exposure (*i.e.*, nervous system malformations or neuropathy, brain weight changes in offspring, functional or behavioral changes in the offspring).

iii. The pesticide elicits a causative association between exposures and adverse neurological effects in human epidemiological studies.

iv. The pesticide evokes a mechanism that is associated with adverse effects on the development of the nervous system (*i.e.*, structure-activity-relationship (SAR) to known neurotoxicants, altered neuroreceptor or neurotransmitter responses).

31. To facilitate the weight-of-evidence determination for the pesticide's mutagenicity, in addition to those specifically listed in this table, the Agency requires submission of other mutagenicity test results that may have been performed. A reference list of all studies and papers known to the applicant concerning the mutagenicity of the test chemical must be submitted with the required studies.

32. Due to the nature of antimicrobials, if testing with bacterial strains has not been conducted, then testing using a mammalian cell assay such as the mouse lymphoma TK ± assay is preferred. If reverse mutation assay testing with bacterial strains has already been conducted, and the testing was conducted at levels that did not cause toxicity to the bacterial strains tested, then the applicant may submit the study to fulfill this data requirement.

33. For the *in vitro* mammalian gene mutation study, there is a choice of assays using either mouse lymphoma L5178Y cell thymidine kinase (tk) gene locus, maximizing assay conditions for small colony expression and detection; Chinese hamster ovary (CHO) or Chinese hamster lung fibroblast (v79) cells, hypoxanthine-guanine phosphoribosyl transferase (hgprt) gene locus, accompanied by an appropriate *in vitro* test for clastogenicity; or CHO cells strains AS52, xanthine-guanine phosphoribosyl transferase (xprt) gene locus.

34. There is a choice of assays, but the micronucleus rodent bone marrow assay is preferred; the rodent bone marrow assays using metaphase analysis (aberrations) are acceptable.

35. Data are required when chronic toxicity or carcinogenicity studies are also required.

36. Data is required if the product label directs that it be applied to domestic animals, such as cats, dogs, cattle, pigs, and horses.

37. In the absence of dermal absorption data or a repeated dose dermal toxicity study, the assumption of 100 percent dermal absorption would be used in a risk assessment to determine if a dermal penetration study is required, and to identify the doses and duration of exposure for which dermal absorption is to be quantified.

38. Required for nonfood uses, if oral exposure could occur.

39. Data may be required if significant adverse effects are seen in available toxicology studies and these effects can be further elucidated by metabolism and pharmacokinetics studies.

§ 158.2240 Nontarget organisms.

(a) *General*. Subpart B of this part and § 158.2201 describe how to use the table in paragraph (c) of this section to determine the terrestrial and aquatic nontarget organisms data requirements for a particular antimicrobial pesticide product. Notes that apply to an individual test, including specific conditions, qualifications, or exceptions are listed in paragraph (d) of this section.

(1) Terrestrial and aquatic nontarget organism data are required to support the registration of most end-use and manufacturing-use antimicrobial products.

(2) Data are generally not required to support end-use products of a gas, highly volatile liquid, highly reactive solid, or a highly corrosive material.

(3) Data on transformation/degradation products or leachate residues of the parent compound are also required to support registration, if the transformation/degradation/degradation products or leachate residues meet one of the following criteria:

(i) More toxic, persistent, or bioaccumulative than the parent;

(ii) Have been shown to cause adverse effects in mammalian or aquatic reproductive studies; or

(iii) The moiety of concern (*i.e.*, functional group in the parent chemical molecule that imparts adverse effects) remains intact.

(4) If an antimicrobial may be applied to a field crop, horticultural crop, or turf, then the data requirements in § 158.630 apply.

(5) For the purpose of determining data requirements, the all other use patterns category includes the following use patterns:

(i) Agricultural premises and equipment.

(ii) Food-handling/storage establishments, premises, and equipment.

(iii) Commercial, institutional and industrial premises and equipment.

(iv) Residential and public access premises.

(v) Medical premises and equipment.

(vi) Human drinking water systems.

(vii) Materials preservatives.

(viii) Swimming pools.

(b) *Key*. MP = Manufacturing use product; EP = End-use product; R = Required; CR = Conditionally required; NR = Not required; TGAI = Technical grade of the active ingredient; TEP = Typical end-use product; PAIRA = Pure active ingredient radiolabeled; a.i. = active ingredient.

(c) *Antimicrobial nontarget organism data requirements table*. The following table shows the data requirements for nontarget organisms. The test notes appear in paragraph (d) of this section.

