

required by paragraph (e) of this section, are required for a serial of autogenous biologic with 50 or fewer containers.

(9) *Miscellaneous*: The number of samples from products not in the categories provided for in paragraphs (b)(1) through (b)(8) of this section shall be prescribed in the filed Outline of Production for the product.

(c) *Prelicensing and Outline of Production changes*: Samples needed to support a license application or a change in the Outline of Production for a licensed product shall be submitted only upon request from the animal and Plant Health Inspection Service. Except for miscellaneous products specified in paragraph (b)(9) of this section, the number of such samples shall be at least one and one-half times the number prescribed for such product in paragraph (b) of this section. Samples of Master Seeds and Master Cell Stocks with a minimum individual volume of 1 ml shall be submitted as follows:

(1) Ten samples of Bacterial Master Seeds.

(2) Thirteen samples of viral Master Seeds or nonviral Master Seeds requiring cell culture propagation. For Master Seeds isolated or passed in a cell line different from the species of intended use, an additional 2 samples are required for each additional species. For Master Seeds grown in cell culture and intended for use in more than one species, an additional 2 samples are required for each additional species.

(3) Thirty-six samples of at least 1 ml each or six samples of at least 1 ml each, one sample of at least 20 ml, and one sample of at least 10 ml of Master Cell Stocks. In the case of Master Cell Stocks which are persistently infected with a virus, an additional four samples of at least 1 ml each are required. If these persistently infected cell stocks are intended for use in more than one species, an additional two samples of at least 1 ml each are required for each additional species.

(4) Four samples of the Master Cell Stock + n (highest passage) cells.

(d) *Sterile diluent*: A sample of Sterile Diluent shall accompany each sample of product, other than Marek's Disease Vaccine, if such diluent is required to rehydrate or dilute the product before use. The volume of diluent shall be an appropriate amount to rehydrate or dilute the product. Samples of Sterile Diluent prepared for use with Marek's Disease Vaccine shall be submitted upon request from the Animal and Plant Health Inspection Service.

(e) Reserve samples shall be selected from each serial and subserial of biological product. Such samples shall be selected at random from final

containers of completed product by an employee of the Department, of the licensee, or of the permittee, as designated by the administrator. Each sample shall:

(1) Consist of 5 single-dose packages, 2 multiple-dose packages, or 2 diagnostic test kits, except that, in the case of diagnostic test kits in which final packaging consists of multiple microtiter test plates or strips, a sample may consist of a specified number of test plates or strips along with all other test reagents as prescribed in a filed Outline of Production;

(2) Be adequate in quantity for appropriate examination and testing;

(3) Be truly representative and in final containers;

(4) Be held in a special compartment set aside by the licensee or permittee for holding these samples under refrigeration at the storage temperature recommended on the labels for 6 months after the expiration date stated on the labels. The samples that are stored in this manner shall be delivered to the Animal and Plant Health Inspection Service upon request.

(Approved by the Office of Management and Budget under control number 0579-0013)

§ 113.309 [Amended]

3. In § 113.309, paragraph (c)(4), the words "Veterinary Services" are removed and the words "Animal and Plant Health Inspection Service" are added in their place.

Done in Washington, DC, this 13th day of March 1995.

Terry L. Medley,

Acting Administrator, Animal and Plant Health Inspection Service.

[FR Doc. 95-6649 Filed 3-16-95; 8:45 am]

Billing Code 3410-34-M

9 CFR Part 113

[Docket No. 92-132-2]

Viruses, Serums, Toxins, and Analogous Products; Revision of Standard Requirements

AGENCY: Animal and Plant Health Inspection Service, USDA.

ACTION: Final rule.

SUMMARY: This rule amends the Standard Requirements concerning Dog Safety Testing; Canine Distemper Vaccine, Killed Virus; Canine Hepatitis Vaccine, Killed Virus; Canine Adenovirus Type 2 Vaccine, Killed Virus; Mink Enteritis Vaccine, Killed Virus; Canine Hepatitis Vaccine, Live Virus; Canine Adenovirus Type 2 Vaccine, Live Virus; and Canine

Distemper Vaccine, Live Virus. The amendments are necessary because new test methods and procedures have been developed that can replace current test requirements and increase the validity of test results. The effect of the amendments is to provide new test methods and procedures and to relax some of the restrictions currently in effect. Also, the Standard Requirement for Canine Distemper Vaccine (Ferret Virulent) is removed because this vaccine is no longer manufactured.

EFFECTIVE DATE: April 17, 1995.

FOR FURTHER INFORMATION CONTACT: Dr. David A. Espeseth, Deputy Director, Animal and Plant Health Inspection Service, Biotechnology, Biologics, and Environmental Protection, Veterinary Biologics, 4700 River Road Unit 148, Riverdale, MD 20737-1228, (301) 734-8245.

SUPPLEMENTARY INFORMATION:

Background

The regulations in 9 CFR part 113 "Standard Requirements", (referred to below as the regulations) consist of test methods, procedures, and criteria established by the Animal and Plant Health Inspection Service (APHIS) for the evaluation of veterinary biological products based upon their purity, safety, potency, and efficacy. The Agency periodically reviews the regulations and amends test methods and procedures as required to ensure that they are consistent with current scientific knowledge. On July 23, 1993, we published in the **Federal Register** (see 58 FR 39467-39473, Docket No. 92-132-1) a proposed rule to update the regulations based upon current scientific knowledge.

