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Identification of factors associated with increased excretion of foot-and-mouth disease virus

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ABSTRACT

We investigated which variables possibly influence the amount of foot-and-mouth disease virus (FMDV) shed in secretions and excretions by FMDV infected animals, as it is likely that the amount of FMDV shed is related to transmission risk. First, in a separate analysis of laboratory data, we showed that the total amount of FMDV in secretions and excretions from infected animals is highly correlated with maximum titres of FMDV. Next, we collected data from 32 published scientific articles in which FMDV infection experiments were described. The maximum titres of FMDV reported in different secretions and excretions (the response variable) and the experimental conditions in which they occurred (the explanatory variables), were recorded in a database and analyzed using multivariate regression models with and without random effects. In both types of models, maximum titres of FMDV were significantly ($p < 0.05$) associated with types of secretions and excretions, animal species, stage of the disease and days post infection. These results can be used to prioritize biosecurity measures in contingency plans.

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1. Introduction

Foot-and-mouth disease (FMD) is a contagious viral disease of cloven-hoofed animals, both domestic (cattle, pigs, sheep, goats and domestic buffalo) and wild (Thomson, 1994). The FMD virus (FMDV) can be transmitted by several routes (Sellers, 1971; Hyslop, 1970), with direct contact between animals considered the most important. The virus can also be transmitted by several indirect routes. In the European Union, an outbreak of FMD invokes an obligatory stand-still of animal transport (OIE, 2012). During such a stand-still, direct contact between infected animals in one farm and non-infected animals in another farm is theoretically impossible, leaving indirect transmission via contaminated material the most likely remaining route of transmission. In this respect, airborne transmission has been also considered (Henderson, 1969).

During epidemics, even when there is a complete standstill of animal transport, transmission between farms has been shown (Bouma et al., 2003). That indirect routes play a role in such transmission is clear from the observation that veterinarians were involved in the transmission of FMDV in outbreaks both in Denmark in 1982, and in Italy in 1993, either by using contaminated surgical equipment or by visiting farms after visiting an infected farm. Similarly, during the 2001 FMD outbreak in the United Kingdom, it was suggested that farmers were involved in the transmission of the virus between sheep flocks (Kitching, 2005). In the 2001 United Kingdom outbreak, the basic reproduction number remained above 1, that is, FMDV transmission continued despite the standstill in animal transport (Woolhouse et al., 2001). Thus indirect transmission of FMDV can have enormous consequences.

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It can be assumed that the risk of indirect transmission of FMDV is related to the total amount of FMDV present in the environment through contamination by secretions and excretions from FMDV infected animals. Here, secretions include material released from glands (e.g. milk, semen, saliva) whereas excretions refer to any other products released from animals (e.g. faeces, material released from the respiratory tract, urine, probang samples, nasal discharge and blood). The concentrations of FMDV in infected secretions and excretions have been reviewed (Pharo, 2002). However we analyzed the quantitative relationship between possible explanatory variables and the amount of FMDV in infected secretions and excretions.

2. Materials and methods

2.1. Materials

2.1.1. Laboratory data

Laboratory reports from animal studies performed at the Central Veterinary Institute (The Netherlands) were mined for all available daily data on virus secretion in milk from cattle (Orsel et al., 2007a) and on virus secretion and excretion in oropharyngeal fluid (OPF) swabs from cattle (Orsel et al., 2005), sheep (Eblé et al., 2008; Orsel et al., 2007b) and pigs (Eblé et al., 2004, 2006a,b, 2007; Orsel et al., 2007c). These data were used to identify the response variable for our multivariate regression analysis.

