§ 630.66 General requirements.

(a) Final container tests. In addition to the tests required pursuant to §610.14 of this chapter, an immunological and virological identity test shall be performed on the final container if it was not performed on each pool or on the bulk vaccine prior to filling.

(b) Dose. These standards are based on an individual human immunizing dose of no less than 1,000 TCID₅₀ of Rubella Virus Vaccine Live, expressed in terms of the assigned titer of the Reference Rubella Virus, Live.

(c) Labeling. In addition to the items required by other applicable labeling provisions of this subchapter, single dose container labeling for vaccine which is not protected against photochemical deterioration shall include a statement cautioning against exposure to light.

(d) Clarification. The rubella virus fluids shall be clarified by following the procedures prescribed in §630.35(c).

(2) In addition to the requirements of paragraph (e)(1) of this section, whenever a new production seed lot is introduced, or whenever the source of cell culture substrate must be reestablished and recertified, samples consisting of no less than 100 milliliters in 10-milliliter volumes, in a frozen state (—60°C), of postclarification bulk vaccine containing stabilizer but no preservative or adjuvant, taken from each of 5 consecutive lots of the bulk vaccine.

(3) The product shall not be issued by the manufacturer until written notification of official release of the lot is received from the Director, Center for Biologics Evaluation and Research.


Subpart H—Smallpox Vaccine

§ 630.70 Smallpox Vaccine.

(a) Proper name and definition. The proper name of this product shall be Smallpox Vaccine, which shall be a preparation of live vaccinia virus obtained from inoculated calves or chicken embryos.

(b) Strains of virus. The strain of seed virus used in the manufacture of Smallpox Vaccine shall be identified by historical records including origin and manipulation, and shall meet the sterility test requirements when tested by the procedure prescribed in §610.12 of this chapter. The strain of seed virus and every third passage shall be tested by a rabbit scarification procedure and shown to maintain its original dermatropic properties. The test procedure is available upon request from the Director, Center for Biologics Evaluation and Research. Any new strain shall be shown not to produce a

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§ 630.71 Production.

Vaccinia virus used for the manufacture of vaccine shall be obtained from vesicles on the skin of an inoculated calf or from inoculated chorioallantoic membranes of chicken embryos, as set forth below:

(a) Virus from calves—(1) Quarantine. Only calves which, prior to being placed in quarantine have reacted negatively to tuberculin, were afebrile and free of ectoparasites, and which shall have met all other applicable quarantine requirements of §600.11(f)(2)(i) of this chapter, shall be used for vaccinia virus production. The quarantine period shall be at least 14 days. During the last 7 days of the quarantine period daily morning and afternoon rectal temperatures shall be taken and calves that do not remain afebrile during that period shall not be used for virus production.

(2) Inoculation. A larger area of the calf than will be used for production purposes shall be prepared in a manner comparable to that appropriate for aseptic surgery, except that the area to be inoculated must be washed free of all antiseptics that may have a deleterious effect on virus propagation. The instrument and method used for scarification must produce a uniform penetration into the epidermis but must not extend through into the corium.

(3) Incubation. The inoculated calf shall remain in the incubation room confined to its stall and daily morning and afternoon rectal temperatures shall be taken to determine that only the expected febrile condition occurs. If any signs of disease other than vesiculation at the inoculation site occur, the virus from that calf shall not be used for vaccine manufacture.

(4) Harvesting. Before harvesting, the calf shall be anesthetized and killed by exsanguination. Prior to harvesting, the inoculated area shall be thoroughly cleansed by aseptic techniques. Only the vesicular material shall be harvested.

(b) Virus from embryonated chicken eggs—(1) Eggs for production. Embryonated chicken eggs used for propagation of vaccinia virus shall be derived from flocks found to be free of, and continuously monitored for freedom from Salmonella pullorum, Mycoplasma species, avian tuberculosis, fowl pox, Newcastle disease virus, Rous sarcoma virus, avian leucosis complex of viruses, and other agents pathogenic for chickens, or appropriate tests shall be performed to demonstrate freedom of the vaccine from such agents.