We solicited comments concerning our proposal for a 60-day comment period ending September 21, 1993. We received four comments by that date. One commenter fully supported the proposal as written. Three commenters suggested changes to certain sections related to the Standard Requirements. These comments are discussed below.

Two commenters suggested changes to § 113.204. Both commenters indicated that the portion of the regulations dealing with the time(s) of feces collection for virus detection required clarification, and suggested that feces collection at some point from day 4 to 8 would be appropriate.

APHIS believes that the above comments have merit. APHIS agrees that feces collection early or late in the collection period, or more than once, is unnecessary. Therefore, APHIS has revised the regulations in § 113.204(b)(2) to specify that feces are

to be collected once from day 4 through day 8.

One of the two commenters stated that the term "virus isolation" should be defined and that methods other than that specified, "virus isolation and/or fluorescent antibody examination," should be allowed for the detection of virus in feces. APHIS agrees that not only "virus isolation" but the whole phrase "virus isolation and/or fluorescent antibody examination" needs clarification. Therefore, we have revised the regulations in § 113.204(b)(2) to specify that cell culture with fluorescent antibody examination is the acceptable method of virus detection. APHIS does not agree, however, with the suggestion that other methods of virus detection should be specified in the regulations presently. We believe that cell culture with fluorescent antibody examination is the most sensitive and specific method of virus detection. Should APHIS become aware of another method that is superior to that indicated, it would consider rulemaking to specify that method in the regulations.

One of the two commenters also stated that unvaccinated control mink in immunogenicity studies should not be considered susceptible to challenge if the animals exhibit clinical signs but do not shed virus. The second commenter stated that the determination of virus shedding in animals used for immunogenicity studies should not be required if four or five of the five unvaccinated control mink exhibit clinical signs. APHIS does not agree with either commenter. We believe that the absence of appropriate clinical signs in vaccinated mink challenged with virulent mink enteritis virus together with clinical signs in unvaccinated control mink after challenge is sufficient evidence of the effectiveness of the challenge. We also believe that an effective vaccine against mink enteritis should prevent virus shedding. No change in the regulations is made in response to these comments.

As a final comment on § 113.204, one commenter criticized what the commenter thought was a Standard Assay Method (SAM) developed by the National Veterinary Services Laboratories for challenging mink with mink enteritis virus. No such SAM has been prepared. What has been prepared is more appropriately termed a "bench protocol." Since the protocol was not addressed in the proposed rulemaking, no change to the regulations is made based on this comment.

Comments on the three other Standard Requirements included in the proposed rule (§§ 113.40, 113.201, and

113.306) were received from another commenter. Two of the comments related to the route of canine distemper virus challenge and the requirements for satisfactory vaccine performance in a repeat immunogenicity study. The commenters requested that § 113.306 concerning live virus vaccines be changed to specify an intranasal rather than the traditional intracerebral route of challenge. No amendments to the route of challenge or repeat immunogenicity requirements were proposed in § 113.306. An intracerebral challenge has been used successfully for many years with the live virus vaccine. It was the proposed amendments to § 113.201 that changed the route of challenge from intranasal to intracerebral for killed virus vaccines to be consistent with that specified for live virus vaccines in § 113.306. Since the commenters focussed only on § 113.306 and that section is not being amended as to route of challenge, no change to the regulations is made in response to these commenters.

The same commenter also claimed that the proposal would result in the overuse of Master Seed and suggested that material obtained after five passages of Master Seed be used instead. APHIS disagrees with this comment. In requiring that Master Seed be used, the proposed change is consistent with other regulations in part 113. We believe that testing the Master Seed is necessary for a satisfactory determination of its use. No change to the regulations is made in response to this commenter.

Based on the rationale set forth in the proposed rule and in this document, we are adopting the provisions of the proposed rule as a final rule, with the changes discussed in this document.

Executive Order 12866 and Regulatory Flexibility Act

This proposed rule has been reviewed under Executive Order 12866. The rule has been determined to be not significant for the purposes of Executive Order 12866 and, therefore, has not been reviewed by the Office of Management and Budget.

This final rule revises the current Standard Requirements for certain vaccines. Sections 113.201 and 113.202 are amended to revise the potency test performed on each serial of product so that fewer dogs will be used and serology will be used instead of virus challenge. Both of these changes will decrease the costs of production to the manufacturer. In § 113.204, the potency test in mink is changed to require that virus shedding be examined. This change should result in only a minimal increase in cost (less than \$100 per test)

to the manufacturer. Other changes to the Standard Requirements generally update the Standard Requirements to reflect current scientific knowledge. We do not expect any increase in cost, except as noted above, to the 200 biologics manufacturers affected by this rule. In most cases, we expect the changes will actually decrease the costs for the manufacturers.

Under these circumstances, the Administrator of the Animal and Plant Health Inspection Service has determined that this action will not have a significant economic impact on a substantial number of small entities.

Executive Order 12372

This program/activity is listed in the category of Federal Domestic Assistance under No. 10.025 and is subject to Executive Order 12372, which requires intergovernmental consultation with State and local officials. (See 7 CFR part 3015, subpart V.)

Paperwork Reduction Act

This rule contains no new information collection or recordkeeping requirements under the Paperwork Reduction Act of 1980 (44 U.S.C. 3501 *et seq.*).

