



October 15, 2012

CENTER FOR VETERINARY BIOLOGICS NOTICE NO. 12-21

**United States
Department of
Agriculture**

Animal and Plant
Health Inspection
Service

Veterinary Services

Center for Veterinary
Biologics

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TO: Biologics Licensees, Permittees, and Applicants
Directors, Center for Veterinary Biologics
Veterinary Services Leadership Team

FROM: Richard E. Hill, Jr. /s/ *Richard E. Hill, Jr.*
Director
Center for Veterinary Biologics

SUBJECT: Conducting Dilution of Preservative Studies for Live Bacterial Vaccines

I. PURPOSE

The purpose of this Notice is to inform interested parties regarding the Center for Veterinary Biologics' (CVB) interpretation of when dilution of preservative data is required for live bacterial vaccines.

II. BACKGROUND

Title 9, Code of Federal Regulations (9 CFR), Part 113.27(b) describes a direct inoculation sterility test to be conducted on live bacterial vaccines in order to detect extraneous bacterial or fungal contamination. With regard to the media volume to be used, 9 CFR 113.27(b) provides a default media volume of 40 mL, but indicates that larger volumes may be needed when 9 CFR 113.25(d) testing indicates product interference. The CVB uses a screening test for dilution of preservative/ product interference when conducting confirmatory sterility testing of products. The screening test was first announced in CVB Notice 09-02, *Dilution of Preservative Screening for Broth-Based Sterility Tests*.

While many live bacterial vaccines lack antibiotics in their final form, a small number of them have antibiotics added which could cause product interference for the detection of bacterial or fungal contamination. The intent of this Notice is to ensure the dilution of preservative requirement in 9 CFR 113.25(d) and 9 CFR 113.27 is consistently applied to live bacterial products.

III. ACTION (or POLICY)

For live bacterial products with antibiotics added, dilution of preservative studies are required in accordance with 9 CFR 113.25(d). For live bacterial products that lack antibiotics at formulation, dilution of preservative studies will not be required at this time. However, in an effort to evaluate this situation, the CVB laboratory has begun



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inoculating an extra media vessel for all live bacterial products utilizing the purity test in accordance with 9 CFR 113.27(b). If a lack of indicator organism growth in the extra tube is seen in check or confirmatory testing, this result will be evaluated utilizing a complete dilution of preservative study in accordance with SAM 903.

For oral products, not covered by 9 CFR 113.27(e), an exemption to dilution of preservative may be supplied upon request. The default amount of media would then be used.

Therefore, each Outline of Production should include information on the dilution of preservative status with the information in Section V regarding the sterility test. The date that the data were accepted, the date an exemption was obtained, or the statement that the product lacks antibiotics, should be included.

IV. IMPLEMENTATION/ APPLICABILITY

The above policies will be implemented 30 days from the publication date of this Notice. As each licensee/permittee is required to review their Outlines of Production at least annually for accuracy and sufficiency in accordance with 9 CFR 114.8(d), we would anticipate that within 18 months of publication, all products and their Outlines will be in compliance with the above. Manufacturers that require additional time to achieve compliance should contact their Reviewer with the request for additional time.