CYCLOHEXIMIDE

Task 2: Topical Discussions

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Final Report

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CYCLOHEXIMIDE

Task 2

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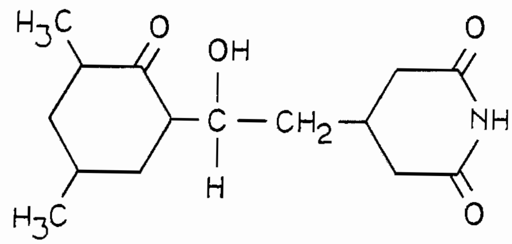

3-{2-(3,5-Dimethyl-2-oxocyclohexyl)-2-hydroxyethyl-}glutarimide

Data requirements are cited from EPA's Guidelines for Registering Pesticides (1981).

(1) DEGRADATION 163.161

(A) Hydrolysis 163.161-1

Hydrolysis data are required to support the registration of each end-use product intended for outdoor use or aquatic impact use, and each manufacturing-use product that may legally be used to formulate such an end-use product.

Three hydrolysis studies were reviewed and considered valid.

Cycloheximide was dehydrated in solution at all pH's tested (Garrett and Notari, 00011594 and 00011595), yielding trans-anhydrocycloheximide and a product, termed P, that could not be conclusively identified but was most likely α-epicycloheximide and/or α-epi-Naramycin B (Figure 1). One or more other isomers were formed and tentatively identified as α-epi-Naramycin B and/or isocycloheximide; the latter would be derived from cis-anhydrocycloheximide (Figure 1). The reaction is pH and temperature dependent. The rate increases with increasing temperature. The dehydration rate is rapid at very low

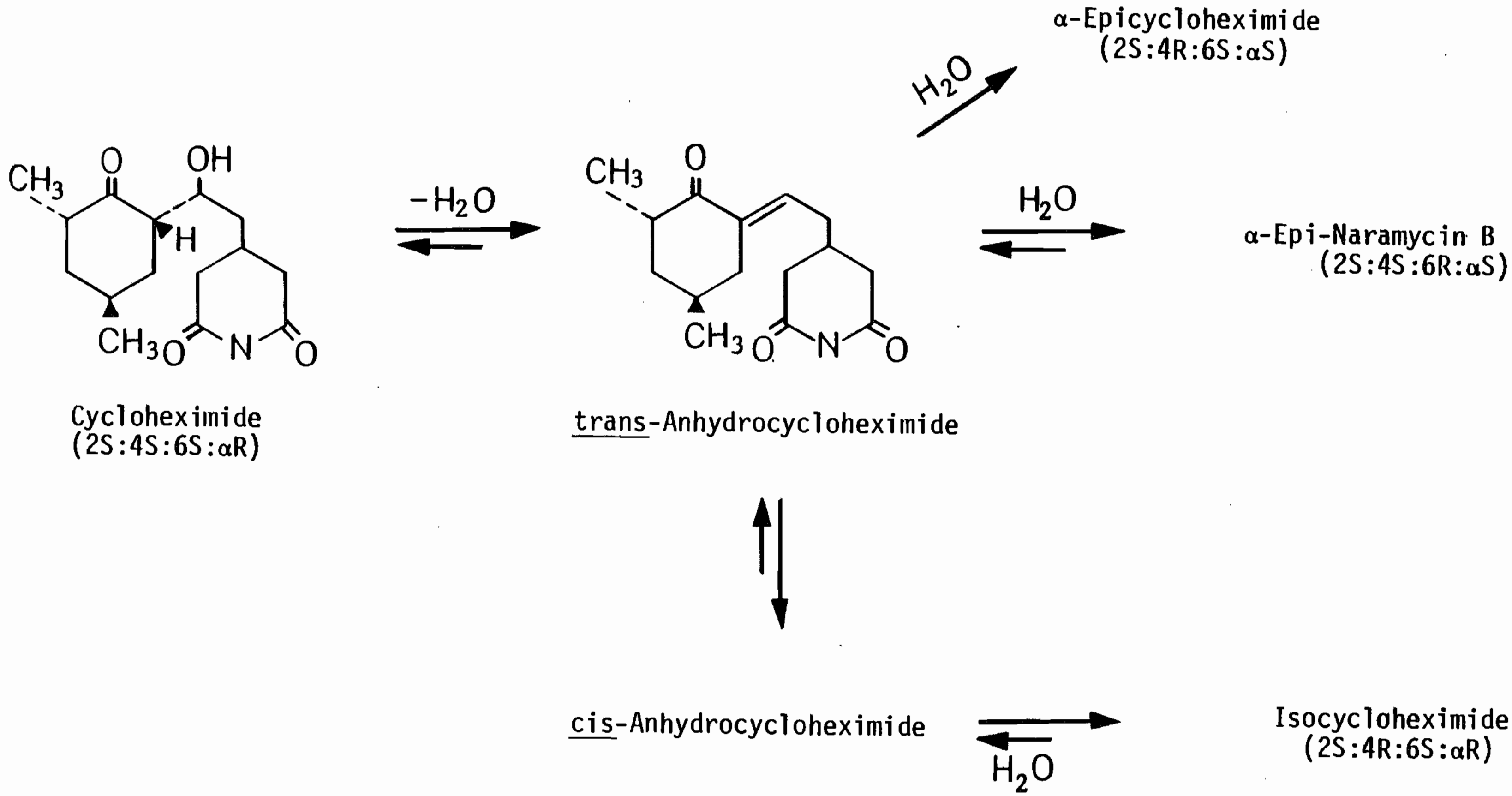

Figure 1. Dehydration of cycloheximide and the probable transformation products formed in acidic solutions.