Executive Order 12778

This final rule has been reviewed under Executive Order 12778, Civil Justice Reform. This rule: (1) Preempts all State and local laws and regulations that are in conflict with this rule; (2) has no retroactive effect; and (3) does not require administrative proceedings before parties may file suit in court challenging this rule.

List of Subjects in 9 CFR Part 113

Animal biologics, Exports, Imports, Reporting and recordkeeping requirements.

Accordingly, 9 CFR part 113 is amended as follows:

PART 113—STANDARD REQUIREMENTS

1. The authority citation for part 113 continues to read as follows:

Authority: 21 U.S.C. 151–159; 7 CFR 2.17, 2.51, and 371.2(d).

2. Section 113.40 is revised to read as follows:

§ 113.40 Dog safety tests.

The safety tests provided in this section shall be conducted when prescribed in a Standard Requirement or in the filed Outline of Production for a biological product recommended for use in dogs. Serials which are not found to be satisfactory when tested pursuant to

the procedures in this section may not be released for shipment.

(a) The dog safety test provided in this paragraph shall be used when the Master Seed Virus is tested for safety.

(1) The test animals shall be determined to be susceptible to the virus under test by a method acceptable to the Animal and Plant Health Inspection Service.

(2) Each of at least 10 susceptible dogs shall be administered a sample of the Master Seed Virus equivalent to the amount of virus to be used in one dog dose of the vaccine, by the method recommended on the label, and the dog shall be observed each day for 14 days.

(3) If unfavorable reactions attributable to the virus occur in any of the dogs during the observation period, the Master Seed Virus is unsatisfactory. If unfavorable reactions occur which are not attributable to the Master Seed Virus, the test shall be declared inconclusive and may be repeated: *Provided*: That, if the test is not repeated, the Master Seed Virus shall be considered unsatisfactory.

(b) The dog safety test provided in this paragraph shall be used when a serial of vaccine is tested for safety before release.

(1) Each of two healthy dogs shall be administered 10 dog doses by the method recommended on the label and the dogs shall be observed each day for 14 days.

(2) If unfavorable reactions attributable to the biological product occur during the observation period, the serial is unsatisfactory. If unfavorable reactions occur which are not attributable to the biological product, the test shall be declared inconclusive and may be repeated: *Provided*, That, if the test is not repeated, the serial shall be considered unsatisfactory.

3. Section 113.201 is revised to read as follows:

§ 113.201 Canine Distemper Vaccine, Killed Virus.

Canine Distemper Vaccine, Killed Virus, shall be prepared from virus-bearing cell culture fluids. Only Master Seed Virus which has been established as pure, safe, and immunogenic shall be used for vaccine production. All serials of vaccine shall be prepared from the first through the fifth passage from the Master Seed Virus.

(a) The Master Seed Virus shall meet the applicable general requirements prescribed in § 113.200.

(b) The immunogenicity of vaccine prepared from the Master Seed Virus in accordance with the Outline of Production shall be established. Vaccine used for this test shall be at the highest

passage from the Master Seed and prepared at the minimum preinactivation titer specified in the Outline of Production.

(1) Twenty-five canine distemper susceptible dogs (20 vaccinates and 5 controls) shall be used as test animals. Blood samples drawn from each dog shall be individually tested for neutralizing antibody against canine distemper to determine susceptibility. A constant virus-varying serum neutralization test in cell culture using 50 to 300 TCID₅₀ of virus shall be used. Dogs shall be considered susceptible if there is no neutralization at a 1:2 final serum dilution.

(i) The 20 dogs used as vaccinates shall be injected with one dose of vaccine by the method recommended on the label. If a second dose is recommended, the second dose shall be administered at the time specified on the label.

(ii) At least 14 days after the last inoculation, the vaccinates and controls shall each be challenged intracerebrally with canine distemper virus furnished or approved by the Animal and Plant Health Inspection Service and observed each day for 21 days.

(iii) If at least four of the five controls do not die and the survivor, if any, does not show clinical signs of canine distemper, the test is inconclusive and may be repeated.

(iv) If at least 19 of the 20 vaccinated do not survive without showing clinical signs of canine distemper during the observation period, the Master Seed Virus is unsatisfactory.

(c) *Test requirements for release.* Each serial shall meet the applicable general requirements prescribed in § 113.200 and the special requirements for safety and potency provided in this section.

(1) *Safety test.* The vaccinates used in the potency test in paragraph (c)(2) of this section shall be observed each day during the postvaccination observation period. If unfavorable reactions occur which are attributable to the vaccine, the serial is unsatisfactory. If unfavorable reactions occur which are not attributable to the vaccine, the test is inconclusive and may be repeated: *Provided*, That, if the test is not repeated, the serial is unsatisfactory.

(2) *Potency test—serum neutralization test.* Bulk or final container samples of completed product shall be tested for potency using five susceptible dogs (four vaccinates and one control) as the test animals. Blood samples drawn from each dog shall be individually tested for neutralizing antibody against canine distemper virus to determine susceptibility.

(i) A constant virus-varying serum neutralization test in tissue culture using 50 to 300 TCID₅₀ of virus shall be used. Dogs shall be considered susceptible if there is no neutralization at a 1:2 final serum dilution.

(ii) *Vaccination.* Each of the four vaccinates shall be injected as recommended on the label. If two doses are recommended, the second dose shall be administered at the time specified on the label. The dogs shall be observed each day for at least 14 days after the last inoculation.

