30.3.2005	Islamabad	Pakistan	HQ439229	This study
O/PAK/2/2006	Cattle	31.1.2006	Karachi	Pakistan	EF494499	Klein et al. (2008)
O/PAK/4/2006	Cattle	31.1.2006	Karachi	Pakistan	EF494500	Klein et al. (2008)
O/PAK/6/2006	Buffalo	31.1.2006	Karachi	Pakistan	EF494501	Klein et al. (2008)
O/PAK/8/2006	Buffalo	31.1.2006	Karachi	Pakistan	EF494502	Klein et al. (2008)
O/PAK/10/2006	Buffalo	31.1.2006	Karachi	Pakistan	EF494503	Klein et al. (2008)
O/PAK/11/2006	Buffalo	1.2.2006	Karachi	Pakistan	EF494504	Klein et al. (2008)
O/PAK/12/2006	Buffalo	1.2.2006	Karachi	Pakistan	EF494505	Klein et al. (2008)
O/PAK/14/2006	Cattle	1.2.2006	Karachi	Pakistan	EF494506	Klein et al. (2008)
Pak 5.2.9	Buffalo	14.9.2006	Karachi	Pakistan	EF494498	Klein et al. (2008)
Pak 24.18	Buffalo	3.4.20007	Karachi	Pakistan	EF444993	Klein et al. (2008)
Pak 23.28	Buffalo	3.4.20007	Karachi	Pakistan	EF444991	Klein et al. (2008)
Pak 23.18	Buffalo	3.4.20007	Karachi	Pakistan	EF444990	Klein et al. (2008)
O/PAK/60/2006	Buffalo	4.1.2006	Sargpdha	Pakistan	FJ798168	Waheed et al. (2010)
O/PAK/61/2006	Cattle	4.1.2006	Sargpdha	Pakistan	FJ798169	Waheed et al. (2010)
O/PAK/63/2006	Buffalo	8.1.2006	Sheikhopura	Pakistan	FJ798170	Waheed et al. (2010)
O/PAK/66/2006	Cattle	15.2.2006	Jhang	Pakistan	FJ798171	Waheed et al. (2010)
O/PAK/67/2006	Buffalo	15.2.2006	Jhang	Pakistan	FJ798172	Waheed et al. (2010)
O/PAK/68/2006	Buffalo	1.2.2006	Hafizabad	Pakistan	FJ798173	Waheed et al. (2010)
O/PAK/70/2006	Cattle	18.2.2006	Arifwala	Pakistan	FJ798174	Waheed et al. (2010)
O/PAK/71/2006	Cattle	18.2.2006	Arifwala	Pakistan	FJ798175	Waheed et al. (2010)
O/PAK/72/2006	Buffalo	16.1.2006	Gujranwala	Pakistan	FJ798176	Waheed et al. (2010)
O/PAK/73/2006	Buffalo	16.1.2006	Gujranwala	Pakistan	FJ798177	Waheed et al. (2010)
O/PAK/74/2006	Cattle	30.1.2006	Okara	Pakistan	FJ798178	Waheed et al. (2010)
O/PAK/53/2007	Cattle	14.1.2007	Sargodha	Pakistan	FJ798179	Waheed et al. (2010)
O/PAK/56/2007	Buffalo	23.1.2007	Sheikhopura	Pakistan	FJ798180	Waheed et al. (2010)
O/PAK/60/2007	Cattle	18.1.2007	Jhang	Pakistan	FJ798181	Waheed et al. (2010)
O/PAK/61/2007	Cattle	7.2.2007	Hafizabad	Pakistan	FJ798182	Waheed et al. (2010)
O/PAK/63/2007	Cattle	22.2.2007	Gujranwala	Pakistan	FJ798183	Waheed et al. (2010)
O/PAK/66/2007A	Buffalo	7.12.2007	Layyah	Pakistan	FJ798184	Waheed et al. (2010)
O/PAK/66/2007B	Buffalo	7.12.2007	Layyah	Pakistan	FJ798185	Waheed et al. (2010)
O/PAK/68/2007	Buffalo	3.12.2007	TT Singh	Pakistan	FJ798186	Waheed et al. (2010)
O/PAK/69/2007	Buffalo	7.12.2007	Layyah	Pakistan	FJ798187	Waheed et al. (2010)
O/PAK/70/2007	Buffalo	7.12.2007	Layyah	Pakistan	FJ798188	Waheed et al. (2010)
O/PAK/71/2007	Cattle	8.12.2007	Lakki Marwat	Pakistan	FJ798189	Waheed et al. (2010)
O/PAK/1/2008	Cattle	3.1.2008	Burki	Pakistan	FJ798190	Waheed et al. (2010)
O/PAK/2/2008	Buffalo	8.1.2008	Patoki	Pakistan	FJ798191	Waheed et al. (2010)
O/PAK/3/2008	Cattle	8.1.2008	Patoki	Pakistan	FJ798192	Waheed et al. (2010)
O/PAK/6/2008	Cattle	24.1.2008	Peshswar	Pakistan	FJ798193	Waheed et al. (2010)
O/PAK/39/2008	NK	2008	NK	Pakistan	GU384685	Xu et al. (Unpublished)
O/PAK/44/2008	NK	2008	NK	Pakistan	GU384682	Xu et al. (Unpublished)
O/PAK/45/2008	NK	2008	NK	Pakistan	GU384683	Xu et al. (Unpublished)
O/PUN/PAK/L1343/2008	Cattle	31.12.2008	Faisalabad	Pakistan	HQ439207	This study
O/PUN/PAK/L1345/2008	Buffalo	31.12.2008	Faisalabad	Pakistan	HQ439213	This study
O/PUN/PAK/L1346/2008	Cattle	22.12.2008	Faisalabad	Pakistan	HQ439209	This study
O/PUN/PAK/L1347/2008	Cattle	22.12.2008	Faisalabad	Pakistan	HQ439206	This study
O/PUN/PAK/L1353/2008	Cattle	26.12.2008	Faisalabad	Pakistan	HQ439211	This study
O/PUN/PAK/L1358/2008	Cattle	22.12.2008	Faisalabad	Pakistan	HQ439208	This study
O/PUN/PAK/L1360/2008	Cattle	26.12.2008	Faisalabad	Pakistan	HQ439210	This study