Adapted from Garrett and Notari (00011594).

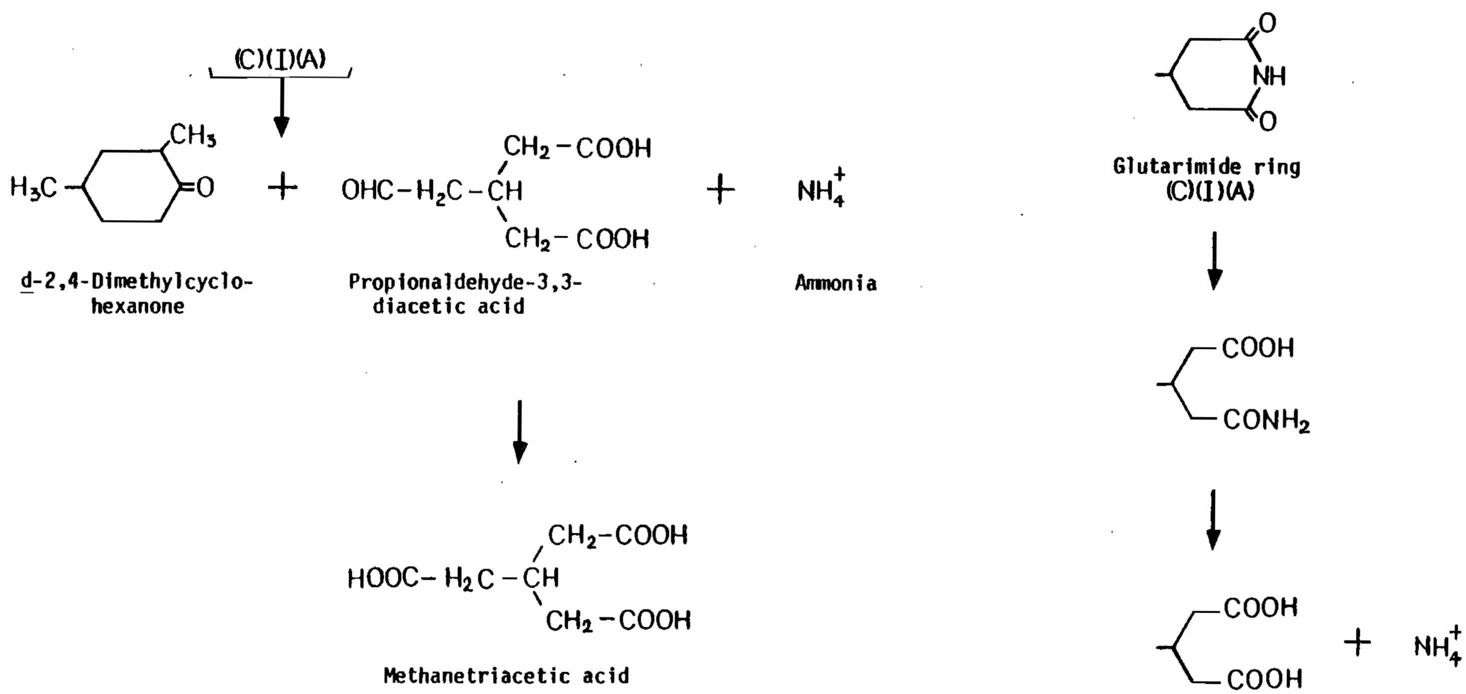

Figure 2. Hydrolysis of cycloheximide (C), its isomers (I), and anhydrocycloheximide (A). Adapted from Garret and Notari (00011594, 00011595) and Kernfeld (05013827).

and imide hydrolysis occurred simultaneously to yield propionaldehyde-3,3-diacetic acid and ammonia (Figure 2). Garrett and Notari (00011594 and 00011595) never found 2,4-dimethylcyclohexanone.

Data Gaps

All data specified in Section 163.161-1 are needed to assess the hydrolysis properties of cycloheximide.

References

Garrett, E.R., and R.E. Notari. 1965. Cycloheximide transformations. II. Kinetics and stability in a pharmaceutically useful pH range. J. Pharm. Sci. 54(2):209-215. (00011595)

Garrett, E.R., and R.E. Notari. 1966. Cycloheximide transformations. I. Kinetics and mechanisms in aqueous acid. J. Org. Chem. 31:425-434. (00011594)

Kornfeld, E.C. 1949. The structure and chemistry of Actidione, an antibiotic from Streptomyces griseus. J. Am. Chem. Soc. 71(1):150-159. (05013827)

(B) Photodegradation in Water 163.161-2

A photodegradation study in water is required to support the registration of each end-use product intended for terrestrial (except greenhouse and domestic outdoor), aquatic, and forestry use and for any aquatic impact use which results in direct discharges into the aquatic environment. Such a study is also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product.

No data on the photodegradation of cycloheximide in water are available.

Data Gaps

All data specified in Section 163.161-2 are needed to assess the photodegradation of cycloheximide in water.

(C) Photodegradation on Soil 163.161-3

Photodegradation studies on soil surfaces are required to support the registration of each end-use product intended for orchard crop use, field and vegetable crop use, or forestry use. Such studies are also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product. However, uses involving injection of the product into the soil or incorporation of the product into the soil upon application are not subject to the requirements of this section.