(iii) *Serology.* At the end of the post vaccination observation period, a second blood sample shall be obtained from each of the five dogs and the serums shall be individually tested for neutralizing antibody against canine distemper virus in the same manner used to determine susceptibility.

(iv) *Interpretation of the serum neutralization test.* If the control has not remained seronegative at 1:2, the test is inconclusive and may be repeated. If at least three of the four vaccinates in a valid test have not developed titers based upon a final serum dilution of at least 1:50 and the remaining vaccinate has not developed a titer of at least 1:25, the serial is unsatisfactory except as provided in paragraphs (c)(2) (v) and (vi) of this section.

(v) *Virus challenge test.* If the results of a valid serum neutralization test are unsatisfactory, the vaccinates and the control may be challenged intracerebrally with a virulent canine distemper virus furnished or approved by the Animal and Plant Health Inspection Service and each animal observed each day for an additional 21 days.

(vi) *Interpretation of the virus challenge test.* For a serial to be satisfactory, all vaccinates must remain free from clinical signs of canine distemper while the control must die of canine distemper. If the control does not die of canine distemper, the test is inconclusive and may be repeated except, that if any of the vaccinates show signs or dies of canine distemper, the serial is unsatisfactory.

4. Section 113.202 is revised to read as follows:

§ 113.202 Canine Hepatitis and Canine Adenovirus Type 2 Vaccine, Killed Virus.

Canine Hepatitis and Canine Adenovirus Type 2 Vaccine, Killed Virus, shall be prepared from virus-bearing cell culture fluids. Only Master Seed Virus which has been established as pure, safe, and immunogenic shall be used for vaccine production. All serials of vaccine shall be prepared from the

first through the fifth passage from the Master Seed Virus.

(a) The Master Seed Virus shall meet the applicable requirements prescribed in § 113.200.

(b) Each lot of Master Seed Virus used for vaccine production shall be tested for immunogenicity by one or both of the following methods. Vaccine used for these tests shall be at the highest passage from the Master Seed and prepared at the minimum preinactivation titer specified in the Outline of Production.

(1) *Immunogenicity for canine hepatitis.* Twenty-five canine hepatitis susceptible dogs shall be used as test animals (20 vaccinates and 5 controls). Blood samples shall be drawn from these animals and individual serum samples tested. The dogs shall be considered susceptible if the results are negative at a 1:2 final serum dilution in a varying serum-constant virus neutralization test using 50 to 300 TCID₅₀ of canine adenovirus.

(i) The 20 dogs to be used as vaccinates shall be injected with one dose of vaccine and the remaining five dogs held as controls. If a second dose is recommended, the second dose shall be administered at the time specified on the label.

(ii) Not less than 14 days after the last inoculation, each vaccinate and control shall be challenged intravenously with virulent infectious canine hepatitis virus furnished or approved by the Animal and Plant Health Inspection Service and observed each day for 14 days.

(iii) If at least four of the five controls do not show severe clinical signs of infectious canine hepatitis, the test is inconclusive and may be repeated.

(iv) If at least 19 of the 20 vaccinates do not survive without showing clinical signs of infectious canine hepatitis during the observation period, the Master Seed Virus is unsatisfactory.

(2) *Immunogenicity for canine adenovirus type 2.* Thirty canine adenovirus type 2 susceptible dogs shall be used as test animals (20 vaccinates and 10 controls). Blood samples shall be drawn from these animals and individual serum samples tested. The dogs shall be considered susceptible if the results are negative at a 1:2 final serum dilution in a varying serum-constant virus neutralization test using 50 to 300 TCID₅₀ of canine adenovirus.

(i) The 20 dogs to be used as vaccinates shall be injected with one dose of vaccine and the remaining 10 dogs held as controls. If a second dose is recommended, the second dose shall be administered at the time specified on the label.

(ii) Not less than 14 days after the last inoculation, the vaccinates and the controls shall be challenged by exposure to a nebulized aerosol of virulent canine adenovirus type 2 furnished or approved by the Animal and Plant Health Inspection Service and observed each day for 14 days postchallenge. The rectal temperature of each animal shall be taken and the presence of respiratory or other clinical signs of canine adenovirus type 2 noted and recorded each day.

(iii) If at least 6 of 10 controls do not show clinical signs of canine adenovirus type 2 infection other than fever, the test is inconclusive and may be repeated.

(iv) If a significant difference in clinical signs in a valid test cannot be demonstrated between vaccinates and controls using a scoring system approved by the Animal and Plant Health Inspection Service, the Master Seed Virus is unsatisfactory.

(c) *Test requirements for release.* Each serial shall meet the applicable general requirements prescribed in § 113.200, the special requirements for safety provided in this section, and the applicable potency tests provided in this section.

(1) *Safety test.* The vaccinates used in the potency test in paragraph (c)(2) and/or (c)(3) of this section shall be observed each day during the postvaccination observation period. If unfavorable reactions occur which are attributable to the vaccine, the serial is unsatisfactory. If unfavorable reactions occur which are not attributable to the vaccine, the test is inconclusive and may be repeated: *Provided*, That, if not repeated, the serial is unsatisfactory.

(2) *Potency test for canine hepatitis—serum neutralization test.* Bulk or final container samples of completed product shall be tested for potency using at least five susceptible dogs (four vaccinates and one control) as the test animals. Blood samples drawn from each dog shall be individually tested for neutralizing antibody against canine adenovirus to determine susceptibility.

