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Foot and Mouth Disease (Ruminants) Vaccine

General Notices

(Foot-and-Mouth Disease (Ruminants) Vaccine (Inactivated), Ph. Eur. monograph 0063)

Ph Eur

1 DEFINITION

Foot-and-mouth disease (ruminants) vaccine (inactivated) is a preparation containing one or more suitable strains of foot-and-mouth disease virus inactivated while maintaining adequate immunogenic properties. This monograph applies to vaccines intended for active immunisation of ruminants against foot-and-mouth disease.

2 PRODUCTION

2-1 PREPARATION OF THE VACCINE

The vaccine virus is grown in cell cultures and then separated from cellular material by filtration or other suitable procedures. The harvested virus is inactivated in suitable conditions and may be concentrated and purified. It is used for the preparation of vaccine immediately or after storage at a temperature shown to be consistent with antigen stability. The vaccine is prepared from inactivated virus by blending with one or more adjuvants. For a given strain, the quantity of 146S antigen blended in each batch of vaccine is not lower than that of a batch of vaccine that has been found satisfactory with respect to Immunogenicity.

2-2 SUBSTRATE FOR VIRUS PROPAGATION

2-2-1 Cell cultures

The cell cultures comply with the requirements for cell cultures for production of veterinary vaccines (*5.2.4*).

2-3 VALIDATION OF THE INACTIVATION PROCEDURE

During inactivation, the virus titre is monitored by a sensitive and reproducible technique. The inactivation procedure is not satisfactory unless the decrease in virus titre, plotted logarithmically, is linear and extrapolation indicates that there is less than 1 infectious virus unit per 10^4 L of liquid preparation at the end of inactivation.

2-4 CHOICE OF VACCINE COMPOSITION

The vaccine is shown to be satisfactory with respect to safety (*5.2.6*) and efficacy (*5.2.7*) for each species for which it is intended.

The following tests for safety (section 2-4-1) and immunogenicity (section 2-4-2) may be used during the demonstration of safety and efficacy.

2-4-1 Safety

Carry out the test for each route and method of administration to be recommended for vaccination and in each category of each species for which the vaccine is intended, using in each case animals of the minimum age to be recommended. Use a representative batch of vaccine containing not less than the maximum antigen content that may be expected in a batch of vaccine.

For each test, use not fewer than 8 animals that do not have antibodies against foot-and-mouth disease virus. Administer to each animal 1 dose of the vaccine. If the schedule to be recommended requires a 2^{nd} dose, administer 1 dose after an interval of at least 14 days. Observe the animals at least daily for at least 14 days after the last administration.

The vaccine complies with the test if no animal shows abnormal local or systemic reactions, or dies from causes attributable to the vaccine.

2-4-2 Immunogenicity

Carry out an immunogenicity test for each strain of foot-and-mouth disease virus that may be included in the vaccine.

Each test is carried out for each route and method of administration to be recommended for vaccination, using in each case cattle not less than 6 months old. The vaccine administered to each cattle is of minimum antigen content.

Either of the following 2 tests is suitable to demonstrate immunogenicity of the vaccine for cattle.

2-4-2-1 PD_{50} challenge test. The potency of the vaccine is expressed as the number of 50 per cent cattle protective doses (PD_{50}) contained in the dose stated on the label. The PD_{50} is determined in cattle given primary vaccination and challenged by the inoculation of 10 000 ID_{50} of virulent bovine virus of the same strain as that used in the preparation of the vaccine in the conditions described below. The vaccine virus may be used for challenge.

Use for the test not fewer than 17 cattle obtained from areas free from foot-and-mouth disease, that have never been vaccinated against foot-and-mouth disease and do not have antibodies neutralising the different strains of foot-and-mouth disease virus. Vaccinate not fewer than 3 groups of not fewer than 5 cattle per group, using a different dose of the vaccine for each group. Administer the different doses by injecting different volumes of the vaccine and not by dilution of the vaccine. Maintain 2 cattle as controls. For example, if the label states that the injection of 2 mL corresponds to the administration of 1 dose of vaccine, a 1/4 dose of vaccine would be obtained by injecting 0.5 mL, and a 1/10 dose would be obtained by injecting 0.2 mL. Challenge all the cattle after 20-22 days by the intradermal route, into at least 2 sites on the upper surface of the tongue (0.1 mL per site), with a dose equivalent to approximately 10 000 ID_{50} of a suspension of a fully virulent virus, obtained from cattle and of the same strain as that used in the preparation of the vaccine. Observe the cattle at least daily for 8 days. In the interest of animal welfare, individual animals may be euthanised before the end of the observation period and considered as unprotected if a vaccinated animal shows lesions of foot-and-mouth disease on at least 1 foot or if a control animal shows lesions of foot-and-mouth disease on at least 3 feet. Unprotected cattle show lesions at sites other than the tongue. Protected cattle may display lingual lesions.

The test is not valid if both control cattle do not show lesions on at least 3 feet. From the number of protected cattle in each group, calculate the PD_{50} content of the vaccine.

The vaccine complies with the test if the potency is not less than that to be stated on the label; the minimum potency to be stated on the label is not less than 3 PD_{50} per dose for cattle.