TABLE—ANTIMICROBIAL NONTARGET ORGANISM DATA REQUIREMENTS

Tier One Testing									
850.2100	Acute avian oral toxicity.	R	R	R	R	R	TGAI	TGAI	1
850.1010	Acute freshwater invertebrates toxicity.	R	R	R	R	R	TGAI	TGAI	2
850.1075	Acute freshwater fish toxicity.	R	R	R	R	R	TGAI	TGAI	3
Higher Tier Testing									
Avian Testing									
850.2200	Avian dietary toxicity	CR	CR	CR	CR	CR	TGAI	TGAI	4
850.2300	Avian reproduction	CR	CR	CR	CR	CR	TGAI	TGAI	1, 6
Aquatic Organisms Testing									
850.1010	Acute freshwater invertebrates toxicity.	CR	CR	CR	CR	CR		TEP	2, 5, 7
850.1075	Acute freshwater fish toxicity.	CR	CR	CR	CR	CR		TEP	3, 5, 7
850.1025 850.1035 850.1045	Acute estuarine and marine organisms toxicity.	CR	R	CR	CR	CR	TGAI	TGAI	8, 9
850.1055 850.1075	Acute estuarine and marine organisms toxicity.	CR	CR	CR	CR	CR		TEP	5, 7, 8
850.1400	Fish early-life stage	R	R	R	R	R	TGAI	TGAI	10
850.1300 850.1350	Aquatic invertebrate life-cycle.	R	R	R	R	R	TGAI	TGAI	10
850.1500	Fish life-cycle	CR	CR	CR	CR	CR	TGAI	TGAI	11, 12
850.1710 850.1730 850.1850	Aquatic organisms, bioavailability, biomagnification, toxicity tests.	CR	CR	CR	CR	CR	TGAI, PAI, degradate.	TGAI, PAI, degradate.	13
850.1950	Simulated or actual field testing for aquatic organisms.	CR	CR	CR	CR	CR	TEP	TEP	14, 15, 16

Sediment Testing									
850.1735	Whole sediment; acute freshwater invertebrates.	CR	CR	CR	CR	CR	TGAI	TGAI	15, 17
850.1740	Whole sediment; acute marine invertebrates.	CR	CR	CR	CR	CR	TGAI	TGAI	15, 17, 19
None	Whole sediment; chronic invertebrates fresh-water and marine.	CR	CR	CR	CR	CR	TGAI	TGAI	15, 18, 19
Insect Pollinator Testing									
850.3020	Honeybee acute contact.	NR	NR	R	NR	CR	TGAI	TGAI	20
850.3030	Toxicity of residues to honeybees.	NR	NR	R	NR	CR	TGAI	TEP or treated wood.	20, 21

(d) *Test notes.* The following test notes apply to the data requirements in the table to paragraph (c) of this section:

1. For industrial processes and water systems, antifoulant paints and coatings, wood preservatives, and aquatic areas, data are required for two avian species: one waterfowl species and one upland game bird species. For the all other use patterns category (as specified in §158.2240(a)(5)), data are required for one avian species.

2. Data are required on one freshwater aquatic invertebrate species.

3. For the industrial processes and water systems, antifoulant paints and coatings, wood preservatives, and aquatic use pattern areas, data are required on two species of fish, one cold water species and one warm water species. For the all other use patterns category (as specified in §158.2240(a)(5)), data are required on one species of fish, either one cold water species or one warm water species. Testing on a second species is required if the active ingredient or principal transformation products are stable in the environment and the LC_{50} in the first species is less than or equal to 1 ppm or 1 mg/L.

4. Data are required on one avian species, either one waterfowl species or one upland game bird species, if the avian acute oral LD_{50} (TGAI testing) is less than or equal to 100 mg/a.i./kg and a.i. residues or its principal transformation products are likely to occur in avian feed items. Data on the second species are required if the avian dietary LC_{50} in the first species tested is less than or equal to 500 ppm a.i. in the diet.

5. If TEP testing cannot be conducted due to the physical characteristics of the test substance (for example, a paint), then the applicant should request a waiver.

6. Data are required if one or more of the following criteria are met:

i. Birds may be subjected to repeated or continued exposure to the pesticide or any of its transformation products, especially preceding or during the breeding season.

ii. The pesticide or any of its major metabolites or degradation products are stable in the environment to the extent that a potentially toxic amount may persist in avian feed.

iii. The pesticide or any of its major metabolites or degradation products are stored or accumulated in plant or animal tissues, as indicated by the octanol/water partition coefficient (K_{ow} is greater than or equal to 1,000), accumulation studies, metabolic release and retention studies, or as indicated by structural similarity to known bioaccumulative chemicals.

iv. Any other information, such as that derived from mammalian reproduction studies, indicates that reproduction in terrestrial vertebrates may be adversely affected by the anticipated use of the pesticide product.