(2) Harvesting. Aseptic techniques shall be used in harvesting the chorioallantoic membranes exhibiting vesicles characteristic of vaccinia infection.

§ 630.72 Reference vaccine.

Reference Smallpox Vaccine and reconstitution fluid shall be obtained from the Center for Biologics Evaluation and Research and shall be used in all tests for determining the potency of Smallpox Vaccine.

§ 630.73 Potency test.

Each filling of Smallpox Vaccine shall be tested for potency by the "pock count" method as follows:

(a) [Reserved]

(b) Pock counting in embryonated chicken eggs—(1) Dilutions shall be made starting with no less than 0.5 ml. of the test vaccine and of the reference vaccine. The same diluent shall be used for all dilutions of both vaccines. The sample of vaccine in capillary tubes shall be obtained by pooling the contents of no less than 50 capillaries into a sterile vessel.

(2) Inoculation of embryonated chicken eggs. One-tenth milliliter of each dilution of test vaccine shall be inoculated onto the chorioallantoic membrane of each of at least five embryonated chicken eggs. The reference vaccine shall be tested in the same manner.
§ 630.74 Tests for safety.

(a) Anaerobes. A 10-milliliter sample representative of the homogenized viral harvest or pool of several viral harvests shall be tested for the presence of anaerobes in the following manner: Before the addition of preservatives other than glycerin, the test sample shall be inoculated into freshly heated Fluid Thioglycollate Medium using a ratio of inoculum to culture medium sufficient for optimal bacterial growth. The test vessels shall be incubated at 35° to 37°C and observed daily for 10 days for evidence of bacterial growth. If bacterial growth is observed, the organism(s) shall be identified as to genus. Within 24 to 48 hours of an indication that there may be anaerobic growth, 1.0-milliliter samples from each vessel showing growth shall be inoculated subcutaneously into each of at least three mice weighing not more than 20 grams each, and into each of three guinea pigs weighing not more than 350 grams each. The animals shall be observed daily for 6 days for signs of tetanus or presence of other anaerobes. If the animals show no signs of tetanus or presence of other anaerobes, additional groups of the same types and numbers of animals shall be injected 9 days after evidence of anaerobic bacterial growth is observed in the original planting with 1.0-milliliter samples from each test vessel showing growth. The animals shall be observed daily for 6 days. If any animals die within 3 days without having shown signs of tetanus or presence of other anaerobes, the test shall be repeated within 18 hours of the deaths, with 0.1-milliliter samples of the culture from which that animal was inoculated. Samples from the culture shall be injected into each of three additional test animals of the same species, and the animals shall be observed daily for 6 days. If there is any evidence of the presence of pathogenic anaerobes, the viral harvest may not be used in the manufacture of Smallpox Vaccine.

(b) [Reserved]

(c) Coliform organisms. A 5.0 ml. sample of bulk vaccine shall be tested for the presence of coliform organisms by the method published by the American Public Health Association, Inc., in “Standard Methods for the Examination of Water and Wastewater” (13th edition, 1971), section entitled “Multiple-Tube Fermentation Technic for
Members of the Coliform Group,” pages 662-678 and any amendments or revisions thereof, which section is hereby incorporated by reference and deemed published herein. Said publication is available at most medical and public libraries and copies of the pertinent section will be provided to any manufacturer affected by the provisions of this part upon request to the Director, Center for Biologics Evaluation and Research, or to the appropriate Information Center Officer listed in 45 CFR part 5. An official historic file of the material incorporated by reference is maintained in the Office of the Director, Center for Biologics Evaluation and Research, or available for inspection at the Office of the Federal Register, 800 North Capitol Street NW., suite 700, Washington, DC 20408. A method different than that contained in the above cited section may be used to test for the presence of coliform organisms upon a showing that it is of equal or greater sensitivity. The ratio of the volume of inoculum to the volume of culture medium shall be such as will dilute the preservative to a level that does not inhibit growth of contaminating micro-organisms. The vaccine is satisfactory if there is no evidence of coliform organisms.