2-4-2-2 PPG test. The following test could also be used to demonstrate immunogenicity of the vaccine for cattle (referred to as the ‘Percentage of protection against generalised foot infection' (PPG test)).

The potency of the vaccine is expressed as the percentage of cattle that do not show lesions on any feet. The PPG is determined in cattle given primary vaccination and challenged by the inoculation of 10 000 ID_{50} of virulent virus of the same strain as that used in the preparation of the vaccine under the conditions described below. The vaccine virus may be used for challenge. Use for the test not fewer than 18 cattle obtained from areas free from foot-and-mouth disease, that have never been vaccinated against foot-and-mouth disease and do not have antibodies neutralising the different strains of foot-and-mouth disease virus. Vaccinate not fewer than 16 cattle with 1 full dose. Maintain 2 cattle as controls. Challenge all the cattle after 20-22 days by the intradermal route, into at least 2 sites on the upper surface of the tongue (0.1 mL per site), with a dose equivalent to approximately 10 000 ID_{50} of a suspension of a fully virulent virus of the same strain as that used in the preparation of the vaccine. Observe the cattle at least daily for 8 days. In the interest of animal welfare, individual animals may be euthanised before the end of the observation period and considered as unprotected if a

vaccinated animal shows lesions of foot-and-mouth disease on at least 1 foot or if a control animal shows lesions of foot-and-mouth disease on at least 3 feet. Unprotected cattle show lesions at sites other than the tongue. Protected cattle may display lingual lesions.

The test is not valid if both control cattle do not show lesions on at least 3 feet. From the number of protected cattle in the vaccinated group, calculate the percentage of protected cattle.

The vaccine complies with the test if the potency is not less than that to be stated on the label; the minimum potency to be stated on the label is not less than 75 per cent.

2-5 MANUFACTURER'S TESTS

2-5-1 Identification

The bulk inactivated antigen is identified by a suitable immunochemical method (*2.7.1*).

2-5-2 Residual live virus

The limit of detection of the cell cultures to be used with respect to the virus to be tested is established by determining the number of $CCID_{50}$ and the 146S antigen content of a sample of live virus. The cells are not suitable if an amount of virus corresponding to 1 µg of 146S antigen has less than 10^6 $CCID_{50}$. A proportion of each batch of bulk inactivated antigen representing at least 200 doses is tested for freedom from live virus by inoculation into suitable cell cultures. A passage is made during culture of the cells. For this purpose, the sample of the inactivated antigen may be concentrated to allow testing of such large samples in cell cultures. It must be shown that the selected concentration and assay systems are not detrimental to detection of infectious virus within the test sample and that the concentrated inactivated antigen does not interfere with virus replication or cause toxic changes. A positive control is included in each test.

2-5-3 Antigen content

The 146S antigen content of each batch of bulk inactivated antigen is determined by an *in vitro* method (for example, by sucrose density gradient centrifugation and ultraviolet spectrophotometry at 259 nm).

2-5-4 Batch potency test

It is not necessary to carry out the potency test (section 3-3) for each batch of vaccine if it has been carried out using a batch of vaccine with a minimum antigen content. Where the test is not carried out, an alternative validated method is used, the criteria for acceptance being set with reference to a batch of vaccine that has given satisfactory results in the test described under Potency and has been shown to be satisfactory with respect to immunogenicity in the target species.

The following test may be used after a satisfactory pass level for a given strain has been established. Once a pass level has been established for a given strain, the same level of antigen may be used when this strain is formulated in combination with any other antigen provided that the formulation of the vaccine differs only in the strains included.

2-5-4-1 Vaccines for use in cattle. Use cattle of the minimum age recommended for vaccination obtained from areas free from foot-and-mouth disease, that have never been vaccinated against foot-and-mouth disease and do not have antibodies neutralising the different strains of foot-and-mouth disease virus. Vaccinate not fewer than 5 cattle by a recommended route. Use a suitable dose of the vaccine for each animal. After a defined period, not greater than 28 days following vaccination, draw a blood sample and determine individually in each serum the level of antibodies against each strain used in the preparation of the vaccine by a validated technique (e.g. sero-neutralisation test, ELISA). The vaccine complies with the test if the geometric mean of the antibody titre in cattle is not significantly lower than the pass level.

2-5-4-2 Vaccines for use in other ruminants. The potency of each batch shall be demonstrated in a suitable, validated test.

EMERGENCY USE: in situations of extreme urgency and subject to agreement by the competent authority, a batch of vaccine may be released before completion of the tests and the determination of potency if a test for sterility has been carried out on the bulk inactivated antigen and all other components of the vaccine and if the determination of potency has been carried out on a representative batch of vaccine prepared from the same bulk inactivated antigen. In this context, a batch is not considered to be representative unless it has been prepared with not more than the amount of antigen or antigens and with the same formulation as the batch to be released.

3 BATCH TESTS

3-1 Identification

The vaccine contains the antigen or antigens stated under Definition.

3-2 Bacteria and fungi

The vaccine and, where applicable, the liquid supplied with it, comply with the test for sterility prescribed in the monograph *Vaccines for veterinary use (0062)*.

3-3 Potency

The vaccine complies with the requirements of the test mentioned under Immunogenicity (section 2-4-2) when administered by a recommended route and method.