7. TEP testing is required for any product which meets one or more of the following conditions:

i. When based on deterministic modeling results: If the Estimated Environmental Concentration (EEC) in the aquatic environment is equal to or greater than one-half the LC_{50}/EC_{50} of the TGAI.

ii. When based on probabilistic modeling results: If the estimated 10th percentile 7Q10 Surface Water Concentration exceeds the acute concentration of concern (*i.e.*, one-half the LC_{50}/EC_{50}).

iii. If an ingredient in the end-use product other than the active ingredient is expected to enhance the toxicity of the active ingredient or to cause toxicity to aquatic organisms.

iv. The end-use antimicrobial product will be applied directly into an aquatic environment.

8. Data are required on one estuarine/marine mollusk, one other estuarine/marine invertebrate, and one estuarine/marine fish species.

9. For the all other use patterns category (as specified in §158.2240(a)(5)), industrial processes and water systems, wood preservatives, and aquatic areas, data are required if the pesticide residues from the parent compound and/or transformation products are likely to enter the estuarine/marine environment.

10. Testing must be conducted with the most sensitive organism (either freshwater or estuarine/marine vertebrates, or freshwater or estuarine/marine invertebrates), as determined from the results of the acute toxicity

tests (acute EC_{50} freshwater invertebrates; acute LC_{50}/EC_{50} estuarine and marine organisms; acute freshwater fish LC_{50}).

11. Data are required on estuarine/marine species if the product is intended for direct application to the estuarine or marine environment, or the product is expected to enter this environment in significant concentrations because of its expected use or mobility patterns.

12. Data are required on freshwater species if the end-use product is intended to be applied directly to water, or is expected to be transported to water from the intended use site, and when one or more of the following conditions apply:

i. When based on deterministic modeling results: If the Estimated Environmental Concentration (EEC) in water is equal to or greater than 0.1 of the no-observed-adverse-effect concentration or no-observed-adverse-effect level (NOAEC/NOAEL) in the fish early-life stage or invertebrate life cycle tests.

ii. When based on probabilistic modeling results: If the estimated 10th percentile 7Q10 Surface Water Concentration based on probabilistic modeling exceeds for 20 days or more the chronic concentration of concern (*i.e.*, one-tenth the NOAEC or NOAEL) determined in the fish early-life stage or invertebrate life cycle tests.

iii. If studies of other organisms indicate that the reproductive physiology of fish may be affected.

13. Not required when:

i. The octanol/water partition coefficients of the pesticide and its major degradates are less than 1,000;

ii. There are no potential exposures to fish and other nontarget aquatic organisms; or

iii. The hydrolytic half-life is less than 5 days at pH 5, 7, and 9.

14. Environmental chemistry methods used to generate data associated with this study must include results of a successful confirmatory method trial by an independent laboratory. Test standards and procedures for independent laboratory validation are available as addenda to the guideline for this test requirement.

15. Protocols must be approved by the Agency prior to the initiation of the study.

16. Data are required if the intended use pattern, and the physical/chemical properties and environmental fate characteristics of the antimicrobial indicate significant potential exposure, and, based on the results of the acute and chronic aquatic organism testing, significant impairment of nontarget aquatic organisms could result.

17. Data are required if the half-life of the pesticide in the sediment is equal to or less than 10 days in either the aerobic soil or aquatic metabolism studies, and if one or more of the following conditions are met:

i. The soil partition coefficient (K_d) is equal to or greater than 50 L/kg.

ii. The log K_{ow} is equal to or greater than 3.

iii. The K_{oc} is equal to or greater than 1,000.

18. Data are required if the EEC in sediment is greater than 0.1 of the acute LC_{50}/EC_{50} values and if one or more of the following conditions are met:

i. The soil partition coefficient (K_d) is equal to or greater than 50 L/kg.

ii. The log K_{ow} is equal to or greater than 3.

iii. The K_{oc} is equal to or greater than 1,000.

19. Sediment testing with estuarine/marine test species is required if the product is intended for direct application to the estuarine or marine environment or the product is expected to enter this environment in significant concentrations either by runoff or erosion, because of its expected use or mobility pattern.

20. For the all other use patterns category (as specified in § 158.2240(a)(5)), data are required only for beehive applications when the beehive (empty or occupied) may be treated.

21. A study similar to ''Honey Bee Toxicity of Residues on Foliage'' is required using treated wood instead of the foliage. Protocols must be approved by the Agency prior to the initiation of the study.

§ 158.2250 Nontarget plant protection.

(a) Subpart B of this part and § 158.2201 describe how to use the table