

EPA Comments on Chemical RTK Challenge Submission:

3,5-Di-*tert*-butyl-4-hydroxyhydrocinnamic acid neopentetetrayl ester

SUMMARY OF EPA COMMENTS

The sponsor, Ciba Specialty Chemicals Corp., submitted Robust Summaries to EPA and a Test Plan that were received July 10, 2000, and a test plan to the HPV Tracking System Web site (www.hpvchallenge.com) for 3,5-Di-*tert*-butyl-4-hydroxyhydrocinnamic acid neopentetetrayl ester (CAS # 6683-19-8). EPA posted the submission on the ChemRTK website on July 20, 2000.

EPA has reviewed this submission and has reached the following conclusions:

1. The submission does not meet minimal standards for data adequacy. There were many inadequacies in the health and ecological effects study summaries, which must be revised to be acceptable for the Challenge Program. EPA has provided specific comments on how to enhance the robust summaries. Sponsors should refer to the Challenge Program guidance.

EPA accepts the submission conditionally, believing that the issue is poor documentation but that enough information may be inferred to make tentative judgements. Eventual full acceptance of the submission is contingent upon the receipt within 90 days of substantially improved robust summaries and other information that can meet the standard set out in EPA's guidance documents.

2. Physicochemical properties and environmental fate. The sponsor supplied calculated data without citing available experimental data. Similarly, the sponsor used only estimated data as inputs into the fugacity model. EPA prefers measured data when available. To estimate transport and distribution, the sponsor used the EPIWIN Level III model which provides estimated values as default inputs. EPA recommends using the EQC level III model from the Canadian Environmental Modeling Centre at Trent University.

3. Health endpoint testing: Most of the robust summaries are inadequate because not enough information is presented to allow for an independent assessment of the data. However, EPA's tentative scientific judgment is that no further testing is needed for the purposes of the U.S. HPV Challenge Program, provided that the sponsor supplies adequate documentation as discussed under Item 1 above.

4. Ecological effects data. Although there were many inadequacies in the study summaries, EPA suggests that an analysis based on this chemical's physicochemical properties, including extremely low water solubility, may support the sponsor's conclusion that no further testing is necessary. EPA will take into account adequate documentation of such an analysis supplied by the sponsor in determining final acceptance of the test plan.

EPA is requesting that the Sponsor advise the Agency within 60 days of any modifications to its submission.

EPA COMMENTS ON THE 3,5-DI-*tert*-BUTYL-4-HYDROXYHYDROCINNAMIC ACID NEOPENTANETETRAYL ESTER CHALLENGE SUBMISSION

General

The submission does not meet minimal standards. There were many inadequacies in the health and ecological effects study summaries, which must be revised to allow final acceptance as an HPV Challenge submission. EPA has provided specific comments on how to enhance the robust summaries to the standard established in EPA's HPV Challenge Program Guidance (<http://www.epa.gov/opptintr/chemrtk/guidocs.htm>).

The test plan on the industry HPV Tracking System Web site and the test plan summary table submitted with the robust summaries to EPA were substantially different. EPA contacted the sponsor and learned that the sponsor was unable to make desired corrections to the Tracking System submission.

Test Plan

Chemistry (melting point, boiling point, vapor pressure, water solubility, and partition coefficient).

EPA believes that no additional test data are needed to satisfy the needs of the HPV Challenge Program.

Fate (photodegradation, stability in water, biodegradation, and transport/distribution).

EPA believes that no additional test data are needed to satisfy the needs of the HPV Challenge Program.

Health Effects (acute toxicity, repeat dose toxicity, genetic toxicity, and reproductive/developmental toxicity).

EPA's tentative judgement is that no additional test data are needed to satisfy the needs of the HPV Challenge Program pending receipt of adequate robust summaries.

Ecological Effects.

EPA's tentative judgement is that no additional test data are necessary to satisfy the needs of the HPV Challenge Program pending receipt of adequate robust summaries. An adequately documented analysis, such as a quantitative structure-activity relationship (QSAR) analysis, based on this chemical's physicochemical properties may provide additional support to the sponsor's conclusion that further aquatic testing is unnecessary.

