# IMMUNITY STATUS OF FOOT-AND-MOUTH DISEASE IN THE BORDER DISTRICTS OF PENINSULAR MALAYSIA

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#### Abstract

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A serological survey for the prevalence of protective level of antibody to Foot-and-mouth disease (FMD) was carried out in 10 border districts in Peninsular Malaysia. A liquid phase blocking ELISA kit prepared and standardized by World Reference Laboratory (WRL) for FMD was used for the testing. A total of 800 serum samples collected by a random process were tested for protective level of antibody for virus types O, A and Asia I. An overall mean prevalence for antibody to FMD in the 'immune-belt' region was found to be 51.0%, 37.3%, 53.6% for virus types Q, A, and Asia I respectively and 28.9% for all the three sero-types. The percentage of cattle population having protective level of antibody was too low to prevent active spread of FMD infection. There was also substantial variation in the prevalence of antibody detected at the district level and varied from a low mean of 18.8% for the State of Kedah and a high of 67.5% for the district of Besut. More than 70% of the population need to have protective level of antibody to effectively prevent disease spread. The States of Kedah and Kelantan had variable levels of vaccination coverage from 1994 and had less than 45% coverage for the year 1996. A coverage of more than 90% would be essential to maintain high herd immunity and the current high variability in the vaccination coverage at the district level will only favour a higher infection on rate in the field.

#### 1. INTRODUCTION

Peninsular Malaysia has faced many outbreaks of foot-and-mouth disease (FMD) since 1973 [1]. These outbreaks have been effectively controlled through vaccination and a modified stamping out method. However, the scenario has changed since 1992 whereby sporadic outbreaks continue to emerge in the border districts. Many reasons have been attributed to this problem including limited success in movement control at the border region and low herd immunity in the 'buffer zone'. Low vaccination coverage has been incriminated as the main reason for this low herd immunity. Mass vaccination is used as a key strategy of control in the Border States of Kelantan, Terengganu, Perlis, Kedah and Perak. Annual vaccination of cattle and buffaloes using a trivalent vaccine is mandatory in these States. However, the vaccination coverage varied widely in these States with the exception of the State of Terengganu. Higher coverage of up to 80% is usually achieved subsequent to an outbreak. Though vaccination has been deployed as a key strategy of control in the 'immune-belt region' the assessment of immunity status in animals in these areas has not been possible (no antibody monitoring capability) and has not been carried out so far. An epidemiological investigation to evaluate the immunity status in this region would be vital for an effective control and eradication programme. The immunity status of the animals in these strategic border districts would reveal valuable information as to the cause of the persistent infection in this region. This could also outline the future strategy of control and eradication.

The objective of this study is to:

- Assess the immunity status to FMD in the high-risk border districts of Peninsular Malaysia.
- Determine correlation between vaccination coverage and antibody prevalence.

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## 2. MATERIALS AND METHODS

# 2.1. Sampling Procedures

Serum samples were collected from cattle in the border and FMD high-risk districts in the States of Kelantan (4), Terengganu (1), Perlis (1), and Kedah (4). Ten districts were picked for this survey based on a high risk of having FMD and being in a strategic location. About 80 sera per district were determined using computer software EPINFO, based on a population size of about 20 000 at 95% confidence interval and 10% accuracy and using an estimated antibody prevalence of 30%. Longitude/latitude gridlines were drawn on the district maps at an interval of one minute (2 km in distance) and each square was numbered. Eight squares from each district were selected at random lot and the exact position of these squares was determined by using a Global Positioning System (GPS) GARMIN GPS II. Serum samples were collected from cattle within one kilometer radius from the position located by the GPS system. The serum samples were tested using ELISA kits provided by the Joint FAO/IAEA Division, which have been prepared and standardized by FMD World Reference Laboratory (WRL) at Pirbright, United Kingdom. The antibody detection kit is a liquid phase blocking ELISA technique for the detection of FMD virus antibodies in serum as described in the literature [2,3]. The assay was used as a single dilution screening assay and as a quantitative titration assay, resulting in an end-point titre determination for each serum. The kit detected antibody for FMD virus types O, A and Asia I. The assay protocols followed are as described by FAO/IAEA Bench Protocol June 1995 and June 1997.