(i) A constant virus-varying serum neutralization test in tissue culture using 50 to 300 TCID₅₀ of virus shall be used. Dogs shall be considered susceptible if there is no neutralization at a 1:2 final serum dilution.

(ii) *Vaccination.* Each of the vaccinates shall be injected as recommended on the label. If two doses are recommended, the second dose shall be administered at the time specified on the label. The dogs shall be observed each day for at least 14 days after the last inoculation.

(iii) *Serology.* At the end of the postvaccination observation period, a

second blood sample shall be obtained from each of the dogs and the serums shall be individually tested for neutralizing antibody against canine adenovirus in the same manner used to determine susceptibility.

(iv) *Interpretation of the serum neutralization test.* If the control(s) has not remained seronegative at 1:2, the test is inconclusive and may be repeated. If at least 75 percent of the vaccinates in a valid test have not developed titers based upon final serum dilution of at least 1:10 and the remaining vaccinate(s) has not developed a titer of at least 1:2, the serial is unsatisfactory except as provided in paragraphs (c)(2) (v) and (vi) of this section.

(v) *Virus challenge test.* If the results of a valid serum neutralization test are unsatisfactory, the vaccinates and the control(s) may be challenged intravenously with a virulent canine hepatitis virus furnished or approved by the Animal and Plant Health Inspection Service and each animal observed each day for an additional 14 days.

(vi) *Interpretation of the virus challenge test.* For a serial to be satisfactory, all vaccinates must remain free of clinical signs of canine hepatitis while the control(s) must show severe clinical signs of canine hepatitis. If the control(s) does not show severe clinical signs of canine hepatitis, the test is inconclusive and may be repeated: *Provided*, That, if any of the vaccinates show signs or die of canine hepatitis, the serial is unsatisfactory.

(3) *Potency test for canine adenovirus type 2.* Bulk or final container samples of completed product shall be tested for potency using eight susceptible dogs (five vaccinates and three controls) as the test animals. Blood samples drawn from each dog shall be individually tested for neutralizing antibody against canine adenovirus to determine susceptibility.

(i) A constant virus-varying serum neutralization test in tissue culture using 50 to 300 TCID₅₀ of virus shall be used. Dogs shall be considered susceptible if there is no neutralization at a 1:2 final serum dilution.

(ii) *Vaccination.* Each of the five vaccinates shall be injected as recommended on the label. If two doses are recommended, the second dose shall be administered at the time specified on the label. The dogs shall be observed each day for at least 14 days after the last inoculation.

(iii) Not less than 14 days after the last inoculation, the vaccinates and the controls shall be challenged by exposure to a nebulized aerosol of virulent canine adenovirus type 2 furnished or

approved by the Animal and Plant Health Inspection Service and observed each day for 14 days postchallenge. The rectal temperature of each animal shall be taken and the presence of respiratory or other clinical signs of canine adenovirus type 2 noted and recorded each day.

(iv) If at least two of three controls do not show clinical signs of canine adenovirus type 2 other than fever, the test is inconclusive and may be repeated.

(v) If a significant difference in clinical signs cannot be demonstrated between vaccinates and controls using a scoring system approved by the Animal and Plant Health Inspection Service and prescribed in the Outline of Production, the serial is unsatisfactory.

5. Section 113.204 is amended by revising paragraphs (b)(2) and (b)(3) to read as follows:

§ 113.204 Mink Enteritis Vaccine, Killed Virus.

* * * * *

(b) * * *

(2) *Challenge.* At least 2 weeks after the last inoculation, the five vaccinates and the five controls shall be challenged with virulent mink enteritis virus and observed each day for 12 days. Fecal material shall be collected on one day between days 4–8 (inclusive) postchallenge from each test animal that remains free of enteric signs and tested for the presence of mink enteritis virus by cell culture with fluorescent antibody examination.

(3) *Interpretation.* A serial is satisfactory if at least 80 percent of the vaccinates remain free of enteric signs and do not shed virus in the feces, while at least 80 percent of the controls develop clinical signs of mink enteritis or shed virus in the feces. If at least 80 percent of the vaccinates remain free of enteric signs and do not shed virus in the feces, while less than 80 percent of the controls develop clinical signs of mink enteritis or shed virus in the feces, the test is considered inconclusive and may be repeated: *Provided*, That, if at least 80 percent of the vaccinates do not remain well and free of detectable virus in the feces, the serial is unsatisfactory.

6. Section 113.305 is revised to read as follows:

§ 113.305 Canine Hepatitis and Canine Adenovirus Type 2 Vaccine.

Canine Hepatitis Vaccine and Canine Adenovirus Type 2 Vaccine shall be prepared from virus-bearing cell culture fluids. Only Master Seed Virus which has been established as pure, safe, and immunogenic shall be used in preparing the production seed virus for vaccine

production. All serials shall be prepared from the first through the fifth passage from the Master Seed Virus.

(a) The Master Seed Virus shall meet the applicable requirements prescribed in § 113.300 except that the dog safety test prescribed in § 113.40(a) shall be conducted by the intravenous route.

(b) Each lot of Master Seed Virus used for vaccine production shall be tested for immunogenicity by one or both of the following methods:

(1) *Immunogenicity for canine hepatitis.* Twenty-five canine hepatitis susceptible dogs shall be used as test animals (20 vaccinates and 5 controls). Blood samples shall be drawn from these animals and individual serum samples tested. The dogs shall be considered susceptible if the results are negative at a 1:2 final serum dilution in a varying serum-constant virus neutralization test using 50 to 300 TCID₅₀ of canine adenovirus.

