

**REPORT ON THE PRODUCTION OF EMERGENCY 01 MANISA FOOT-  
AND-MOUTH DISEASE VACCINE FOR THE UNITED KINGDOM BY THE  
INTERNATIONAL VACCINE BANK (IVB), 22<sup>nd</sup> MARCH – 4<sup>th</sup> APRIL 2001**

**Paul V Barnett**

**Institute for Animal Health, Pirbright Laboratory,  
Ash Road, Pirbright, Surrey GU24 0NF, U.K.**

### **Summary**

On the 20th February 2001 the United Kingdom confirmed its first case of foot-and-mouth disease (FMD) for 20 years and during the initial stages, the International Vaccine Bank (IVB), Pirbright, prepared itself for the possibility of producing emergency FMD vaccine. Serological evaluation and sequencing data undertaken at Pirbright had indicated that the most suitable vaccine strain against this particular field isolate would be O<sub>1</sub> Manisa. On the 22<sup>nd</sup> March, the Department of Environment, Food and Rural Affairs (DEFRA), formerly the Ministry of Agriculture, Fisheries and Food (MAFF) requested the formulation of 500,000 bovine doses of aqueous aluminium hydroxide/saponin O<sub>1</sub> Manisa vaccine. This was the first time in the Banks' history that it had been summoned to produce vaccine in 'anger'. This report details the subsequent manufacture of vaccine following request.

### **Manufacture of emergency FMD vaccine**

The 500,000 bovine doses of O<sub>1</sub> Manisa vaccine were formulated over four separate batch runs. The first three runs consisting of 150,000 cattle doses, and a final run of 50,000 bovine doses. Because of the volumes required, the 500 litre vessel OV1 vessel was used for the blending of each of the 150,000 dose batches (equivalent to 450 litres), whilst the final 50,000 dose run (equivalent to 150 litres) was blended in the 300 litre vessel LH1. Each individual run took three days to complete from preparation and sterilisation to filling and capping. In accordance with Good Manufacturing Practice (GMP), records were made of each manufacturing stage, which were countersigned by the appropriate member of staff and any problems entailed were also noted.

Some 26 staff members were either directly or indirectly involved in assisting in the production of the vaccine. This included 6 bottling personnel, 5 personnel for dispatch (1 transporting vaccine to hatch, 2 adding documents, freezer packs and checking, 2 strapping boxes and 3 personnel packing). Bottling rate of the vaccine was approximately 280 units per hour and a filling run into nominal 300 ml polypropylene bottles of 150,000 bovine doses took approximately 6 - 6.5 hours. Some 494,657 bovine doses were finally dispensed, which were hand labelled, packed appropriately in 20 unit amounts and stored in the IVB's + 4°C cold room. The only dispatched vaccine, Batch 1/01, which was transported to Penrith in Cumbria, also included cool packs (1 per box), an aqueous vaccine package insert/data sheet and a disclosure sheet notifying the user of the number of doses per vaccine bottle. These sheets were similarly produced for the other 3 batches awaiting dispatch. The number of doses per bottle tended to vary slightly from run to run.

The minimum of at least 30 retention samples were kept from each batch for subsequent analyses or sterility testing. In addition, during the transfer of components and blending of each batch, in-line samples were also taken for sterility checks. The vaccine, Batch 1/01, which was dispatched to Cumbria was subsequently returned to Pirbright and all four batches of vaccine are still currently stored in the IVB's +4°C fridge.

## Quality control

Safety, according to current European Pharmacopoeia guidelines, and potency of the emergency vaccine by serology, were undertaken in-house. Sterility of the final product was carried out independently by a third party to full European Pharmacopoeia compliance. Two production batches were used for the various tests. Batch 1/01, which was dispatched to Penrith in Cumbria, underwent sterility and safety testing. Batch 2/01 was used for potency analysis in 8 cattle. In addition, Batch 4/01 has been used to monitor the stability of the final product at +4°C.

## Results

**a) Safety test on emergency 0<sub>1</sub> Manisa vaccine Batch 1/01-** Carried out in accordance to the European Pharmacopoeia safety test for veterinary vaccines. Briefly, two cattle were inoculated with 2 x bovine dose (6 ml) of aqueous AI (OH)<sub>3</sub>/saponin vaccine, Batch 1/01, subcutaneously. Body temperatures were recorded and the animals were monitored daily for well being and local reactions.

**Table 1 Results of safety test on emergency 0<sub>1</sub> Manisa vaccine Batch 1/01**

Animal	5/4/01	6/4/01	7/4/01	8/4/01	9/4/01	10/4/01	11/4/01	12/4/01	13/4/01	4/4/01
UJ70	38.6°C	39.0°C	39.1°C	38.8°C	38.9°C	38.5°C	38.4°C	38.5°C	38.7°C	38.6°C
UJ71	38.2°C	39.0°C	38.2°C	38.5°C	38.7°C	38.4°C	38.2°C	38.2°C	38.4°C	38.4°C

No adverse reactions were observed following vaccination and animals remained healthy and body temperatures remained normal during period of monitoring.

**b) Cattle potency test of emergency 0<sub>1</sub> Manisa vaccine Batch 2/01** - Using the same vaccine formulation, this antigen was originally potency tested for acceptance into the IVB in 1991 and was found to have a PD<sub>50</sub> value 112. Batch 2/01 was therefore tested in accordance to a mini IVB cattle potency test which is routinely undertaken every fifth anniversary following acceptance. Briefly, eight cattle were subcutaneously vaccinated with a 1/10 cattle dose of antigen as a 3 ml aqueous AI (OH)<sub>3</sub>/saponin vaccine. At 21 days post-vaccination animals were bled for serology and a PD<sub>50</sub> value estimated from the neutralising antibody titres at 21 days by computer model analysis using logistic regression.