### 2.2. Assay interpretation

The diagnostic threshold for the screening assay was set at 50% percentage inhibition (50 PI) at serum dilution of 1:32. Serum samples positive to the test (PI  $\ge$  50) were further tested by titration assay to assess 'protective level' of antibody. The serum samples were tested at two fold dilutions of 1:64 and 1:128 to determine protective level of antibody. An antibody titre of  $\ge$  90 is considered protective, which indicates that the animal, at the time of bleeding, was protected against infection from homologous antigen of the particular FMD virus sero-type. Antibody prevalence for all three sero-types was calculated based on the presence of protective level of antibody in all three sero-types.

A spreadsheet was used to tabulate the results and plot the charts. The vaccination coverage from August 1996 to July 1997 of each sub-district where samples were collected was analysed for correlation with immunity status. Correlation analysis was done using a statistical program SPSS.

# 3. RESULTS AND DISCUSSION

Table I shows the prevalence of cattle with protective level of antibody for FMD. The State of Kelantan had a mean antibody prevalence of 49.7%, 32.8%, 59.7% for virus types O, A and Asia I respectively and 25.9% for all three sero-types. Kedah had a mean antibody level of 44.1%, 30.3%, 40.9% for virus types O, A, Asia I and 18.8% for all sero-types. The State of Perlis had 63.8%, 47.5%, 56.3% for virus types O, A, Asia I respectively and 42.5% for all sero-types. The district of Besut had a level of 71.3%, 72.5%, 77.5% for virus types O, A, Asia I, respectively and 67.5% for all sero-types. The overall mean prevalence of protective level of antibody for the districts in the immune belt region was found to be 51.0%, 37.3%, 53.6% for virus types O, A, Asia I, respectively and 28.9% for all sero-types.

The antibody prevalence for virus types O, A and Asia I varied widely in this survey. The trivalent vaccine used in the national FMD control programme had been purchased from a reputable supplier and induced good immune response to all three sero-types (Palanisamy – unpublished data). Antibody monitoring programme of imported cattle and buffalo at quarantine stations also conforms to this finding (Palanisamy – unpublished data). Immunity due to natural infection could be the main cause for the wide variation between the sero-types. The incidence of FMD outbreak due to virus type A had been rare, restricted to Selama Perak and Kedah in 1995 and 1996. As such, percentage of animals detected with antibody for virus type A and antibody for all three sero-types together is more likely to be of vaccine origin rather than from natural infection in the field. Generally, higher antibody prevalence has been observed for sero-types Asia I and O compared to type A. The districts of Pasir

Puteh, and Bachok, in the State of Kelantan had much higher prevalence for Asia I due to widespread infection with this virus type in 1995 and 1996. The districts of Kubang pasu, Kota Setar and Padang Terap however, had much higher prevalence of antibody for virus type O. This could be due to the protracted outbreak with virus type O since 1995 and 1996.

	Sample	Туре О	Туре А	Type Asia I	All Sero-type
	Size	%	%	%	%
KELANTAN					
TUMPAT	80	56.3%	28.8%	50.0%	22.5%
PASIR MAS	80	37.5%	31.3%	47.5%	18.8%
ВАСНОК	80	46.3%	36.3%	67.5%	33.8%
PASIR PUTEH	80	58.8%	35.0%	73.8%	28.8%
Mean for State		49.7%	32.8%	59.7%	25.9%
<u>TERENGGANU</u>					
BESUT	80	71.3%	72.5%	77.5%	67.5%
PERLIS					
PERLIS	80	63.8%	47.5%	56.3%	42.5%
<u>KEDAH</u>					
KUBANG PASU	80	53.8%	30.0%	45.0%	13.8%
KOTA SETAR	80	31.3%	11.3%	20.0%	10.0%
PADANG TERAP	80	65.0%	43.8%	52.5%	35.0%
PENDANG	80	26.3%	36.3%	46.3%	16.3%
Mean for State		44.1%	30.3%	40.9%	18.8%
Overall Mean		51.0%	37.3%	53.6%	28.9%