(d) Hemolytic streptococci and coagulase-positive staphylococci. Each of three 1.0 ml. samples of bulk vaccine shall be spread uniformly on the surface of separate blood agar plates. The plates shall be incubated for 48 hours at 35° to 37° C. The vaccine is satisfactory if there is no evidence of the presence of either hemolytic streptococci or coagulase-positive staphylococci.

(e) Viable bacteria—(1) Vaccine intended for multiple pressure administration. Samples of each lot of both bulk and final container vaccine shall be tested for viable bacteria by a procedure designed to detect both aerobic and anaerobic growth through a period of 7 days. At least three 1.0 ml. samples of bulk vaccine and three 0.2 ml. samples of vaccine derived from not less than three final containers or dilutions thereof shall be inoculated into a volume of culture medium sufficient for optimal bacterial growth. The vaccine is satisfactory if it contains no more than 200 viable organisms per ml.

(2) Vaccine intended for jet injection. Samples of each lot of both bulk and final container vaccine shall be tested for viable bacteria in Fluid Thioglycollate Medium prepared in accordance with §610.12(e)(1)(i) of this chapter for at least a 7-day test period. A sample of at least 10.0 ml. of bulk vaccine and 1.0 ml. from each of at least 20 final containers shall be tested. The ratio of the volume of the inoculum to the volume of culture medium shall be such as will dilute the preservative in the inoculum to a level that does not inhibit growth of contaminating micro-organisms. The vaccine is satisfactory if it contains no more than one organism per 100 doses of vaccine.

(f) Sterile vaccine. The tests prescribed in paragraphs (c), (d), and (e) of this section need not be performed on a lot of Smallpox Vaccine that meets the sterility requirements prescribed in §610.12 of this chapter.

§ 630.75 General requirements.

(a) General safety. Each lot of vaccine shall be tested for safety as prescribed in §610.11 of this chapter and shall meet the safety requirements of that section, except that for liquid Smallpox Vaccine distributed in capillaries, the test may be performed with a sample of bulk vaccine taken at the time of filling into final containers.

(b) Preservative. A preservative that meets the requirements of §610.15 of this chapter may be used, provided that if the preservative is phenol, its concentration shall not exceed 0.5 percent.

(c) Labeling. In addition to complying with all other applicable labeling provisions of this subchapter the package label shall bear the following:

(1) Vaccine intended for jet injection. (i) A conspicuous statement that the vaccine is intended for administration by jet injector.

(ii) A statement that the vaccine has been shown by appropriate test methods to contain not more than one organism per 100 doses or reference to an enclosed circular that contains such...
information, except that such a statement is not required for vaccine which meets the sterility requirements of §610.12 of this chapter.

(2) Vaccine intended for multiple pressure administration. A statement that the vaccine has been shown by appropriate test methods to contain not more than 200 organisms per ml. or reference to an enclosed circular that contains such information, except that such a statement is not required for vaccine which meets the sterility requirements of §610.12 of this chapter.

(d) Samples; protocols; official release.

(1) For each lot of vaccine the following shall be submitted to the Director, Center for Biologics Evaluation and Research, Food and Drug Administration, 8800 Rockville Pike, Bethesda, MD 20892:

(i) A protocol which consists of a summary of the history of manufacture of each filling including all results of each test for which test results are requested by the Director, Center for Biologics Evaluation and Research.

(ii) Three hundred capillaries from the first filling of a lot of liquid vaccine, and 200 capillaries from each subsequent filling.

(iii) Two 10 ml. samples of bulk liquid vaccine to be submitted along with the capillaries from the first filling and taken from the same vessel from which such capillaries were filled.

(iv) For vaccine intended for jet gun injection, a sample from each drying consisting of no less than eight 100-dose vials or eight 500-dose vials of vaccine in final labeled containers, plus sufficient diluent in final labeled containers to reconstitute the vaccine.

(v) For vaccine intended for multiple pressure administration, a sample from each drying consisting of no less than eighty 10-dose vials, ninety 25-dose vials, or eighty 100-dose vials of vaccine in final labeled containers, plus sufficient diluent in final labeled containers to reconstitute the vaccine.

(2) The product shall not be issued by the manufacturer until written notification of official release of the lot is received from the Director, Center for Biologics Evaluation and Research.