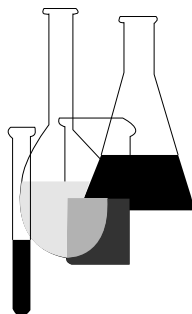




Microbial Pesticide Test Guidelines

OPPTS 885.5400 Expression in a Marine or Estuarine Environment



INTRODUCTION

This guideline is one of a series of test guidelines that have been developed by the Office of Prevention, Pesticides and Toxic Substances, United States Environmental Protection Agency for use in the testing of pesticides and toxic substances, and the development of test data that must be submitted to the Agency for review under Federal regulations.

The Office of Prevention, Pesticides and Toxic Substances (OPPTS) has developed this guideline through a process of harmonization that blended the testing guidance and requirements that existed in the Office of Pollution Prevention and Toxics (OPPT) and appeared in Title 40, Chapter I, Subchapter R of the Code of Federal Regulations (CFR), the Office of Pesticide Programs (OPP) which appeared in publications of the National Technical Information Service (NTIS) and the guidelines published by the Organization for Economic Cooperation and Development (OECD).

The purpose of harmonizing these guidelines into a single set of OPPTS guidelines is to minimize variations among the testing procedures that must be performed to meet the data requirements of the U. S. Environmental Protection Agency under the Toxic Substances Control Act (15 U.S.C. 2601) and the Federal Insecticide, Fungicide and Rodenticide Act (7 U.S.C. 136, *et seq.*).

Final Guideline Release: This guideline is available from the U.S. Government Printing Office, Washington, DC 20402 on *The Federal Bulletin Board*. By modem dial 202-512-1387, telnet and ftp: fedbbs.access.gpo.gov (IP 162.140.64.19), internet: <http://fedbbs.access.gpo.gov>, or call 202-512-0132 for disks or paper copies. This guideline is also available electronically in ASCII and PDF (portable document format) from the EPA Public Access Gopher (gopher.epa.gov) under the heading "Environmental Test Methods and Guidelines."

OPPTS 885.5400 Expression in a marine or estuarine environment.

(a) **Scope—(1) Applicability.** This guideline is intended to meet testing requirements of the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) (7 U.S.C. 136, *et seq.*).

(2) **Background.** The source material used in developing this harmonized OPPTS test guideline is OPP guideline 155A-12.

(b) **Test standards.** This guideline is to be used with OPPTS 885.5000.

(1) Tests shall be conducted in a simulated marine or estuarine environment (e.g., aquarium with bottom sediment) to determine whether the microbial pest control agent (MPCA) is able to survive, persist, and/or replicate in a marine or estuarine environment consisting of seawater or brackish water and bottom sediment representative of the proposed use site. The following parameters should be varied to determine their effect on the survival and growth of the MPCA population:

(i) Temperature.

(ii) pH.

(iii) Nutrients.

(iv) Salinity.

(v) Sunlight.

(vi) Oxygen content.

(vii) Turbulence.

(2) The values selected for each parameter listed should be selected to approximate the conditions expected at the intended use site.

(3) Specialized laboratory studies designed to determine the MPCA's growth requirements (e.g., temperature, pH, sunlight, and oxygen) may supplement the study described in paragraph (d)(1)(i) of OPPTS 885.5000. Specialized laboratory studies may demonstrate that the MPCA will be unable to survive and persist in a marine or estuarine environment. In such instances, the Agency will consider studies on an individual basis in lieu of the study described in this guideline.

(c) **Test substance.** A typical end-use product or the technical grade of the active ingredient shall be tested.

(d) **Test duration.** Data to establish a population decline curve should be collected at intervals until two half-life determinations have been made or until data establish that the microbial agent population is able to main-

tain itself in a marine or estuarine environment at or above the level present immediately after test initiation.

(e) **Reporting and evaluation of data.** The reporting and evaluation provisions are the same as those set forth in OPPTS 885.5000. In addition, any changes in morphology of the microorganism in response in to changes in salinity should be reported.

(f) **Tier progression.** If results of this study and use pattern information indicate that the MPCA is likely to enter and is able to persist in a marine or estuarine environment such that the susceptible nontarget organisms tested in Tier I is likely to be exposed, the appropriate testing in Tier III (OPPTS 885.4450 through 885.4850) is required as specified by 40 CFR 158.740.

(g) **References.** The following references contain useful for developing acceptable protocols:

(1) Anthony, D.W. et al. Field tests with *Nosema algerae* Vavra and Undeen (Microsporida, Nosematidae) against *Anopheles albinamus* Wiedemann in Panama. *Miscellaneous Publications of the Entomological Society of America* 11:17–28 (1978).

(2) Brand, R.J. et al. Methods for assessing field persistence of *Bacillus thuringiensis* spores. *Journal of Invertebrate Pathology* 25:199–208 (1975).

(3) EPA. Impact of the use of microorganisms on the aquatic environment. EPA publication 660–3–75–001. Technical Publications Office, Environmental Protection Agency, National Environmental Research Center, Corvallis, OR 97330 (1975).

(4) Hostetter, D.L. et al. Persistence of formulations of *Bacillus thuringiensis* spores and crystals on eastern red cedar foliage in Missouri. *Journal of the Kansas Entomological Society* 48:189–193 (1975).

(5) Ignoffo, C.N. et al. Stability of *Bacillus thuringiensis* and *Baculovirus heliothis* on soybean foliage. *Environmental Entomology* 3:117–119 (1974).

(6) Kaya, H.K. Persistence of spores of *Pleistophora schuber* (Onidospora: Microsporida) in the field and their application in microbial control. *Journal of Invertebrate Pathology* 26:329–332 (1975).

(7) Pinnock, D.E. et al. The field persistence of *Bacillus thuringiensis* spores. *Journal of Invertebrate Pathology* 18:405–411 (1971).

(8) Sinclair, J.L. and M. Alexander. Role of resistance to starvation in bacterial survival in sewage and lake water. *Applied Environmental Microbiology* 48:410–415 (1984).

(9) Young, S.Y. Pre- and posttreatment assessment of virus levels. Selected papers from EPA-USDA Working Symposium. M. Summers, R. Engler, L. Falcon, and P. Vail (eds.) American Society of Microbiology (1975).