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.

IDENTIFICATION
The serum of a susceptible animal that has been immunised with the vaccine neutralises the serotypes of the virus used to prepare the vaccine, when tested by a suitably sensitive method.

TESTS
Safety. Use 2 non-vaccinated animals of one of the species for which the vaccine is intended, not less than 6 months old, having serum free from foot-and-mouth disease antibodies and coming from regions free from foot-and-mouth disease. Administer a double dose of vaccine to each animal by a recommended route. Observe the animals for 14 days. No abnormal local or systemic reaction occurs.

Sterility. It complies with the test for sterility prescribed in the monograph on Vaccines for veterinary use (0062).

POTENCY
The potency of the vaccine is expressed as the number of 50 per cent cattle protective doses (PD₅₀) contained in the dose stated on the label. The PD₅₀ is determined in animals given primary vaccination and challenged by the inoculation of 10 000 ID₅₀ of virulent bovine virus of the same serotype as that used in the preparation of the vaccine in the conditions described below. The vaccine strain may be used for challenge.

The potency is not less than that stated on the label; the minimum potency stated on the label is not less than 3 PD₅₀ per dose for cattle.

Carry out a potency test for each serotype of foot-and-mouth disease virus that may be included in the vaccine. The Potency test carried out for a particular serotype will be valid for other vaccines provided that they have the same basic composition and that they have a batch potency with regard to that particular serotype that is not lower than that of the vaccine that has given satisfactory results in the Potency test.

Use cattle not less than 6 months old, obtained from areas free from foot-and-mouth disease, which have never been vaccinated against foot-and-mouth disease and are free from antibodies neutralising the different serotypes of foot-and-mouth disease virus. Vaccinate not fewer than 3 groups of not fewer than 5 cattle per group by the route stated on the label. Use 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. 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.

3 weeks after the vaccination, challenge the vaccinated animals and a control group of 2 animals with a suspension of virus that has been obtained from cattle and that is fully virulent and of the same serotype as that used in the preparation of the vaccine, by inoculating a dose equivalent to approximately 10 000 ID₅₀ intradermally into 2 sites on the upper surface of the tongue (0.1 ml per site). Observe the animals for 8 days and then slaughter. Unprotected animals show lesions at sites other than the tongue. Protected animals may display linguistic lesions. The test is not valid unless each control animal shows lesions on at least 3 feet. From the number of protected animals in each group, calculate the PD₅₀ content of the vaccine.

LABELLING
The label states:
- the serotypes of virus used to prepare the vaccine,
- the minimum potency (PD₅₀ per dose) for each serotype of virus used to prepare the vaccine.

04/2005:1945

FOWL CHOLERA VACCINE (INACTIVATED)

Vaccinum cholerae aviaeavum inactivatum

1. DEFINITION
Fowl cholera vaccine (inactivated) is a preparation of 1 or more suitable strains of 1 or more serovars of Pasteurella multocida. This monograph applies to vaccines intended for the active immunisation of chickens, turkeys, ducks and geese against acute fowl cholera.

2. PRODUCTION
2-1. PREPARATION OF THE VACCINE
The seed material is cultured in a suitable medium. If the vaccine contains more than 1 strain of bacterium, the different strains are grown and harvested separately. The bacterial harvests are inactivated. The vaccine may contain an adjuvant.

2-2. CHOICE OF VACCINE COMPOSITION
The vaccine is shown to be satisfactory with respect to safety (5.2.6) and efficacy (5.2.7) for the species for which it is intended. The following tests for safety (section 2-2-1) and immunogenicity (section 2-2-2) may be used during the demonstration of safety and efficacy.

2-2-1. Safety. The test is carried out for each route of administration to be recommended for vaccination and for each avian species for which the vaccine is intended. For each test, use not fewer than 20 birds not older than the minimum age to be recommended for vaccination. In the case of chickens, use chickens from a flock free from specified pathogens (SPF) (5.2.2) and in the case of turkeys, ducks or geese, use birds that have not been vaccinated and that are free from antibodies against P. multocida. Administer by a recommended route and method to each bird a double dose of vaccine. If the recommended schedule requires a second dose, administer 1 dose after the recommended interval. Observe the birds at least daily until 21 days after the last administration of the vaccine. The test is not valid if more than 10 per cent of the birds show abnormal clinical signs of disease or die from causes not attributable to the vaccine. The vaccine complies with the test if no bird shows abnormal clinical signs of disease or dies from causes attributable to the vaccine.