2.1.2. Literature data

Data on FMDV in secretions and excretions were collected from 32 scientific articles published between 1965 and 2007 (see Annex) found in internal databases and through the electronic (external) databases Scopus[1] and PubMed[2] in 2010, all reporting experimental trials involving FMDV infection. The electronic databases were explored using the keywords: foot-and-mouth disease, virus, infection and excretion. References cited in retrieved articles were reviewed to identify additional ones. The articles had to meet two basic criteria for their inclusion in the analysis: be written either in English, Spanish or French, and contain data on animal experiments with FMDV. They needed to contain information on the maximum titre(s) of FMDV detected in secretions and/or excretions, and additional information on one or more of the following: the type(s) of secretion or excretion in which the virus was detected, route of infection, animal species, FMDV serotype, stage of disease (clinical and non-clinical), dose of infection and/or days post infection at which the maximum secretion or excretion occurred. Missing data on one or more of these variables were recorded as not available (N.A.). These data were used as the response and possible explanatory variables for our multivariate regression analysis.

Per individual animal, the maximum titre of FMDV (including the experimental conditions) was recorded. Virus titres were reported as ^{10}log $TCID_{50}$/ml. Plaque forming units (PFU) were converted to $TCID_{50}$ (Horzinek, 1985). Median doses, such as 50% cattle infection dose (CID_{50}), 50% mouse infection dose (MID_{50}) or 50% mouse lethal dose (MID_{50}) per ml, were considered equal to 50% tissue culture infective dose ($TCID_{50}$/ml) (House and Yedloutschnig, 1982). The maximum recorded titre was the maximum titre over time for an individual animal. If the maximum titre was reported per group of animals, this resulted in one observation (from blood in Alexandersen et al. (2003); from airborne excretion in Alexandersen et al. (2002), Alexandersen and Donaldson (2002), Donaldson et al. (1970, 1981, 1982), Gloster et al. (2007), and Sellers and Parker (1969); from probang, milk, faeces and blood in Burrows (1968); from milk in Burrows (1971); and, from probang and nasal discharge in Burrows (1972)). Data on airborne excretion were recorded as ^{10}log $TCID_{50}$/animal/day.

The recorded secretion or excretion types were airborne, faeces, milk, probang, semen, urine, blood, nasal discharge, oropharyngeal fluid (OPF) swabs, and saliva. The category faeces contains data on material collected from the rectum (Burrows et al., 1968) and from rectal swabs (Garland, 1974). Probang refers to oropharyngeal samples that were obtained after scraping the oropharynx with a sampling cup.

Routes of infection were recorded as: contact (if an infected donor and a susceptible contact animal shared a common experimental unit); intranasal (IN, if the animals were infected via the intranasal route) or parenteral (if the animals were infected intravenously (IV), intramuscularly (IM), intralingually, intracutaneously (IC), intramammary or intradermally (ID)).

Animal species were recorded as cattle (bull, steer, ox, cow, calf and heifer), swine (pigs) or small ruminants (sheep, lambs and goats). The FMD viruses used for infection were recorded based on FMDV serotype, i.e. A, O, C, Asia 1, SAT 1, SAT 2, SAT 3, but no subdivision was made to the level of subtypes. The stage of disease was recorded as 'clinical' when lesions or clinical signs (including fever) were reported; otherwise it was recorded as 'non-clinical'.

Dose of infection (ranging from 0.95 to 10.15 $TCID_{50}$/ml) was recorded. Days post infection was recorded as the day when the maximum titres in the secretion or excretion were observed (ranging from 0.33 to 28 dpi).

2.2. Methods

2.2.1. Identifying the response variable for the multivariate regression analysis

A proxy for the total amount of FMDV secreted and excreted by the infected animals was established using available laboratory data from OPF swab samples and milk samples. The total amount of secreted and excreted FMDV (per individual animal) was calculated by summing the observed viral amounts (without logarithmic transformation) from consecutive observations (area under the curve, AUC). In a univariate regression analysis, the logarithm of the AUC (^{10}log AUC) was used as the response variable. Three explanatory variables were analyzed: (1) the maximum virus titre (max ^{10}log $TCID_{50}$/ml), (2) the time when the maximum virus titre occurred (^{10}log

[1] http://www.scopus.com/.
[2] http://www.ncbi.nlm.nih.gov/pubmed/.

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