(i) A geometric mean titer of the dried vaccine produced from the highest passage of the Master Seed Virus shall be established before the immunogenicity test is conducted. The 20 dogs to be used as vaccinates shall be injected with a predetermined quantity of vaccine virus and the remaining five dogs held as uninjected controls. To confirm the dosage calculations, five replicate virus titrations shall be conducted on a sample of the vaccine virus dilution used.

(ii) Not less than 14 days postinjection, the vaccinates and the controls shall each be challenged intravenously with virulent infectious canine hepatitis virus furnished or approved by the Animal and Plant Health Inspection Service and observed each day for 14 days.

(A) If at least four of the five controls do not show severe clinical signs of canine hepatitis, the test is inconclusive and may be repeated.

(B) If at least 19 of the 20 vaccinates do not survive without showing clinical signs of infectious canine hepatitis during the observation period, the Master Seed Virus is unsatisfactory.

(iii) The Master Seed Virus shall be retested for immunogenicity for canine hepatitis in 3 years unless use of the lot previously tested is discontinued. Ten susceptible dogs (8 vaccinates and 2 controls) shall be used in the retest. Susceptibility shall be determined in the manner provided in paragraph (b)(1) of this section.

(A) Each vaccine shall be injected with a predetermined quantity of vaccine virus as provided in paragraph (b)(1)(i) of this section.

(B) At least 14 days postvaccination, a second serum sample shall be drawn from each dog and tested for neutralizing antibody to canine adenovirus in the same manner used to determine susceptibility.

(C) If the two controls have not remained seronegative at 1:2, the test is inconclusive and may be repeated.

(D) If at least six of the eight vaccinates in a valid test do not develop titers of at least 1:10 based upon final serum dilution, the Master Seed Virus is unsatisfactory except as provided in paragraph (b)(1)(iii)(E) of this section.

(E) If the results of a valid serum neutralization test are unsatisfactory, the vaccinates and the controls may be challenged as provided in paragraph (b)(1)(ii) of this section. A Master Seed is satisfactory if all vaccinates remain free of clinical signs of canine hepatitis, while both controls develop severe clinical signs of canine hepatitis. If both controls do not show severe clinical signs of canine hepatitis, the test is inconclusive and may be repeated: *Provided*, That, if any of the vaccinates show such signs, the Master Seed Virus is unsatisfactory.

(2) *Immunogenicity for canine adenovirus Type 2.* Thirty canine adenovirus type 2 susceptible dogs shall be used as test animals (20 vaccinates and 10 controls). Blood samples shall be drawn from these animals and individual serum samples tested. The dogs shall be considered susceptible if the results are negative at a 1:2 final serum dilution in a varying serum-constant virus neutralization test using 50 to 300 TCID₅₀ of canine adenovirus.

(i) A geometric mean titer of the dried vaccine produced from the highest passage of the Master Seed Virus shall be established before the immunogenicity test is conducted. The 20 dogs to be used as vaccinates shall be injected with a predetermined quantity of vaccine virus and the remaining 10 dogs held as uninjected controls. To confirm the dosage calculations, five replicate virus titrations shall be conducted on a sample of the vaccine virus dilution used.

(ii) Not less than 14 days postinjection, the vaccinates and the controls shall be challenged by exposure to a nebulized aerosol of virulent canine adenovirus type 2 furnished or approved by the Animal and Plant Health Inspection Service and observed each day for 14 days postchallenge. The rectal temperature of each animal shall be taken and the presence of respiratory or other clinical signs of canine adenovirus type 2 noted and recorded each day.

(A) If at least 6 of 10 controls do not show clinical signs of canine adenovirus type 2 infection other than fever, the test is inconclusive and may be repeated.

(B) If a significant difference in clinical signs in a valid test cannot be demonstrated between vaccinates and controls using a scoring system approved by the Animal and Plant Health Inspection Service, the Master Seed Virus is unsatisfactory.

(iii) the Master Seed Virus shall be retested for immunogenicity in 3 years unless use of the lot previously tested is discontinued. Either 10 vaccinates and 6 controls or 5 vaccinates and 3 controls shall be used in the retest.

(A) If less than 4 of 6 or 2 of 3 of the controls show clinical signs of canine adenovirus type 2 other than fever, the test is inconclusive and may be repeated.

(B) A significant difference in clinical signs shall be demonstrated between vaccinates and controls in a valid test as prescribed in paragraph (b)(2)(ii)(B) of this section.

(iv) an Outline of Production change shall be made before authorization for use of a new lot of Master Seed Virus shall be granted by the Animal and Plant Health Inspection Service.

(c) *Test requirements for release.* Each serial and subserial shall meet the requirements prescribed in § 113.300 and in this paragraph. Final container samples of completed product shall be tested. Any serial or subserial found unsatisfactory by a prescribed test shall not be released.

(1) *Virus titer requirements.* Final container samples of completed product shall be tested for virus titer using the titration method used in paragraph (b)(1)(i) and/or (b)(2)(i) of this section. To be eligible for release, each serial and each subserial shall have a virus titer sufficiently greater than the titer of vaccine virus used in the immunogenicity test(s) prescribed in paragraph (b) of this section to assure that when tested at any time within the expiration period, each serial and subserial shall have a virus titer of $10^{0.7}$ greater than that used in such immunogenicity test(s) but not less than $10^{2.5}$ TCID₅₀ dose. If both immunogenicity tests in paragraph (b) of this section are conducted and a different amount of virus is used in each test, the virus titer requirements shall be based on the higher of the two amounts.