2-2-2. Immunogenicity. The test is carried out for each route of administration to be recommended for vaccination, for each avian species for which the vaccine is intended and for each serovar of P. multocida against which protection is claimed. Use for each test not fewer than 30 birds not older than the youngest age to be recommended for vaccination.

Use birds that have not been vaccinated and that are free from antibodies against P. multocida. For each test, administer to each of not fewer than 20 birds a quantity of the vaccine not greater than 1 dose. If re-vaccination is recommended, repeat this operation after the recommended interval. Maintain not fewer than 10 birds as controls. Challenge each of the birds of both groups 21 days after the last administration by the intramuscular route with a sufficient quantity of virulent...
Fowl cholera vaccine (inactivated)

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P. multocida. Observe the birds at least daily for 14 days after challenge. The test is not valid if during the observation period after challenge, fewer than 70 per cent of the control birds die or show signs of infection (such as either clinical signs or bacterial re-isolation in organs) or if during the period before challenge, more than 10 per cent of the birds from the control group or from the vaccinated group show abnormal clinical signs of disease or die from causes not attributable to the vaccine. The vaccine complies with the test if, at the end of the observation period after challenge, not fewer than 70 per cent of the birds from the vaccinated group survive and show no clinical signs of disease. Mild signs that do not persist beyond the observation period may be tolerated.

2-3. Batch potency. It is not necessary to carry out the Potency test (section 3-4) for each batch of the vaccine if it has been carried out using a batch of vaccine with minimum potency. 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 Immunogenicity (section 2-2-2). The following test may be used.

Use not fewer than 15 SPF chickens (5.2.2), 3 to 4 weeks old. Collect serum samples from each vaccinated and control chicken just before vaccination and check the absence of antibodies against each serovar of P. multocida in the vaccine. Administer to each of 10 chickens 1 dose of the vaccine by the subcutaneous route. Maintain 5 chickens as controls. Collect serum samples 5 weeks after vaccination from each vaccinated and control chicken. Measure the titres of serum antibodies against each serovar of P. multocida stated on the label using a suitable validated serological method. Calculate the mean titres for the group of vaccinates. The test is not valid if specific P. multocida antibodies are found: before vaccination in 1 or more sera from chickens to be vaccinated or from controls; in 1 or more sera from control chickens 5 weeks after the time of administration of the vaccine. The vaccine complies with the test if the mean antibody titres of the group of vaccinates are equal to or greater than the titres obtained with a batch that has given satisfactory results in the test described under Immunogenicity (section 2-2-2).

2-4. Bacterial endotoxins. A test for bacterial endotoxins (2.6.14) is carried out on the final lot or, where the nature of the adjuvant prevents performance of a satisfactory test, on the bulk antigen or the mixture of bulk antigens immediately before addition of the adjuvant. The maximum acceptable amount of bacterial endotoxins is established and validated in the test described under Immunogenicity (section 2-2-2). The following test may be used.

3. BATCH TESTS

3-1. Identification. The vaccine injected into SPF chickens (5.2.2) stimulates the production of antibodies against each of the serovars of P. multocida in the vaccine.

3-2. Bacteria and fungi. The vaccine complies with the test for sterility prescribed in the monograph Vaccines for veterinary use (0062).

3-3. Safety. For vaccines recommended for use in chickens, use not fewer than 10 chickens from an SPF flock (5.2.2) and of the minimum age recommended for vaccination. For vaccines recommended for use only in turkeys, ducks or geese, use not fewer than 10 birds of the species likely to be most sensitive to fowl cholera, that do not have antibodies against P. multocida and of the minimum age recommended for vaccination. Administer to each bird by a recommended route a double dose of the vaccine. Observe the birds at least daily for 21 days. The test is not valid if more than 20 per cent of the birds show abnormal clinical signs or die from causes not attributable to the vaccine. The vaccine complies with the test if no bird shows notable clinical signs of disease or dies from causes attributable to the vaccine.

3-4. Potency. The vaccine complies with the requirements of the test mentioned under Immunogenicity (section 2-2-2) using 1 dose of the vaccine administered by a recommended route.

LABELLING

The label states:
– the serovar(s) used to prepare the vaccine,
– the serovar(s) against which protection is claimed.