No data on the photodegradation of cycloheximide on soil are available.

Data Gaps

All data specified in Section 163.161-3 are needed to assess the photodegradation of cycloheximide on soil.

(D) Photodegradation in Air 163.161-4

A laboratory photodegradation study in the vapor phase will be required on a case-by-case basis to support the registration of an end-use product with orchard or field and vegetable crop uses that involve potentially significant exposure to workers. Data from such a study will also be required to support the registration of a manufacturing-use product which legally could be used to make such an end-use product. The Agency will make an assessment of what constitutes a significant inhalation exposure to workers based on the information required by Section 163.163-2(b)(2).

No data on the photodegradation of cycloheximide in air are available.

Data Gaps

All data specified in Section 163.161-4 may be needed to assess the photodegradation of cycloheximide in air if a significant inhalation exposure to workers is indicated based on the information required by Section 163.163-2(b)(2).

(2) METABOLISM 163.162

Data on metabolism are required to determine the nature and availability of pesticide residues to rotational crops and to help in the assessment of potential disposal and reentry hazards.

(A) Aerobic Soil 163.162-1

An aerobic laboratory soil metabolism study is required to support the registration of each end-use product intended for terrestrial or forestry use. Such a study is also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product.

Four aerobic soil metabolism studies were reviewed and considered valid.

Cycloheximide had a half-life of about 2-3 days in sandy soil treated at 0.13 ppm and incubated outdoors in Florida during January (Petzold et al., 00012843). In another study, 92 and >99% of the applied cycloheximide (10 ppm) had dissipated after 11 days of incubation in a loam (at 28 C) and an unidentified soil (at 24 C), respectively. (Gottlieb et al., No MRID). At the same time, 53 and 70% had dissipated from sterilized portions of the respective soils, demonstrating that cycloheximide dissipation in soil is a result of both physicochemical mechanisms and microbial metabolism. Petzold and Chapman (00012845) obtained similar results with different soils, but because the presterilized soil became contaminated no precise comparative data were available. In their study, cycloheximide had a half-life of ∿1.5 days in a mixture of three Florida soils treated with [^{14}C]-cycloheximide at 0.2 ppm and incubated aerobically at 35 C. Extractable ^{14}C had a half-life of 2.25 days. More than 80% of the applied ^{14}C dissipated in the form of volatile compounds after 9 days of incubation, and <15% was unextractable. Cycloheximide's decline pattern was shown to be biphasic in another study by Petzold and Chapman (00011196) using the same soil mixture. At 35 C, the initial half-life was ~2.8 days, and the second and third half-lives were 4.5-5 days; 90% dissipated by the 13th day.

In summary, cycloheximide dissipates rapidly in soil. Precise quantitation of decline patterns is not possible because the studies by Petzold and Chapman were done at high incubation temperatures that do not reflect usual environmental conditions, and the other studies presented very limited data. However, the combined data indicate that cycloheximide should have a half-life of only a few days, perhaps a week, in most soils. Microbial metabolism and physicochemical transformations all play a role in the dissipation of cycloheximide. Products formed in soil were not identified, but most of the products (and possibly the parent compound) are probably volatile and significant accumulation of unextractable products is not expected.

Data Gaps

All data specified in Section 163.162-1 are needed to assess the aerobic soil metabolism of cycloheximide.

References

Gottlieb, D., P. Siminoff, and M.M. Martin. 1952. The production and role of antibiotics in soil. IV. Actidione and clavacin. Phytopathology 42(9):493-496. (No MRID)

Petzold, E.N., and D.D. Chapman. 1970. The stability of cycloheximide in a controlled soil environment: Report No. 120-9760-30. (Unpublished study received July 28, 1970, under 1023-EX-27; submitted by Upjohn Co., Kalamazoo, MI; CDL:210033-D). (00011196)

Petzold, E.N., and D.D. Chapman. 1971. Fate of cycloheximide when incorporated into sterile vs. nonsterile soil: Report No. 120-9760-51.

(Unpublished study received Mar. 22, 1972, under 2F1252; submitted by Upjohn Co., Kalamazoo, MI; CDL:095124-F). (00012845)

Petzold, E.N., A.W. Neff, and D.D. Chapman. 1971. Effect of repeated applications of water upon the migration and persistence of cycloheximide in a treated plot of Florida soil: Report No. 120-9760-52. (Unpublished study received Mar. 22, 1972, under 2F1252; submitted by Upjohn Co., Kalamazoo, MI; CDL:095124-D). (00012843)

(B) <u>Anaerobic Soil 163.162-2</u>

An anaerobic soil metabolism study is required to support the registration of each end-use product intended for field or vegetable crop use. Such a study is also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product. However, an anaerobic soil metabolism study shall not be required if an anaerobic aquatic metabolism study has been conducted in accordance with the requirements of Section 163.162-3.

No data on the anaerobic soil metabolism of cycloheximide are available.

No data on the anaerobic soil metabolism of cycloheximide are required because cycloheximide is not intended for use on field and vegetable crops.

(C) <u>Anaerobic Aquatic 163.162-3</u>

An anaerobic aquatic metabolism study is required to support the registration of each end-use product intended for aquatic use, forestry use, or for any aquatic impact use which results in direct discharges into the aquatic environment. Such a study is also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product. The anaerobic soil metabolism study in Section 163.162-2 may not be substituted for this study.

