Biopesticides and

Pollution Prevention Division

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Microbial Pesticides

78 registered

- Bacteria (39)
  - 17 *Bacillus thuringiensis* subspecies
  - 10 other *Bacillus* species
  - 10 *Pseudomonas* species
  - 2 *Agrobacterium* isolates
- Fungi (29)

  - Insect pathogens, anti-fungals & mycoherbicides
- Insect, bacterial & plant viruses (7)
- Yeast (2)
- Protozoa (1)

Microbial Data Product Analysis and Human Health Data Requirements

Human Health Tier I

Toxicity/Pathogenicity

& Toxicity studies

Product Analysis

If significant toxicity or persistence is seen in Tier I testing
Also for toxic metabolites

Human Health Tier II

Acute & subchronic tox

If use patterns might result in residues
and
If testing or analysis indicates potential for significant health effects or mammalian toxin

If significant toxicity/infectivity seen at Tier II or to assess suspected human pathogens or viruses

Residue Data

Human Health Tier III

Additional Toxicity testing and/or Toxicity/infectivity analysis

Microbial Product Analysis Data Requirements		
Guideline Number	**Data Requirement**	
Product Chemistry and Composition		
885.1100	Product Identity	Relation to pathogens ecological niche, strain origin
885.1200	Manufacturing process	Quality control, seed cultures
	Deposition of a sample in a nationally recognized culture collection	
885.1300	Discussion of formation of unintentional ingredients	Potential for toxin production, microbial contaminants
Analysis and Certified Limits		
885.1400	Analysis of samples	Multiple batch analysis
885.1500	Certification of limits	
Physical and Chemical Characteristics		
830.6302	Color	
830.6303	Physical state	
830.6304	Odor	
830.6313	Stability to normal and elevated temperatures, metals and metal ions	
830.6317	Storage stability	Important for shelf life claims
830.6319	Miscibility	Only for emulsifiable liquid forms
830.6320	Corrosion Characteristics	Only when packaged in susceptible containers
830.7000	pH	
830.7100	Viscosity	Only for liquid forms
830.7300	Density/relative density/bulk density (specific gravity)	

Guideline Number	Data Requirement	Test Notes
Tier I		
885.3050	Acute oral toxicity/pathogenicity	Can be combined with the acute oral using EP or MP
885.3150	Acute pulmonary tox/path	--
885.3200	Acute injection tox/path intravenous or intraperitoneal	Not required for viruses ip for larger microbes
885.3400	Hypersensitivity incidents	Reporting requirement
885.3500	Cell culture	For viruses only
870.1100	Acute oral toxicity	Primarily for analyzing inerts
870.1200	Acute dermal toxicity	Primarily for analyzing inerts
870.1300	Acute inhalation toxicity	Primarily for analyzing inerts - if inhalable
870.2400	Acute eye irritation	Primarily for analyzing inerts
870.2500	Primary dermal irritation	Primarily for analyzing inerts

Changes to Microbial Product Analysis and Human Health Data Requirements

New data requirements (at Tier III):

- Infectivity/pathogenicity analysis and immunotoxicity (viruses)

Revised data requirements:

- Test notes better describe when studies are needed
- Listed studies required for Physical/chemical properties and Residues
- Samples to be submitted to a culture collection instead of Repository
- Dermal tox/pathogenicity study replaced by acute dermal toxicity

Deleted data requirements:

- Hypersensitivity studies, i.c. tox/path, Immune response; Mutagenicity, Teratogenicity, Virulence enhancement, Chronic feeding, and Tier II ip/ic tox/path, and Primary eye & dermal

Initial Assessment

(pre-submission and prior to full review)

- Is the taxonomic description accurate?
- Is it related to any well-known microbials?
  - Do any have adverse clinical effects?
  - Are any known to product toxic metabolites?
  - Are any commonly used in food?
  - Are any ubiquitous with no reported adverse effects?
- Do we need all the normally-required data?
- Do we need unique data to check for potential problems indicated by the initial assessment?

Toxicity/Pathogenicity Testing

- 3 animals/sex at each interim & final sacrifice
- Continue until a pattern of clearance is seen
- Maximum hazard testing
  - 10^8 units via oral & pulmonary routes
  - 10^7 units if injected
    - i.v. bacteria, viruses
    - i.p. for fungi, protozoa)

Toxicity/Pathogenicity Testing

- Clinical examination of animals
  - mortality
- Body weights
- Necropsy
- Enumeration of microbial active ingredient
  - kidney, brain, liver, lung, spleen, blood, etc.

Microbial Pesticides Toxicology Tier II & III

Tier II - routes of exposure corresponding to Tier I trigger effects		
885.3550	Acute toxicology	When Tier I oral, injection or pulmonary show toxicity without pathogenicity/infectivity
885.3600	Subchronic toxicity/pathogenicity	When infectivity/unusual persistence is seen without toxicity or pathogenicity at Tier I -or to test toxins or contaminants
Tier III	Test standards may have to be modified depending on the characteristics of the microorganism. Consult with the agency.	
885.3650	Reproductive fertility effects	If one or more of criteria 1, 2, 3, or 4 (below) are met
870.4200	Carcinogenicity	For products that may contain carcinogenic viruses or for microbial components that are identified as having significant toxicity in Tier II testing.
870.7800	Immunotoxicity	For products that may contain viruses that can interact in an adverse manner with components of the mammalian immune system
885.3000	Infectivity/pathogenicity analysis	To analyze pathogenic characteristics if seen in lower Tiers, or if related to known human pathogens

1. Significant infectivity was observed in test animals in the Tier II subchronic study and in which no significant signs of toxicity or pathogenicity were observed.
2. The microbial is a virus which can persist or replicate in mammalian cell culture lines.
3. The microbial is not amenable to thorough taxonomic classification, and is related to organisms known to be parasitic for mammalian cells.
4. The microbial preparation is not well purified, and may contain contaminants which are parasitic for mammals.

Risk Assessment

- Hazard
  - Toxicity, Infectivity, Pathogenicity potential
- Exposure
  - Population dynamics, infectivity, residues
  - Used less for microbial pesticides
    - Difficult analysis for microbials that multiply in the environment
      - Extremely variable test results
      - Environmental populations are very competitive
      - Generally drop to background levels
    - Microbial toxins may need analysis
- Risk
  - Generally try to use the Hazard Assessment to rule out potential human pathogens or ones that produce significant human toxins
  - And/or use risk mitigation involving use limitations

Microbial Pesticides
Regulatory Challenges

- Killed microbials
  - Case-by-case analysis – need microbial expertise to assure it is sufficiently killed, but may need conventional pesticide analysis of the toxins
- Microbial mixtures
  - Need to identify each active as a separate a.i., but may test the consortium for testing
- Compost / Manure tea
  - Would require extensive batch monitoring to assure no non-target or human pathogens were present
- Classical Biocontrol
  - Still needs registration
- Microbial Rodenticides, e.g. BioRat
  - Normally not eligible for registration – but?