TABLE I. CATTLE WITH PROTECTIVE LEVEL OF ANTIBODY TO FMD IN THE BORDER DISTRICTS OF PENINSULAR MALAYSIA – AUGUST 1997

There is substantial variation in the prevalence of antibody to FMD in the Districts surveyed and varied from a low mean of 18.8% for the State of Kedah and a high of 67.5% for Besut, Terengganu. The overall mean for the districts in the 'buffer zone' was found to be only 28.9%. Too low a level to prevent an active spread of the disease in the cattle population as substantial numbers of susceptible animals are maintained for infection to persist. Gleeson et al, (1993) [4] have Stated that in order to prevent an epidemic 70% of the population need to be protected, but to absolutely prevent an outbreak on a herd basis 95% protection is required [5].

Poor vaccination coverage is the main factor for the low protective level of antibody in the border districts. This is evident from Table II. The district of Besut in Terengganu has had maintained a high coverage of more than 90% for the last 3 years, hence a good protective level of antibody of >70% for the three homologous sero-types. However, there is substantial variation in the coverage for the districts in the States of Kelantan and Kedah. Coverage of less than 45% for 1996 for the above States warrants a serious look at the problem. Work in Thailand indicates that vaccinating 70% of the village cattle and buffaloes twice a year is unlikely to produce a level of herd immunity sufficient to

prevent spread of FMD virus. Reasons for this low level of immunity includes poor response to initial vaccination, decline in titres between vaccination and increase in susceptible population through birth and this requires coverage of nearly 100% to be effective [6]. Vaccination coverage of >80% is crucial for an effective control of the disease.



FIG. 1. Cattle with protective level of antibody to different virus types of FMD in border districts of peninsular Malaysia – August 1997

The reported vaccination figures (Table II) do not truly reflect the immune status of the population, as the percentage of animals truly protected would be much lower. The protection is dependent on the potency of the vaccine (the correct strain and optimum antigenic content), maintenance of 'cold-chain' and vaccine delivery to the animal. The current vaccination coverage is too low to prevent any active spread of the disease in the Border States.

	Years						
State	1992	1993	1994	1995	1996		
PERLIS	94.3	71.9	86.2	30.5	92.0		
KEDAH							
K.Setar	-	_	61.5	2.1	52.6		
Pendang		-	71.6	17.6	23.4		
K.Pasu	-	-	84.5	18.7	27.9		
P.Terap	_	-	-	14.0	32.2		
Mean		-	72.5	13.1	34.0		
TERENGGANU							
Besut	72.9	86.9	94.7	90.3	97.6		
KELANTAN							
Tumpat	24.8		80.1	79.2	52.1		
P.Mas	40.5	_	47.0	-	37.5		
K.Bharu	33.5	-	33.8	68.7	26.1		
Bachok	27.9	-	24.4	79.5	50.4		
P.Puteh	40.5	-	75.7	27.4	41.1		
Mean	33.4	-	52.2	51.0	41.4		

TABLE II. FMD VACCINATION COVERAGE (%) IN STRATEGIC DISTRICTS OF PENINSULAR MALAYSIA 1992–1996

There was poor correlation between vaccination coverage at sub-district and antibody prevalence for the three sero-types which was found to be 0.1216, 0.1553 and 0.1396 for sero-types O, A and Asia I respectively. This could have been influenced by faulty animal census figures at sub-District level. This is reflected by the poor correlation coefficient and many outliers with way-off values of more than 100% for the coverage. However, a good correlation of 0.7813 and 0.6101 was found for vaccination coverage (Fig. 2) at district level with that of antibody prevalence for all three sero-types and type A respectively. This supports the view of high vaccination coverage resulting in high prevalence for protective level of antibody in the cattle population.



FIG. 2. Correlation plot of vaccination coverage (%) and antibody level

FMD detection in about 8 consignments of animals at the checkpoints and quarantine stations in 1996 reflects the constant threat of disease introduction into the country. Numerous illegal movement routes and increased smuggling activity during festive seasons have contributed to many outbreaks. As such, FMD control and eradication strategy have to focus on mass vaccination to maintain a high herd immunity of more than 80% to prevent future outbreaks. This could be achieved by maintaining a vaccination coverage of >90% in all the districts with minimal variability in the coverage at the village level.

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