1-13-94

MRID No. 416138-10

DATA EVALUATION RECORD

1. **CHEMICAL:** Benefin.
Shaughnessey No. 084301.

2. **TEST MATERIAL:** Benefin (Balan®) technical; N-butyl-N-ethyl-ααα-trifluoro-2,6-dinitro-p-toluidine; Lot No. 317EF2; 95.6% active ingredient.

3. **STUDY TYPE:** 123-1(a) Non-Target Plants: Seedling Emergence Phytotoxicity Test - Tier 2. Species Tested: Soybean, Sunflower, Cotton, Cabbage, Cucumber, Radish, Sorghum, Wheat, Corn, and Onion.

4. **CITATION:** Waldrep, T.W. 1989. Influence of Benefin Preemergence Spray on Seedling Emergence and Vegetative Vigor of Ten Crop Plants. Laboratory Report ID. No. 61989015. Conducted and submitted by DowElanco, Greenfield, IN. EPA MRID No. 416138-10.

5. **REVIEWED BY:**

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7. **CONCLUSIONS:** This study is not scientifically sound and does not meet the guideline requirements for a Tier 2 seedling emergence non-target plant phytotoxicity test. Multiple species were planted per tray, leading to competition between species.

8. **RECOMMENDATIONS:** If conducted again, species should be planted separately in containers that allow proper development of the test plants.

9. **BACKGROUND:**

10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A.

11. **MATERIALS AND METHODS:**

A. **Test Plants:** Dicotyledon plants were represented by six species from five families (i.e., soybean, sunflower, cotton, cabbage, cucumber, and radish). Monocotyledon plants were represented by four species from two families (i.e., corn, sorghum, wheat, and onion). Cultivars, lot number, and seed source were provided in the report.

B. **Test System:** Approximately 5,500 g of a 1:1 mixture of builders sand and sandy loam soil (resulting organic matter content of 1.0%) was sprayed with 10 ml of each test solution using a hand-held compressed air sprayer at 5 psi. The soil was then blended in a cement mixer for 5 minutes to distribute the test material uniformly. Five-thousand grams of this soil were placed in a plastic tray (31.5 x 21.5 x 8 cm). Trays were either seeded with corn, sorghum, onion, and wheat or cotton, cabbage, sunflower, cucumber, radish, and soybean. The seeds were covered with the remaining 500 g of soil.

After seeding, the trays were placed in a greenhouse with a 14-hour photoperiod and a temperature of 21-29°C. Trays were initially top-watered after planting and subirrigated thereafter.

C. **Dosage:** The highest test solution was prepared by diluting either 31.38 mg (for monocots) or 62.76 mg (for dicots) of the test material to 8 ml of 1:1 acetone:ethanol. To this, 72 ml of deionized water was added for a total of 80 ml of solution. Ten ml of this solution were sprayed per tray (160 gallons/A) for the highest test concentration. The remaining 40 ml of this solution were used to create the lower spray concentrations by serial dilution. The nominal rates were 1.0, 0.5, 0.25, 0.125, and 0.062 lb active ingredient (ai)/A for dicots and 0.5, 0.25, 0.125, 0.062, and 0.031 lb ai/A for monocots. Control trays were sprayed with a 10% acetone:ethanol solution.

D. **Design:** Each treatment/crop combination was replicated four times (i.e., 10 or 25 seeds/tray, 4 trays/ treatment or control). Seedling emergence was evaluated 7, 14, and 21 days after treatment (DAT).

The plants were rated for damage (using a 0-100 scale) at these same time periods. Twenty-one DAT, plant height was measured and above ground fresh weight per pot was assessed.

E. **Statistics:** Analysis of variance (ANOVA) and Dunnett's test were used to determine significant treatment differences on mean data or transformed (arcsine) data. If differences were observed, regression analysis was conducted to estimate EC_{25} values and 95% confidence intervals.

12. **REPORTED RESULTS:** Twenty-one days after treatment with benefin, seedling emergence was not significantly reduced from the untreated control for any of the ten plants tested. However, a 25% reduction was noted for cotton and cucumber at the 1.0 lb ai/A rate, for soybean at 1.0, 0.5, and 0.25 lb ai/A, and for sunflower at all rates.

The only injury symptom was stunting of growth. Based on 21 DAT observations, benefin at 0.25 lb ai/A caused slight injury to onion, wheat, and sorghum, and only slight injury was observed on corn, onion, and wheat at 0.5 lb ai/A. At 0.25 and 0.5 lb ai/A, sorghum was slightly and moderately injured, respectively. Corn, onion, sorghum, and wheat plants were not visually injured at 0.125 lb ai/A or lower rates of benefin. Cotton and sunflower were not injured at any treatment rate. At 1.0 lb ai/A, radish and soybean injury was slight and cabbage and cucumber damage was moderate. Cabbage, cotton, cucumber, radish, soybean, and sunflower plants were not injured with 0.25 lb ai/A or lower rates of benefin.

Benefin at the maximum tested rate did not reduce height of cotton, and height of radish, soybean, sunflower, and wheat plants was reduced slightly (less than 25%). The remaining plants demonstrated at least 25% damage at one or more treatment levels. The EC_{25} values, in order of increasing sensitivity, for the test species (in lb ai/A), are:

cucumber (0.941) < cabbage (0.703) < onion (0.599) < corn (0.575) < sorghum (0.255).

Onion, cotton, radish, and sunflower demonstrated no significant reductions in fresh weight at the maximum tested rate of benefin. However, a 25% reduction was still apparent for rates greater than 0.5 lb ai/A for cotton and radish, and 0.062 lb ai/A for sunflower. The remaining six species were significantly reduced and EC_{25} values were

species was soybean with a 12 and 7% reduction in emergence at the 0.5 and 1.0 lb ai/A rates, respectively.

Three-week injury ratings indicated that sorghum was the most sensitive species. However, NOEC values were not presented by the author. From the raw data, it appears that the NOEC would have been 0.062 lb ai/A.

Sorghum was the most sensitive plant tested and had the lowest EC_{25} with respect to plant height (0.255 lb ai/A). Corn and onion demonstrated responses that were less than 25%; therefore, the EC_{25} values derived are invalid.

Sorghum also was the most sensitive plant tested and had the lowest EC_{25} for plant weight (0.117 lb ai/A). Corn and soybean demonstrated responses that were less than 25%; therefore, the EC_{25} values derived are invalid. Sorghum demonstrated greater than 50% inhibition of fresh weight at some tested rate of benefin and the EC_{50} value could have been determined for this species (reviewer's EC_{50} was 0.293 lb ai/A).

In several cases, the author stated that there were no significant differences for a certain parameter between treatments and the controls. However, the rates used were not tested up to the maximum labeled rate [i.e., 3 lb ai/A based on the seed germination report (EPA MRID No. 416138-08)]. The test material should be applied at the maximum labeled rate in these cases to show that the NOEC lies above the maximum labeled rate.

This study is not scientifically sound and does not meet the guideline requirements for a Tier 2 seedling emergence non-target plant phytotoxicity test.

D. **Adequacy of the Study:**

(1) **Classification:** Invalid.

(2) **Rationale:** Improper design (i.e., multiple plants per tray) precluded valid results from being drawn from the data due to competition.

(3) **Repairability:** No.

15. **COMPLETION OF ONE-LINER:** N/A.