Animal and Plant Health Inspection Service, USDA

§ 113.208 Avian Encephalomyelitis

Vaccine, Killed Virus.

Avian Encephalomyelitis Vaccine (Killed Virus) shall be prepared from virus-bearing tissues or fluids obtained from embryonated chicken eggs. Each serial shall meet the general requirements prescribed in § 113.200 and the requirements prescribed in this section. Any serial found unsatisfactory by a prescribed test shall not be released.

(a) Safety test. Each of at least ten 6 to 12 hour old chickens shall be injected subcutaneously with 0.5 ml of the product and the chickens observed each day for 10 days.

(2) If unfavorable reactions attributable to the product occur in the chickens during the observation period, the test is unsatisfactory. If unfavorable reactions not attributable to the product occur, the test is inconclusive and may be repeated: Provided, That, if the test is not repeated, the serial is unsatisfactory.

(b) Potency test. Fourteen to 21 days after the second injection, serum samples from each vaccinate and each control shall be tested by a plaque reduction, serum neutralization test using Vero 76 cells.

(3) If the control serum samples show a titer of less than 1:40 for the Eastern type fraction, less than 1:40 for the Western type fraction, or less than 1:4 for the Venezuelan type fraction, the second stage of the test may be used for the relevant fraction(s): Provided, That, if a fraction is found acceptable by the first stage of the test, the second stage need not be conducted for that fraction.

(4) If two or three of the vaccinate serum samples show a titer of less than 1:40 for the Eastern type fraction, less than 1:40 for the Western type fraction, or less than 1:4 for the Venezuelan type fraction, the serial or subserial is unsatisfactory.

(5) The results shall be evaluated according to the following table:

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<tr>
<th>CUMULATIVE TOTALS</th>
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<td>Stage</td>
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(i) Chicken Embryo Test. Each of 15 or more AE susceptible 5 or 6 day old embryos shall be injected in the yolk sac with 0.2 ml of the vaccine. For a valid test, at least 80 percent of the embryos shall survive for 48 hours post-inoculation (PI). Eleven to 13 days PI, all embryos surviving the 48 hour PI period shall be examined for gross lesions of AE; all these embryos shall be normal or the serial is unsatisfactory. Concurrently, five additional embryos from the same source shall be injected with live AE virus of the production strain to serve as positive controls. At least 4 of the 5 embryos shall show evidence of AE virus infection during the 11 to 13 day PI period or the test shall be considered inconclusive and repeated: Provided. That, if the test is not repeated, the serial shall be declared unsatisfactory.

(ii) Chicken test. Each of 10 or more AE susceptible 7 day old chickens shall be injected intracerebrally with 0.1 ml vaccine each. The chickens shall be observed each day for 28 days. If any chickens show clinical signs of AE, the serial is unsatisfactory. Concurrently, 5 additional chickens from the same source shall be injected intracerebrally with live AE virus of the production strain to serve as positive controls. At least 4 of the 5 controls shall show evidence of AE virus infection during the observation period or the test shall be inconclusive and may be repeated: Provided. That, if the test is not repeated, the serial shall be unsatisfactory.

(b) Potency test. Bulk or final container samples of completed product from each serial or one subserial shall be tested. Ten or more AE-susceptible chickens (vaccinates), 4 weeks or older, properly identified and obtained from the same source and hatch, shall be injected as recommended on the label. At least 10 additional AE-susceptible chickens, properly identified and obtained from the same source and hatch shall be kept in isolation as controls.

(1) At least 28 days post-inoculation, the vaccinates and the controls shall be challenged intramuscularly with a virulent AE virus and the chickens observed each day for 21 days.

(2) If at least 80 percent of the controls do not show clinical signs of or die from AE infection, the test is inconclusive and may be repeated.

(3) If at least 80 percent of the vaccinates do not remain normal, the serial is unsatisfactory.