SPECIFIC COMMENTS ON ROBUST SUMMARIES

Chemistry

All the physicochemical property data (estimated using EPIWIN) are acceptable, except for melting point, which is much higher than the measured value.

The estimated melting point value of 349.8 EC, provided by the sponsor, does not match the measured value of 115–118 EC (decomposition) (Aldrich Catalog Handbook of Fine Chemicals). For chemicals with a molecular weight greater than about 200 or with more than 15 carbons, EPIWIN almost always predicts a melting point much higher than observed.

The sponsor calculated log P_{ow} values of 19 and 23 using two different programs. Both values are so much higher than for the most hydrophobic materials (typically log P_{ow} = 14) that the difference is not significant.

Fate

The environmental fate data have been reviewed. The Robust Summary information is satisfactory and the sponsor's approach is reasonable. Since this chemical has an extremely low water solubility and very low vapor pressure, it is reasonable to present calculated values; however, a measured melting point is available for input to estimation models.

EPA recommends using measured data as much as possible. The sponsor used the EPIWIN Level III model, which provides estimated values as default inputs. In order to estimate environmental fate endpoints, however, EPA recommends using the EQC level III model from the Canadian Environmental Modeling Centre at Trent University. This model can be found at the following Web address: <http://www.trentu.ca/academic/aminss/envmodel/>.

Health Effects

EPA evaluated 11 health endpoint robust summaries and found all of them to be inadequate for the purposes of the U.S. HPV Challenge Program. Nine of the 11 summaries describe non-GLP, non-guideline studies (the exceptions were the 13-week dog study and the two-generation rat study). EPA's scientific judgment is that new studies are not necessary for the purposes of the U.S. HPV Challenge Program, pending receipt of adequate robust summaries.

The following EPA comments reflect the information in the robust summaries (the full study reports may

address these comments):

Acute Toxicity (oral, rats). (1) The year of the study is given as 1968–1974. The summary states that these findings validate several earlier studies that found the LD₅₀ to be greater than 10,000 mg/kg. One assumes that these earlier studies were done during 1968–74 and that this study is the last in the series. However, this point should be clarified. (2) This is a three-dose study with 2 animals/sex/dose and 2 batches of test material at each dose level. At best there is a question of the statistical reliability of the data with only 2 animals/sex/dose level. (3) There is no indication of the manner of delivery of the test material (i.e., gavage). (4) Although a necropsy was done at the end of the 14-day observation period, no details are presented as to organs examined or what was found.

Acute toxicity (inhalation, rats). (1) There is no indication of clinical signs monitored, food consumption, initial and final body weight, or whether or not a necropsy was performed. (2) There is no discussion of whether or not mortality occurred at any dose level; therefore the summary is lacking in sufficient detail to allow for verification of the conclusion presented.

Acute toxicity (dermal, rabbits). The summary states that the study meets generally accepted scientific standards. However, insufficient data are presented to verify either the adequacy of the methodology or the accuracy of the results.

Genotoxicity (dominant lethal assay, mice). (1) No positive or negative controls were reported. (2) There is no indication of male:female mating ratio or length of time animals were allowed to mate. (3) In addition, although the statement is made that no evidence of dominant lethality was observed, no supporting data are presented.

Genotoxicity (somatic mutation assay, Chinese hamsters). (1) The number of animals/dose group is low and it is uncertain from the summary whether chromosomal aberrations or nuclear anomalies were scored. (2) There is no indication of the number of cells scored for either anomalies or aberrations. (3) No data are presented to support the claim that there is no difference between treated and control cells in numbers of anomalies.

Genotoxicity (somatic mutation assay, Chinese hamsters). (1) There is no indication of the methodology used to prepare slides for scoring of aberrations nor of the method used to score the slides, i.e. were slides coded? (2) No data were submitted to support the claim that the chemical is nonmutagenic,.

Genotoxicity (Ames test). (1) There is no rationale given for dose selection; (2) the background revertant colony counts are not reported; (3) there is no indication whether positive controls were used; (4) there is no indication of the solvent or vehicle used; (5) there is no indication of incubation time or temperature or method of counting, e.g. by hand or electronic colony counter; and (6) No data are presented to support the claim that there is no increase in reverse mutation with or without S9 fraction.