7. Section 113.306 is revised to read as follows:

§ 113.306 Canine Distemper Vaccine.

Canine Distemper Vaccine shall be prepared from virus-bearing cell culture fluids or embryonated chicken eggs.

Only Master Seed Virus which has been established as pure, safe, and immunogenic shall be used for preparing the production seed virus for vaccine production. All serials of vaccine shall be prepared from the first through the fifth passage from the Master Seed Virus.

(a) *Master Seed Virus.* The Master Seed Virus shall meet the applicable requirements prescribed in § 113.300 and the requirements prescribed in this section.

(1) To detect ferret virulent canine distemper virus, each of five canine distemper susceptible ferrets shall be injected with a sample of the Master Seed Virus equivalent to the amount of virus to be used in one dog dose and observed each day for 21 days. If undesirable reactions are observed during the observation period, the lot of Master Seed is unsatisfactory.

(2) Master Seed Virus propagated in tissues or cells of avian origin shall be tested for pathogens by the chicken embryo test prescribed in § 113.37. If found unsatisfactory, the Master Seed Virus shall not be used.

(b) Each lot of Master Seed Virus used for vaccine production shall be tested for immunogenicity. The selected virus dose from the lot of Master Seed Virus shall be established as follows:

(1) Twenty-five canine distemper susceptible dogs shall be used as test animals (20 vaccinates and 5 controls). Blood samples shall be drawn from these animals and individual serum samples tested. The dogs shall be considered susceptible if the results are negative at a 1:2 final serum dilution in a varying serum-constant virus neutralization test using 50 to 300 TCID₅₀ of canine distemper virus.

(2) A geometric mean titer of the dried vaccine produced from the highest passage of the Master Seed Virus shall be established before the immunogenicity test is conducted. The 20 dogs used as vaccinates shall be injected with a predetermined quantity of vaccine virus and the remaining five dogs held as uninjected controls. To confirm the dosage calculations, five replicate virus titrations shall be conducted on a sample of the vaccine virus dilution used.

(3) At least 14 days post-injection, the vaccinates and the controls shall each be challenged intracerebrally with virulent canine distemper virus furnished or approved by the Animal and Plant Health Inspection Service and observed each day for 21 days.

(i) If at least four of the five controls do not die and the survivor, if any does not show clinical signs of canine

distemper the test is inconclusive and may be repeated.

(ii) If at least 19 of the 20 vaccinates do not survive without showing clinical signs of canine distemper during the observation period, the Master Seed Virus is unsatisfactory.

(4) The Master Seed Virus shall be retested for immunogenicity in 3 years unless use of the lot previously tested is discontinued. Ten susceptible dogs (8 vaccinates and 2 controls) shall be used in the retest. Susceptibility shall be determined in the manner provided in paragraph (b)(1) of this section.

(i) Each vaccine shall be injected with a predetermined quantity of vaccine virus as provided in paragraph (b)(2) of this section.

(ii) At least 14 days postvaccination, a second serum sample shall be drawn from each dog and tested for neutralizing antibody to canine distemper virus in the same manner used to determine susceptibility.

(iii) If the two controls have not remained seronegative at 1:2, the test is inconclusive and may be repeated.

(iv) If at least 6 of the 8 vaccinates in a valid test do not develop titers of at least 1:50 based upon final serum dilution, the Master Seed Virus is unsatisfactory, except as provided in paragraph (b)(4)(v) of this section.

(v) If the results of a valid serum neutralization test are unsatisfactory, the vaccinates and the controls may be challenged as provided in paragraph (b)(3) of this section. A Master Seed is satisfactory if all vaccinates remain free of clinical signs of canine distemper, while the two controls die with clinical signs of canine distemper. If the two controls do not die with clinical signs of canine distemper, the test is inconclusive and may be repeated:

Provided, That, if any of the vaccinates show such signs, the Master Seed Virus is unsatisfactory.

(5) An Outline of Production change shall be made before authorization for use of a new lot of Master Seed Virus shall be granted by the Animal and Plant Health Inspection Service.

(c) *Test requirements for release.* Except for § 113.300(a)(3)(ii), each serial and subserial shall meet the requirements prescribed in § 113.300 and in this paragraph. Final container samples of completed product shall be tested. Any serial or subserial found unsatisfactory by a prescribed test shall not be released.

(1) The test for pathogens prescribed in § 113.37 shall be conducted on each serial or one subserial of avian origin vaccine.

(2) *Virus titer requirements.* Final container samples of completed product

shall be tested for virus titer using the titration method used in paragraph (b)(2) of this section. To be eligible for release, each serial and subserial shall have a virus titer sufficiently greater than the titer of vaccine virus used in the immunogenicity test prescribed in paragraph (b) of this section to assure that when tested at any time within the expiration period, each serial and subserial shall have a virus titer of $10^{0.7}$ greater than that used in such immunogenicity test but not less than $10^{2.5}$ TCID₅₀ per dose.

§ 113.307 [Removed]

8. Section 113.307 is removed.

Done in Washington, DC, this 13th day of March 1995.