**Table 2 Cattle potency test on emergency 0<sub>1</sub> Manisa vaccine Batch 2/01**

Animal Number	Antibody titres (Log SN50 @100 TCID50) at 21 days p.v.	Probit - % Probability of Protection
UJ91	1.505	40.7%
UJ92	1.95*	>90%
UJ93	1.95	>90%
UJ94	1.95	>90%
UJ95	1.806	79.7%
UJ96	1.95	>90%
UJ97	1.95	>90%
UJ98	1.95	>90%

**Expected protection for 0<sub>1</sub> Manisa Batch 2/01 = 6.606/8 (>10 PD<sub>50</sub>) Variance = 0.942 t - statistic - 2.6849606**

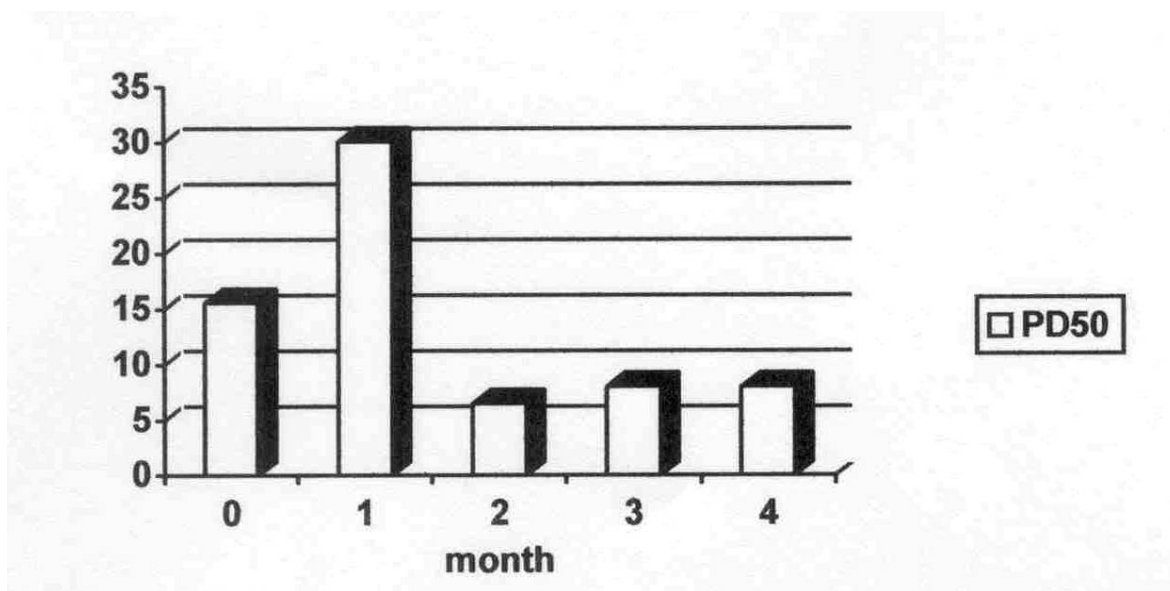
chance that PD<sub>50</sub> > than dilution used (10) = 98.61%

\* - all the 1.95 titres were >1.95 and therefore the calculation is lower than the probable value, however, the potency was in excess of its requirement.

c) **Sterility test on emergency 0<sub>1</sub> Manisa vaccine Batch 1/01** - The testing regime which was done in accordance to the current European Pharmacopoeia and undertaken independently found Batch 1/01 to be sterile. In addition, all line samples taken during the different stages of manufacture showed no evidence of contamination.

d) **Guinea pig potency/stability test on emergency 0<sub>1</sub> Manisa vaccine Batch 4/01** - The testing regime followed that previously described (1) except that the animals receiving a specific dilution of vaccine were always in groups of five and the vaccines were only diluted threefold up to 1/27. Testing was repeated monthly over a 4 month period. PD<sub>50</sub> values were calculated by the method of Karber (2).

**Figure 1 Guinea pig potency/stability values of Batch 4/01 stored over 4 months at +4°C**



### Discussion

The emergency foot-and-mouth disease vaccine requested on the 22<sup>nd</sup> March 2001 by the Department of Environment, Food and Rural Affairs (DEFRA), formerly the Ministry of Agriculture, Fisheries and Food (MAFF), and produced by the IVB at Pirbright, was shown to be safe, sterile and of the required potency with a PD<sub>50</sub> value in excess of 10. A dossier of all the relevant quality control testing of 0<sub>1</sub> Manisa antigen was compiled for scrutiny by the Veterinary Medicines Directorate.

## References

1. P. V. Barnett and R. J. Statham Long Term Stability and Potency of Antigen Concentrates Held by the International Vaccine Bank. 1998, *Report, Session of the Research Group of the Standing Technical Committee of the European Commission for the Control of Foot-and-Mouth Disease and the Foot-and-Mouth Disease Subgroup of the Scientific Veterinary Committee of the Commission of the European Community, United Kingdom (1998), Appendix 38, pages 272-275.*
2. Karber, G. *Beitrag zur kollektiven Behandlung pharmakologischer Reihenversuche.* Arch. Exp. Pathol. Pharmacol. 1931, **162**, 480.