bearing tissues or fluids obtained from embryonated chicken eggs or cell cultures. With the exception of §113.200(c)(2)(iii), each serial shall meet the applicable general requirements prescribed in §113.200 and special requirements prescribed in this section. A serial found unsatisfactory by a prescribed test shall not be released.

(a) *Safety test.* The prechallenge part of the potency test in paragraph (b) of this section shall constitute a safety test. If unfavorable reactions attributable to the product occur in any of the vaccinates, the serial is unsatisfactory. If unfavorable reactions which are not attributable to the product occur, the test shall be declared inconclusive and may be repeated: *Provided*, That, if the test is not repeated, the serial shall be declared unsatisfactory.

(b) *Potency test.* A vaccination-challenge test shall be conducted using susceptible chickens 2 to 6 weeks of age at time of vaccination, properly identified and obtained from the same source and hatch.

(1) Ten or more chickens shall be vaccinated as recommended on the label and kept isolated under observation for at least 14 days.

(2) After at least 14 days post-vaccination, the vaccinates and at least 10 unvaccinated chickens that have been kept isolated as controls shall be challenged with a virulent strain of Newcastle disease virus supplied by or approved by Veterinary Services and the vaccinates observed each day for 14 days.

(3) If at least 90 percent of the controls do not show typical signs of Newcastle disease or die, the test is inconclusive and may be repeated. If at least 90 percent of the vaccinates do not remain normal, the serial is unsatisfactory.

[39 FR 27428, July 29, 1974. Redesignated at 55 FR 35562, Aug. 31, 1990, as amended at 56 FR 66786, Dec. 26, 1991]

§113.206 Wart Vaccine, Killed Virus.

Wart Vaccine, Killed Virus, shall be prepared from virus-bearing epidermal tumors (warts) obtained from a bovine. Each serial shall meet the requirements prescribed in this section and any serial found unsatisfactory by a prescribed test shall not be released.

(a) *Purity.* Final container samples of completed product shall meet the requirements for purity as prescribed in §113.200 (c)(1) and (3).

(b) *Safety.* Bulk or final container samples of completed product shall meet the requirements for safety as prescribed in §§113.33(b) and 113.38.

(c) *Formaldehyde content.* Bulk or final container samples of completed product shall meet the requirements for formaldehyde content as prescribed in §113.200(f).

(d) *Potency and efficacy.* The efficacy of wart vaccine has been demonstrated to the satisfaction of Veterinary Services as being a valuable biological product. The inherent nature of the product precludes the possible development of serial to serial potency tests and none is required: *Provided*, That,

(1) The vaccine shall be a tissue extract representing at least 10 percent weight to volume suspension of wart tissue; and

(2) The vaccine shall be limited to use in the prevention of warts in cattle. Labeling recommendations shall be in accordance with §112.7(i).

[40 FR 14084, Mar. 28, 1975, as amended at 40 FR 23989, June 4, 1975; 40 FR 30803, July 23, 1975. Redesignated at 55 FR 35562, Aug. 31, 1990, as amended at 56 FR 66786, Dec. 26, 1991]

§113.207 Encephalomyelitis Vaccine, Eastern, Western, and Venezuelan, Killed Virus.

Encephalomyelitis Vaccine, Eastern, Western, and Venezuelan, Killed Virus, shall be prepared from virus-bearing cell culture fluids. Each serial or subserial shall meet the requirements prescribed in this section and the general requirements prescribed in §113.200, except those in §113.200(d). Any serial or subserial found unsatisfactory by a prescribed test shall not be released.

(a) *Safety test.* Bulk samples of completed product from each serial shall be tested for encephalomyelitis virus inactivation.

(1) 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 serial 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.* Bulk or final container samples of completed product from each serial shall be tested for potency in accordance with the two-stage test provided in this paragraph. For each fraction contained in the product—Eastern type, Western type, or Venezuelan type—the serological interpretations required in this test shall be made independently. A serial or subserial found unsatisfactory for any of the fractions shall not be released.

(1) For this test, a guinea pig dose shall be one-half the amount recommended on the label for a horse and shall be administered as recommended for a horse. Each of 10 healthy guinea pigs (vaccinates) shall be injected with two guinea pig doses with an interval of 14 to 21 days between doses. Two additional guinea pigs from the same source shall be held as controls.

(2) 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 1:4 or greater for any fraction, the test is inconclusive for that fraction and may be repeated: *Provided*, That, if four or more 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 without further testing.

(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 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.

(5) If the second stage is used and four or more of the vaccinate serum samples show a titer of less than 1:40 for the Eastern type fraction or the Western type fraction, or less than 1:4 for the Venezuelan type fraction, the serial or subserial is unsatisfactory.

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

CUMULATIVE TOTALS

Stage	Vaccinates	Failures for acceptance	Failures for rejection
1	10	1 or less	4 or more.
2	20	3 or less	Do.

[39 FR 44714, Dec. 27, 1974, as amended at 40 FR 14084, Mar. 28, 1975; 42 FR 45284, Sept. 9, 1977. Redesignated at 55 FR 35562, Aug. 31, 1990, as amended at 56 FR 66786, Dec. 26, 1991; 61 FR 67930, Dec. 26, 1996]

§ 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 tests.* (1) The prechallenge part of the potency test prescribed in paragraph (b) of this section shall constitute a safety test. If any of the vaccinates develop clinical signs of disease or die due to causes attributable to the product, the serial is unsatisfactory.

(2) An inactivation test for viable avian encephalomyelitis (AE) virus shall be conducted on each serial. The test shall be conducted using susceptible chicken embryos: *Provided*, That, if a non-embryo adapted virus is used for vaccine production, the test shall be conducted in susceptible chickens.

(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