Repeat dose toxicity (oral via the diet, dogs). This study was reported as GLP and no adverse effects were reported. However, there is a report of an increase in total bilirubin concentration at certain time points during the study, but the magnitude and the dose levels are not provided. This information is necessary to interpret its significance.

Reproductive toxicity (oral via the diet, rats). In this GLP study, the summary does not present enough information to verify the results presented nor to judge the significance of the higher mean litter weights of the high-dose groups in the F₁ and F₂ generations. Since this can be a significant, treatment-related finding, more data are needed to judge its significance before a NOEL or LOEL for the F₁ or F₂ generation can be established. Likewise the narrative establishing the parental NOEL should be supported by data. A summary table, for example, would help a great deal.

Developmental toxicity (oral via gavage, rats). (1) There is no indication of the total number of females treated or mated. Five mated females to a cage is somewhat high, although depending on cage size it may be acceptable. (2) It is reported that there was an increase in food consumption in the dams in the low and intermediate dose groups and higher rates of ossification of the phalangeal nuclei of the hind limb and calcanei in the fetuses of the same two dose groups. Data should be presented to support both of these statements and to allow one to judge the effect of treatment upon other parameters such as number of successful matings, rate of implantation and resorption, litter weight, number of litters/treatment group, etc.

Developmental toxicity (oral via gavage, mice). (1) There is no indication of the total number of females

treated or mated. Five mated females per cage is somewhat high, although depending on cage size it may be acceptable. (2) In the low dose group incidences of ossification of the phalangeal nuclei of the hind limb and calcanei were "significantly" higher than controls. Since this same effect was also observed in rats, more information than a simple statement is needed to judge its significance. (3) At the high dose there was an increase in the number of incompletely ossified sternebrae. This statement is made twice and EPA questions whether it is a typographical error, or if it should say the mid- and high-dose. (4) The actual data should also be presented to allow one to judge the effect of treatment upon other parameters such as number of successful matings, rate of implantation and resorption, litter weight, number of litters/treatment group, etc.

Ecotoxicity Studies

The comments below reflect the information presented in the robust summaries; information in the full study report may address some of the issues identified.

Acute Aquatic Toxicity. Robust summaries were submitted for studies on fish, daphnia, and green algae. Many critical experimental details were omitted from the robust summaries, which effectively rendered them inadequate. EPA's tentative scientific judgment is that new studies are not necessary for the purposes of the U.S. HPV Challenge Program, pending receipt of adequate robust summaries and adequate analysis of potential effects.

Fish. Information missing from the robust summary includes: number of replicates/test, water chemistry, solvent/vehicle used, test substance purity, temperature, pH, TOC, and hardness. Test concentrations were above the predicted water solubility limit. Because this chemical has low water solubility, the information provided does not allow a conclusion to be made as to how much of the test was conducted according to the Guidance Document on Aquatic Toxicity Testing of Difficult Substances and Mixtures (OECD, June 2000, available at <http://www.oecd.org/ehs/test/monos.htm>). The vehicle concentration was above the recommended concentration of #100 mg/L.

Aquatic plants. Information missing from the submitted algal inhibition test robust summary includes: total hardness, pH, TOC, exposure vessel type, size, lighting, temperature, vehicle concentration and dissolved oxygen. The chemical was tested above the calculated water solubility limit. The vehicle appears to be an emulsifier, which is inappropriate for aquatic toxicity testing; emulsions may exert physical toxicity and interfere with test substance concentration measurement. For more information on how to test difficult to test substances refer to the OECD Guidance Document on Aquatic Toxicity Testing of Difficult Substances and Mixtures.

Aquatic invertebrates. Information missing from the submitted acute daphnid test robust summary includes: a description of the dilution water to include source, temperature, TOC, dissolved oxygen, pH, test substance purity, hardness, alkalinity, total organic carbon, vehicle used, and number of organisms per concentration tested. The vehicle concentration was 800 times that allowed by OECD test guidelines. The chemical was tested above the calculated water solubility limit, and test duration was only 24 hours instead of the recommended 48 hours.

Followup Activity

EPA requests that the Sponsor submit adequate robust summaries and other modifications to its submission within 90 days.