No data on the anaerobic aquatic metabolism of cycloheximide are available.

No data on the anaerobic aquatic metabolism of cycloheximide are required because cycloheximide is not intended for aquatic use, forestry use, or any use that results in direct discharges into the aquatic environment.

(D) Aerobic Aquatic 163.162-4

An aerobic aquatic metabolism study is required to support the registration of each end-use product intended for aquatic use or for any aquatic impact use which results in direct discharges into the aquatic environment. Such a study is also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product.

Two studies containing aerobic aquatic metabolism data were reviewed and considered valid. One of these was a fish accumulation study reported under two MRIDs and reviewed together (Petzold and Chapman, 00012880 and 00012864).

The half-life of cycloheximide at 0.09 ppm in aquarium water (containing bluegills) at 17 C was 4.5 - 4.75 days and there was virtually no accumulation in the fish. In another study (Petzold et al., 00012843), irrigation ditch water was treated with cycloheximide at 0.2 ppm and incubated outdoors in Florida in January; cycloheximide was present at 0.12 ppm 5 days later.

No data on the aerobic aquatic metabolism of cycloheximide are required because cycloheximide is not intended for aquatic use or for any use that results in direct discharge into the aquatic environment.

References

Petzold, E.N., and D.D. Chapman. 1971. Residues of ^{14}C-cycloheximide in bluegills from exposure via water for a month: Report No. 120-9764-48. (Unpublished report submitted by Upjohn Co., Kalamazoo, MI). (00012880)

Petzold, E.N., and D.D. Chapman. 1972. Residues of ^{14}C-cycloheximide in bluegills from exposure via water for a month: Report No. 120-9764-48 (Revised). (Unpublished report submitted by Upjohn Co., Kalamazoo, MI). (00012864)

Petzold, E.N., A.W. Neff, and D.D. Chapman. 1971. Effect of repeated applications of water upon the migration and persistence of cycloheximide in a treated plot of Florida soil: Report No. 120-9760-52. (Unpublished study received Mar. 22, 1972, under 2F1252; submitted by Upjohn Co., Kalamazoo, MI; CDL:095124-D). (00012843)

(3) MICROBIOLOGICAL

The requirement for the submission of microbiological data is currently being reserved.

(A) Effects of Microbes on Pesticides

Two studies containing data on the effects of microbes on cycloheximide were reviewed and considered valid.

More than 90% of the applied cycloheximide (10 ppm) dissipated after 11 days of incubation in two types of soils, whereas 50-70% dissipated from sterile samples (Gottlieb et al., No MRID). In a similar study (Petzold and Chapman, 00012845), [^{14}C]cycloheximide applied to a mixture of three Florida soils at 0.2 ppm had a half-life of 1.5 days, and ^{14}C had a half-life of 2.25 days. In presterilized samples, the respective half-life values were 9 and 17 days, even though these samples became contaminated during the experiment.

References

Gottlieb, D., P. Siminoff, and M.M. Martin. 1952. The production and role of antibiotics in soil. IV. Actidione and clavacin. Phytopathology 42(9):493-496. (No MRID)

Petzold, E.N., and D.D. Chapman. 1971. Fate of cycloheximide when incorporated into sterile vs. nonsterile soil: Report No. 120-9760-51. (Unpublished study received Mar. 22, 1972, under 2F1252; submitted by Upjohn Co., Kalamazoo, MI; CDL:095124-F). (00012845)

(B) Effects of Pesticides on Microbes

Nineteen studies on the effects of cycloheximide on microbes were reviewed and 17 were considered valid.

Cycloheximide at 33 ppm inhibited O_2 uptake by the protozoan Tetrahymena pyriformis at 29.5 C by 35-60% of control levels, depending on the substrates (Mefferd and Loefer, 05017110). Respiration in two types of soils was inhibited 6-31% over a 6-hour period by cycloheximide at 4,000 ppm (Vancura and Kunc, 05016745). In another study (Anilkumar and Chakravarti, 05019056), corn seedlings were treated with cycloheximide at 1 ppm and then transplanted into untreated soil. After 7 days, the population of rhizosphere fungi was reduced by 50% relative to controls, but bacterial populations were not affected.

Cycloheximide inhibits and delays nodulation on roots of Trifolium subterraneum (clover) seedlings inoculated with Rhizobium trifolii (Robinson, 05013679). Nodules were not formed during 18 days after treatment with cycloheximide at 100 ppm, and only 25% formed nodules after treatment at 20 ppm. In another study, nitrogen fixation by the blue-green alga Anabaena doliolum (in culture at 30 C) was inhibited about 30 and 70% by cycloheximide at 50 and 100 ppm, respectively (Kapoor and Sharma, 05021783). Growth in an elemental nitrogen medium was inhibited by cycloheximide at >350 ppm, but it was stimulated at 50-150 ppm.

The growth of the halophilic blue-green alga Aphanotheca halophytica (in culture at 22 C) was not inhibited by cycloheximide at 0.5-10 ppm (Yopp et al., 05018934). Growth was inhibited 10 and 25% by cycloheximide at 50-100 and 500 ppm, respectively. Cycloheximide, at 28 ppm, completely inhibited rhizoid formation by spherical cells of the marine green alga Boergesenia forbesii (Ishizawa et al., 05021055).

Cycloheximide is toxic to fungi. The growth of 11 yeast species from soil, 7 from leaves, and 7 of uncertain habitat (6 Candida, 5 Cryptococcus, 4 Rhodotorula, 3 Hansenula, 2 Trichosporon, 1 Torulopsis, 1 Sporobolomyces, 1 Saccharomyces, 1 Debaryomyces, and 1 Schizoblastosporion species) was inhibited by cycloheximide at 1,000 mg/disc on agar plates at 20 C (Di Menna, 05014316). The growth of eight of nine species of mycorrhizal fungi (5 Amanita, 2 Boletus, 1 Russula, and 1 Cenococcum species) was significantly inhibited on agar containing cycloheximide at 1 ppm and at 21 C (Hacskaylo, 05013615). At 0.1 ppm, cycloheximide had relatively little effect on growth. In a study by Partridge (05016063), cycloheximide at 50 ppm completely inhibited the growth of 11 forest fungi (3 Boletus and 2 Fomes species, and 1 species of Poria, Boletinus, Armillaria, Echinodontium, Polyporus and Suillus) in culture at room temperature. All but one were inhibited by cycloheximide at 10 ppm, and half were markedly inhibited at 0.1 ppm.

The growth of four Pythium strains (P irregulare, P. rostratum, and two strains of P. ultimum) at 20 C was inhibited 10-70% and 40-80% on water agar media containing cycloheximide at 1 and 3 ppm, respectively (Vaartaja and Agnihotri, 05013346). In another study, a strain of P. debaryanum was completely inhibited on potato dextrose agar and in soil containing cycloheximide at 2.5 and 5 ppm, respectively (Gottlieb et al., No MRID). In the same study, the growth of the yeast Saccharomyces pastorianus was inhibited 90% in soil containing cycloheximide at 5-20 ppm. Coursen and Sisler (05013934) determined that cycloheximide at 0.018 ppm was required to inhibit growth of S. pastorianus by 50% in broth culture at room temperature.

Sporulation of Pythium strains on water agar was inhibited by cycloheximide at 1-5 ppm (Vaartaja and Agnihotri, 05013346). Germination spores was inhibited about 40, 60, and 80% by cycloheximide at 1, 3, and 5 ppm, respectively. In the presence of yeast extract, sucrose, or asparagine, there was only 5-20% inhibition. Germination in soil was completely inhibited at 1-5 ppm, and the nutrients had a negligible counter-inhibitory effect. Spore germination in Penicillium notatum and Trichoderma lignorum was inhibited by 90-100% during 3 days of incubation at 26 C in broth containing cycloheximide at 50 ppm (Martin and Nicolas, 05012807). Cycloheximide at 10-100 ppm in broth temporarily inhibited conidial germination in Aspergillus nidulans at 37 C (Ryder and Peberdy, 05021327). The inhibitory effect virtually disappeared after 24 hours. Germination of Colletotrichum lagenarium spores was inhibited over a 24-hour period at 24 and 32 C by cycloheximide at 1 ppm (Tani et al., 05013498).

Cycloheximide induced mutations in extrachromosomal DNA in Saccharomyces cerevisiae. At 0.7 ppm, cycloheximide induced the elimination of gene(s) responsible for the killer phenotype (Naumova and Naumov, 05016140).

In summary, cycloheximide is highly toxic to fungi. Based on the available data, cycloheximide in soil at ≥1 ppm can be expected to markedly inhibit the growth of most fungi in soil. However, the growth of most bacteria should not be affected by cycloheximide at low concentrations. Although the data are limited, they indicate that the growth of some blue-green algae is inhibited by cycloheximide at high concentrations (≥50 ppm). Nitrogen fixation can be inhibited by cycloheximide at relatively high concentrations. In vitro nitrogen fixation by A. doliolum was inhibited by cycloheximide at ≥50 ppm. Only one symbiotic nitrogen-fixing association was studied (Rhizobium trifolii and clover), and nodule formation was inhibited and delayed by cycloheximide at ≥20 ppm.

References

Anilkumar, T.B., and B.P. Chakravarti. 1970. Effect of root feeding with antibiotics on rhizosphere microflora of maize seedlings. Biochem. Physiol. Pflanz. 161(5):442-446. (05019056)

Coursen, B.W., and H.D. Sisler. 1960. Effect of the antibiotic, cycloheximide, on the metabolism and growth of Saccharomyces pastorianus. Am. J. Bot. 47(7):541-549. (05013934)

Di Menna, M.E. 1962. The antibiotic relationships of some yeasts from soil and leaves. J. Gen. Microbiol. 27:249-257. (05014316)

Gottlieb, D., P. Siminoff, and M.M. Martin. 1952. The production and role of antibiotics in soil. IV. Actidione and clavacin. Phytopathology 42(9):493-496. (No MRID)

Hacskaylo, E. 1961. Influence of cycloheximide on growth of mycorrhizal fungi and on mycorrhizae of pine. For. Sci. 7(4):376-379. (05013615)

Ishizawa, K., S. Enomoto, and S. Wada. 1979. Germination and photo-induction of polarity in the spherical cells regenerated from protoplasm fragments of Boergesenia forbesii. Bot. Mag. 92(1027): 173-186. (05021055)

Kapoor, K., and V.K. Sharma. 1979. Effects of cycloheximide and maleic hydrazide on a nitrogen-fixing cyanophyte. Biochem. Physiol. Pflanz. 174(5/6):509-512. (05021783)

Martin, J.F., and G. Nicolas. 1970. Physiology of spore germination in Penicillium notatum and Trichoderma lignorum. Trans. Br. Mycol. Soc. 55(1):141-148. (05012807)

Mefferd, R.B., Jr., and J.B. Loefer. 1954. Inhibition of respiration in Tetrahymena pyriformis S by Actidione. Physiol. Zool. 27:115-118. (05017110)

Naumova, T.I., and G.I. Naumov. 1974. Induced elimination of killer cytogenes (k_1) and (k_2) and the neutral cytogene (n) of the Saccharomyces yeasts. Biol. Sci. (Moscow) 17(2):108-110. (05016140)

Partridge, A.D. 1966. Some effects of cycloheximide on selected forest fungi. Plant Dis. Rep. 50(7):497-499. (05016063)

Robinson, A.C. 1968. The effect of anti-fungal antibiotics on the nodulation of Trifolium subterraneum and the estimation of Rhizobium trifolii populations. Aust. J. Exp. Agric. Anim. Husb. 8(32):327-331. (05013679)

Ryder, N.S., and J.F. Peberdy. 1979. Chitin synthetase activity and chitin formation in conidia of Aspergillus nidulans during germination and the effect of cycloheximide and 5-fluorouracil. Exp. Mycol. 3(3): 259-269. (05021327)

Tani, M., N. Ishida, and I. Furusawa. 1977. Effects of temperature and antibiotics on appressorium formation in spores of Colletotrichum lagenarium. Can. J. Microbiol. 23(5):626-629. (05013498)

Vaartaja, O., and V.P. Agnihotri. 1969. Interaction of nutrients and four antifungal antibiotics in their effects on Pythium species in vitro and in soil. Plant Soil 30(1):49-61. (05013346)

Vancura, V., and F. Kunc. 1977. The effect of streptomycin and Actidione on respiration in the rhizosphere and non-rhizosphere soil. Zentralbl. Bakteriol. Parasitenkd. Infektionskr. Hyg., Abt. II 132(5/6): 472-478. (05016745)

Yopp, J.H., G. Albright, and D.M. Miller. 1979. Effects of antibiotics and ultraviolet radiation on the halophilic blue-green alga, Aphanotheca halophytica. Bot. Mar. 22(5):267-272. (05018934)

(C) Activated Sludge

No data on the activated sludge metabolism of cycloheximide are available.

(4) MOBILITY 163.163

The movement of pesticide residues by means of leaching through the soil may cause contamination of food, result in a loss of usable land and water resources to man due to contamination of groundwater supplies, or cause habitat loss to wildlife. Therefore, studies are required to ascertain the extent of pesticide leaching through soil, which, in turn, provides a basis for assessing the mobility potential of a pesticide.

(A) Leaching 163.163-1

Data are required to support the registration of each end-use product intended for domestic outdoor use, greenhouse use, terrestrial non-crop use, orchard crop use, field or vegetable crop use, forestry use, aquatic use, and aquatic impact use involving direct discharge only. Such data are also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product.

For terrestrial non-crop uses, orchard crop uses, field or vegetable crop uses, and forestry uses, the mobility of the test substance and its degradates in soil shall be assessed either by soil thin-layer chromatography, soil column, or batch equilibrium (adsorption/desorption). For domestic outdoor uses, greenhouse uses, aquatic uses, and aquatic impact uses, the mobility of the test substance and its degradates in soil shall be assessed only by batch equilibrium (adsorption/desorption).

Five studies containing leaching data were reviewed and four were considered valid.

[^{14}C]Cycloheximide is very mobile in sand soil. About 96% of the applied radioactivity leached through a 12-inch column eluted with the equivalent of 20 acre-inches of water (Staten et al., 00011189).

About 64% of the ^{14}C was in the form of cycloheximide. When residues were aged for 30 days (Staten et al., 00011190), about 27% of the applied ^{14}C was lost during aging. The leachate contained 52% of the applied radioactivity and the soil contained 15%, half of which remained in the top 2 inches. Cycloheximide was undetectable (<0.015 ppm) in the leachate. In a soil TLC study (Helling et al., 05001190), cycloheximide was mobile to very mobile (R_f 0.89) in a silty clay loam soil sieved to <250 μm.

In a field study (Petzold et al., 00012843), cycloheximide was not detectable (<0.017 ppm) in the 4-12 inch layer of sandy soil for 5 days after treatment with cycloheximide (4.23% ai WP) at ∿0.19 lb ai/A and irrigation with 2 inches of water. Cycloheximide also was not detectable (<0.012 ppm) at any time in the groundwater (<20 inches from surface) sampled from the center of the treated area. Although the study was conducted in a valid manner, the leaching potential of cycloheximide and its degradation products cannot be adequately assessed from this study because the study period was too short, too little water was applied, and products were not sought.

These studies partially fulfill data requirements in Section 163.163-1 by providing leaching information for cycloheximide in sand (rapid and aged leaching) and silty clay loam soils.

Data Gaps

Two additional studies using a sandy loam, clay, or clay loam, and conducted as specified in Section 163.163-1, are needed to fully assess the leaching potential of cycloheximide. If suitable K_d (soil/water relationship) values cannot be calculated from soil column or soil thin-layer chromatography data, batch equilibrium (adsorption/desorption) studies will be needed to support domestic outdoor uses.

References

Helling, C.S., D.G. Dennison, and D.D. Kaufman. 1974. Fungicide movement in soil. Phytopathology 64:1091-1100. (05001190)

Petzold, E.N., A.W. Neff, and D.D. Chapman. 1971. Effect of repeated applications of water upon the migration and persistence of cylcoheximide in a treated plot of Florida soil: Report No. 120-9760-52. (Unpublished study received Mar. 22, 1972, under 2F1252; submitted by Upjohn Co., Kalamazoo, MI; CDL:095124-D). (00012843)

Staten, F.W., A.M. Thornton, and W.M. Wright. 1974. Soil leaching studies on Florida soil fortified with ^{14}C-cycloheximide and aged 30 days: Report No. 120-9760-92. (Unpublished study including letter dated Jan. 24, 1974, from F.W, Staten. A.M. Thornton, and W.M. Wright to A.W. Neff, received Sept. 29, 1977, under 1023-50; submitted by Upjohn Co., Kalamazoo, MI; CDL:097214-J). (00011190)

Staten, F.W., W.M. Wright, and A.M. Thornton. 1974. Soil leaching studies on cycloheximide: Report No. 120-9760-93. (Unpublished study including letter dated Jan. 25, 1974, from F.W. Staten, W.M. Wright, and A.M. Thornton to A.W. Neff, received Sept. 29, 1977, under 1023-50; submitted by Upjohn Co., Kalamazoo, MI; CDL:097214-I). (00011189)

(B) <u>Laboratory Volatility 163.163-2</u>

A laboratory volatility study will be required on a case-by-case basis to support the registration of each end-use product intended for commercial greenhouse, orchard, or field and vegetable crop uses that involve significant inhalation exposure to workers. Data from such a study will also be required to support the registration of each manufacturing-use product which legally could be used to make any end-use product for which laboratory volatility data are required.

No data on the volatility of cycloheximide under laboratory conditions are available.

Data Gaps

All data specified in Section 163.163-2 may be needed to assess the volatility of cycloheximide under laboratory conditions if significant inhalation exposure to workers is indicated based on the information required by Section 163.163-2(b)(2). The data requirements of this section may be satisfied by data produced from a study that meets the test standards contained in Section 163.163-3.

(C) Field Volatility 163.163-3

A volatility study conducted on-site in a commercial greenhouse and/or in the field will be required on a case-by-case basis only for those pesticides that the Agency considers pose a potentially significant inhalation exposure to workers (see Section 163.163-2(b)) and, based on the results of the laboratory study described in Section 163.163-2, that also demonstrate, in the Agency's opinion, a significant rate of volatilization from soil.

No data on the field volatility of cycloheximide are available.

Data Gaps

All data specified in Section 163.163-3 may be needed to assess the volatility of cycloheximide under field conditions if significant inhalation exposure to workers is indicated based on the information required by Section 163.163-2(b)(2).

(5) DISSIPATION 163.164

(A) Field Dissipation - Terrestrial 163.164-1

A terrestrial field dissipation study is required to support the registration of each end-use product intended for any terrestrial use (except greenhouse use). Such data are also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product.

Fifteen terrestrial field dissipation studies were reviewed and one was considered valid.

Cycloheximide dissipated rapidly under field conditions (Petzold et al., 00012843). Cycloheximide had a half-life of 2 days in the top 4 inches of sandy soil treated with cycloheximide (4.23% ai WP) at ∿0.19 lb ai/A. Cycloheximide did not move below 4 inches over a 5-day period in which the soil was irrigated with 2 inches of water. Data on metabolites were not obtained.

Data Gaps

All data specified in Section 163.164-1 are needed to assess the terrestrial field dissipation of cycloheximide.

References

Petzold, E.N., A.W. Neff, and D.D. Chapman. 1971. Effect of repeated applications of water upon the migration and persistence of cycloheximide in a treated plot of Florida soil: Report No. 120-9760-52. (Unpublished study received Mar. 22, 1972, under 2F1252; submitted by Upjohn Co., Kalamazoo, MI; CDL:095124-D). (00012843)

(B) Field Dissipation - Aquatic and Aquatic Impact 163.164-2

An aquatic field dissipation study is required to support the registration of each end-use product intended for aquatic food crop uses, aquatic non-crop uses, and for any aquatic impact use which results in direct discharges into the aquatic environment. Such a study is also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product.

No data on the aquatic field dissipation of cycloheximide are available.

No data on aquatic field dissipation are required because cycloheximide is not intended for aquatic use or any use that results in direct discharges into the aquatic environment.

(C) <u>Dissipation - Forestry 163.164-3</u>

Field dissipation studies for forestry uses are required to support the registration of each end-use product intended for forestry use and of each manufacturing-use product which legally could be used to make such an end-use product.

No data on the dissipation of cycloheximide in forests are available.

No data on the dissipation of cycloheximide in forests are required because the use pattern indicates that introduction of cycloheximide into a forest environment would not occur.

(D) <u>Dissipation - Combinations and Tank Mixes 163.164-4</u>

A laboratory or field soil dissipation study may be required on a case-by-case basis to support the registration of an end-use product containing more than one active ingredient, intended for use as a component in tank mixtures, or customarily applied serially with another pesticide product.

No data on the dissipation of multiple active ingredient formulations of cycloheximide are available.

Data requirements for combinations and tank mixes containing cycloheximide are not cited here because this standard deals only with the single active ingredient.

(E) Dissipation - Long Term 163.164-5

A long-term soil dissipation study will be required to support the registration of the end-use products of any pesticide that has been shown not to readily dissipate in a soil environment. Such a study may also be required to support the registration of any manufacturing-use product that legally could be used to make such an end-use product.

No data on long-term dissipation of cycloheximide in soil are available.

No data are required on the long-term dissipation of cycloheximide because cycloheximide will not be used to treat field and vegetable crops or aquatic food crops.

(6) <u>ACCUMULATION 163.165</u>

(A) <u>Confined Accumulation - Rotational Crops 163.165-1</u>

Confined accumulation studies on rotational crops are required to support the registration of each end-use product intended for field or vegetable crop use, aquatic crop use, or use on any other site on which it is reasonably forseeable that any food or feed crop may be produced after application of a pesticide. Such studies are also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product.

No data on the confined accumulation of cycloheximide in rotational crops are available.

No data on the confined accumulation of cycloheximide in rotational crops are required because the use pattern indicates that cycloheximide is used on crops which are not grown in rotation with other crops.

(B) <u>Field Accumulation - Rotational Crops 163.165-2</u>

A field accumulation study to determine the uptake of soil residues by rotational crops is required when (1) the confined accumulation study (Section 163.165-1) identifies the ^{14}C residues in the crop as either parent compound, closely-related degradates, metabolites, and/or their conjugates or (2) a subsequent crop is treated with the same active ingredient as the initial crop.

No data on the field accumulation of cycloheximide in rotational crops are available.

No data on the field accumulation of cycloheximide in rotational crops are required because the use pattern indicates that cycloheximide is used on crops which are not grown in rotation with other crops.

(C) Accumulation - Irrigated Crops 163.165-3

A study of residue accumulation in irrigated crops under actual field use conditions is required to support the registration of each end-use product intended for aquatic food crop or aquatic non-crop uses, for uses in and around holding ponds used for irrigation purposes, or for uses that involve effluents and other discharges which in turn are used to irrigate crops. Such a study is also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product.

No data on the accumulation of cycloheximide in irrigated crops are available.

No data on the accumulation of cycloheximide in irrigated crops are required because the use pattern indicates that crops are not irrigated with cycloheximide-treated water.

(D) Laboratory Studies - Fish 163.165-4

A fish accumulation study is required to support the registration of each end-use product intended for outdoor use (except domestic outdoor and greenhouse uses), or aquatic impact use resulting in direct discharge into aquatic environments, and for each manufacturing-use product that legally could be used to produce such a product, except when the criteria below are satisfied.

Fish accumulation data will not normally be required in situations where the registrant can offer acceptable evidence showing that the active ingredient and/or its principal degradation product(s):

- Will not reach water, or
- Will not persist in water (i.e., a half-life of approximately 4 days or less) and has properties suggesting:

- A relatively low potential for accumulation in fish (i.e., an octanol/water partition coefficient less than ~1,000) or

- A lack of accumulation in the organs and tissues of mammals or birds.

One fish accumulation study, reported under two MRIDs (Petzold and Chapman, 00012880 and 00012864), was reviewed and considered valid.

Cycloheximide does not accumulate in bluegill sunfish. The maximum bioaccumulation factor was <10 for bluegills in a static system exposed to two treatments of [^{14}C]cycloheximide, 10 days apart, at 0.09 ppm each. Maximum ^{14}C residue levels over a 1-month exposure period were <0.14 ppm in tissues of whole fish, and <0.035 ppm in chloroform extracts (would contain cycloheximide). ^{14}C residue levels declined to ≤0.01 ppm after 3 days of depuration.

Further data on the accumulation of cycloheximide in fish are not required for the following reasons. The data discussed in this section and in Sections 163.161-1 and 163.162-4 indicate that cycloheximide rapidly dissipates in water, with a half-life of about 3-7 days observed in natural systems without sediments. Cycloheximide is very soluble in water (2.1% at 2 C; The Merck Index, 9th ed., Merck & Co., Rahway, NJ, 1976), and therefore should have a low octanol/water partition coefficient. The fish study showed little accumulation, and rapid depuration, of ^{14}C residues in bluegills exposed to [^{14}C]cycloheximide.

References

Petzold, E.N., and D.D. Chapman. 1971. Residues of ^{14}C-cycloheximide in bluegills from exposure via water for a month: Report No. 120-9764-48. (Unpublished report submitted by Upjohn Co., Kalamazoo, MI). (00012880)

Petzold, E.N., and D.D. Chapman. 1972. Residues of ^{14}C-cycloheximide in bluegills from exposure via water for a month: Report No. 120-9764-48

(Revised). (Unpublished report submitted by Upjohn Co., Kalamazoo, MI). (00012864)

(E) <u>Field Accumulation - Aquatic Non-Target 163.165-5</u>

Field accumulation studies in aquatic non-target organisms are required to support the registration of each end-use product.

- Which is intended for forestry use, aquatic non-crop use, or aquatic impact use that results in direct discharges;
- For which data from a laboratory fish accumulation study are required by Section 163.165-4; and
- For which no tolerance or action level for fish has been granted.

Such studies are also required to support the registration of each manufacturing-use product which legally could be used to make such an end-use product.

No data on the accumulation of cycloheximide in aquatic non-target organisms are available.

No data on the accumulation of cycloheximide are required because cycloheximide is not intended for forestry use, aquatic uses, or aquatic impact uses that result in direct discharges.

(7) <u>REENTRY (SUBPART K)</u>

Reentry data may be required to support the registration of manufacturing-use and end-use cycloheximide. Requirements will be decided on a case-by-case basis.