Terry L. Medley,

Acting Administrator, Animal and Plant Health Inspection Service.

[FR Doc. 95-6647 Filed 3-16-95; 8:45 am]

BILLING CODE 3410-34-M

DEPARTMENT OF TRANSPORTATION

Federal Aviation Administration

14 CFR Part 71

[Airspace Docket No. 93-ACE-02]

Amendment to Class E Airspace; Harvard, NE

AGENCY: Federal Aviation Administration [FAA], DOT.

ACTION: Final rule.

SUMMARY: This amendment modifies the Class E airspace area at Harvard, NE to accommodate a planned Very High Frequency Omnidirectional Range/Distance Measuring Equipment (VOR/DME) Standard Instrument Approach Procedure (SIAP) at the Harvard State Airport. This action will provide for additional controlled airspace necessary for the planned VOR/DME SIAP. It will also change the airport status from Visual Flight Rules (VFR) to Instrument Flight Rules (IFR).

EFFECTIVE DATE: 0901 UTC May 25, 1995.

FOR FURTHER INFORMATION CONTACT: Kathy Randolph, Air Traffic Operations Branch, ACE-530c, Federal Aviation Administration, 6021 E. 12th St., Kansas City, MO, 64106; telephone (816) 426-3408.

SUPPLEMENTARY INFORMATION:

History

On January 7, 1994, the FAA proposed to amend part 71 of the Federal Aviation Regulations (14 CFR part 71) by modifying the Class E

airspace area at Harvard, NE (59 FR 3032). The proposed action would provide additional controlled airspace to accommodate a VOR/DME SIAP to Runway 35 at the Harvard State Airport.

Interested parties were invited to participate in this rulemaking proceeding by submitting written comments on the proposal to the FAA. No comments objecting to the proposal were received. Class E airspace areas extending from 700 feet or more above the surface of the earth are published in paragraphs 6005 of FAA order 7400.9B, dated July 8, 1994, and effective September 16, 1994, which is incorporated by reference in 14 CFR 71.1. The Class E airspace designation listed in this document will be published subsequently in the Order.

The Rule

This amendment to part 71 of the Federal Aviation Regulations (14 CFR part 71) amends the Class E airspace area at Harvard, NE, by providing additional controlled airspace for aircraft executing the VOR/DME runway 35 SIAP at the Harvard State Airport.

The FAA has determined that this regulation only involves an established body of technical regulations for which frequent and routine amendments are necessary to keep them operationally current. Therefore, this regulation—(1) is not a “significant regulatory action” under Executive Order 12866; (2) is not a “significant rule” under DOT Regulator Policies and Procedures (44 FR 11034; February 26, 1979); and (3) does not warrant preparation of a Regulatory Evaluation as the anticipated impact is so minimal. Since this is a routine matter that will only affect air traffic procedures and air navigation, it is certified that this rule will not have a significant economic impact on a substantial number of small entities under the criteria of the Regulatory Flexibility Act.

List of Subjects in 14 CFR Part 71

Aviation, Incorporation by reference, Navigation (air).

Adoption of the Amendment

In consideration of the foregoing, the Federal Aviation Administration amends 14 CFR part 71 as follows:

PART 71—[AMENDED]

1. The authority citation for part 71 continues to read as follows:

Authority: 49 U.S.C. app. 1348(a), 1510; E.O. 10854, 24 FR 9565, 3 CFR, 1959-1963 Comp., p. 389; 49 U.S.C. 106(g); 14 CFR 11.69.

§ 71.1 [Amended]

2. The incorporation by reference in 14 CFR 71.1 of Federal Aviation Administration Order 7400.9B, Airspace Designations and Reporting Points, dated July 18, 1994 and effective September 16, 1994, is amended as follows:

Paragraph 6005 Class E airspace areas extending from 700 feet or more above the surface of the earth.

* * * * *

ACE NE E5 Harvard, NE [Revised]

Harvard State Airport, NE
(Lat. 40°39'15" N, long. 98°04'31" W)

That airspace extending upward from 700 feet above the surface within 6.4-mile radius of the Harvard State Airport and within 2 miles each side of the 180° bearing of the Harvard State Airport extending from the 6.4-mile radius to 10 miles south of the airport.

* * * * *

Issued in Kansas City, MO on February 21, 1995.

Clarence E. Newbern,

Manager, Air Traffic Division, Central Region.

[FR Doc. 95-6685 Filed 3-16-95; 8:45 am]

BILLING CODE 4910-13-M

14 CFR Part 97

[Docket No. 28135; Amdt. No. 1654]

Standard Instrument Approach Procedures; Miscellaneous Amendments

AGENCY: Federal Aviation Administration (FAA), DOT.

ACTION: Final rule.

SUMMARY: This amendment establishes, amends, suspends, or revokes Standard Instrument Approach Procedures (SIAPs) for operations at certain airports. These regulatory actions are needed because of the adoption of new or revised criteria, or because of changes occurring in the National Airspace System, such as the commissioning of new navigational facilities, addition of new obstacles, or changes in air traffic requirements. These changes are designed to provide safe and efficient use of the navigable airspace and to promote safe flight operations under instrument flight rules at the affected airports.

DATES: An effective date for each SIAP is specified in the amendatory provisions.

Incorporation by reference approved by the Director of the **Federal Register** on December 31, 1980, and reapproved as of January 1, 1982.

ADDRESSES: Availability of matters incorporated by reference in the amendment is as follows: