

US EPA ARCHIVE DOCUMENT

DATA EVALUATION RECORD

1. **CHEMICAL:** Prometon.
Shaughnessey No. 080804.

2. **TEST MATERIAL:** Prometon technical; 2,4-bis(isopropylamino)-6-methoxy-s-triazine; CAS No. 1610-18-0; 98.5% purity; a white powder.

3. **STUDY TYPE:** Non-Target Plants: Seed Germination Phytotoxicity Test - Tier 2. Species Tested: Soybean, Lettuce, Carrot, Tomato, Cucumber, Cabbage, Corn, Oat, Ryegrass, Onion.

4. **CITATION:** Chetram, R.S. 1990. Tier 2 Seed Germination Nontarget Phytotoxicity Study Using Prometon Technical. Laboratory Report No. LR90-04. Conducted by Pan-Agricultural Laboratories, Inc., Madera, CA. Submitted by Ciba-Giegy Corporation, Greensboro, NC. EPA MRID No. 417253-02.

5. **REVIEWED BY:**

Mark A. Mossler, M.S.
Agronomist
KBN Engineering and
Applied Sciences, Inc.

Signature: *Mark Mossler*
Date: 5/28/91

6. **APPROVED BY:**

Pim Kosalwat, Ph.D.
Senior Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature: P. Kosalwat
Date: 5/29/91

Henry T. Craven, M.S.
Supervisor, EEB/HED
USEPA

Cynthia Monte 12.3.91
Signature: *Henry T. Craven*
Date:

10 hrs

7. CONCLUSIONS:

This study is scientifically sound and meets the requirements for a Tier 2 seed germination test using non-target plants. Based on percent germination, all ten tested species were equally tolerant of prometon at the maximum rate of 30 lb ai/A. Due to this tolerance, no dose responses were exhibited and consequently, no EC values could be determined.

Three species (soybean, carrot, and oat) were equally tolerant to the maximum rate of 30 lb ai/A with respect to radicle length. The NOEC for lettuce and onion was 15 lb ai/A. Cabbage, tomato, and ryegrass had an NOEC value of 7.5 lb ai/A. The NOEC values for corn and cucumber (the most sensitive species) were 3.75 and 0.938 lb ai/A, respectively. The EC₂₅ values were able to be calculated for five of the ten tested species (Table 3, attached).

8. RECOMMENDATIONS: N/A.**9. BACKGROUND:****10. DISCUSSION OF INDIVIDUAL TESTS: N/A.****11. MATERIALS AND METHODS:**

A. **Test Plants:** Dicotyledon plants were represented by six species from six families (i.e., soybean, lettuce, carrot, tomato, cucumber, and cabbage). Monocotyledon plants were represented by four species from two families (i.e., corn, oat, ryegrass, and onion). Cultivars, seed sources, lot numbers, and germination ratings were provided in the report.

B. **Test System:** Two circles of blue blotter were placed in the bottom of a glass petri plate (100 mm in diameter and 15 mm in height). The test solutions were prepared with deionized water and acetone, and then diluted with water. Twelve milliliters of the test solution were added to each plate of soybean, cucumber, oat, and corn. Ten milliliters were added to plates of lettuce, carrot, tomato, cabbage, ryegrass, and onion. Ten seeds of each crop were added to each petri plate

after the test solution was absorbed into the paper. The plates containing crops with the same concentration were then randomly placed in plastic boxes (12.25 x 9.0 x 4.1 inches) with tightly fitting lids to prevent moisture loss. The petri plates were incubated in the dark at 25 ±1°C for 7 days, except lettuce, which was incubated at 20 ±1°C.

- C. **Dosage:** Prometon was applied at a rate of 30.0, 15.0, 7.5, 3.75, 1.88, 0.938, 0.469, and 0.0 lb ai/A to all plant species.
- D. **Design:** Each treatment/crop combination was replicated four times (i.e., 10 seeds/plate, 4 plates/ treatment). After 7 days of incubation, the seeds were removed from the petri plates and the radicle lengths were measured to the nearest millimeter. Percent seed germination and mean radicle length were calculated for all germinated seeds. Seeds were considered germinated if the radicle was at least 5 mm long.
- E. **Statistics:** All data were entered into a Lotus 1-2-3 spreadsheet. The spreadsheet calculated replicate and treatment means, standard deviations, and analysis of variance tables. Treatment means were used to calculate the percent effect resulting from the treatment. The percent effect was calculated using the following equation:

$$\% \text{ effect} = \frac{(\text{treatment mean} - \text{control mean})}{\text{control mean}} \times 100$$

An analysis of variance table was constructed using the Lotus 1-2-3 raw data spreadsheet. A one-way analysis of variance model for data with equal subsamples was used to analyze the data (percent germination and radicle length). Means were separated by using Duncan's New Multiple Range Test.

The percent effect values were input into a probit analysis program. The program ignored positive values and transformed the dose by natural logarithms. For seed germination, the probit was calculated using all data points; for all other parameters, the probit was calculated using replicate means.

12. **REPORTED RESULTS:**

Seed Germination: There was no significant ($p < 0.05$) difference between the control and any concentration for

soybean, lettuce, carrot, tomato, cucumber, cabbage, oat, ryegrass, corn, and onion. The no-effect concentration as defined by this study for each of the ten crops was 30.0 lb ai/A. Due to the lack of significant rate effects and a lack of a true dose response, a probit analysis was neither conducted nor EC values determined for all ten species.

Radicle Length: Soybean, carrot, and oat demonstrated no significant difference between the control and any of the rates tested. The NOEC value for these three species was the maximum rate of 30.0 lb ai/A. Lettuce and onion showed a significant difference between the control and the maximum concentration of 30.0 lb ai/A, therefore, the NOEC was reached at 15.0 lb ai/A. Cabbage, tomato, and ryegrass demonstrated a significant reduction in radicle length at the 15.0 lb ai/A rates, thus the NOEC value for these three species was 7.5 lb ai/A. Corn and cucumber showed significant reductions in radicle length at the 7.5 and 1.88 lb ai/A rates of prometon, respectively. The NOEC values for these two species were therefore 3.75 and 0.938 lb ai/A, respectively.

Due to a lack of significant rate effects and a lack of true dose response for soybean, lettuce, carrot, oat, and ryegrass, a probit analysis was neither conducted nor EC values determined. Tomato, cucumber, cabbage, corn, and onion did demonstrate rate responses, but only EC₂₅ values could be determined due to the magnitude of the rate effect. The EC₂₅ values (in lb ai/A) for these five species, in increasing sensitivity, are:

onion (24.146) < cabbage (17.134) < tomato (16.764) < corn (12.238) < cucumber (8.643).

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:**

"A no-effect concentration was achieved on both measurement parameters (germination and radicle length) for soybean, lettuce, carrot, tomato, cucumber, cabbage, oat, corn, ryegrass, and onion. EC₂₅ values were determined for radicle length of tomato, cucumber, cabbage, corn, and onion."

The Quality Assurance Unit of Pan-Agricultural Laboratories, Inc., was responsible for the assurance of compliance with Good Laboratory Practice (GLP) Standards. Statements of compliance to GLPs and QA were enclosed in the report.

14. REVIEWER'S DISCUSSION AND INTERPRETATION OF STUDY RESULTS:

- A. Test Procedure: The test procedures followed the SEP and Subdivision J guidelines.
- B. Statistical Analysis: Dunnett's pairwise comparison test and probit analysis was conducted on the data for cucumber radicle length (the most sensitive species and parameter). The results (attached) are in general agreement with the author's.
- C. Discussion/Results: This study meets the requirements for a Tier 2 seed germination study using non-target plants.

Seed Germination: All tested species were equally tolerant of the maximum rate of 30 lb ai/A of prometon. The NOEC value was therefore 30 lb ai/A for all test species. No EC values could be determined.

Radicle length: Three of the ten tested species (soybean, carrot, and oat) were equally tolerant of the maximum rate of prometon, with a subsequent NOEC value of 30 lb ai/A. The remaining seven species had NOEC values less than the maximum tested rate. The NOEC for lettuce and onion was 15 lb ai/A. Cabbage, tomato, and ryegrass had an NOEC value of 7.5 lb ai/A. The NOEC values for corn and cucumber (the most sensitive species) were 3.75 and 0.938 lb ai/A, respectively.

The EC₂₅ values were able to be computed for onion, cabbage, tomato, corn, and cucumber. These values were 24.15, 17.13, 16.76, 12.24, and 8.64 lb ai/A, respectively.

Page 21 of the report lists the NOEC values for tomato, cabbage, and ryegrass as 0.75 and corn as 0.375 lb ai/A. This is a typographical error. The two numbers should be 7.5 and 3.75 lb ai/A, respectively.

- D. Adequacy of the Study:
 - (1) Classification: Core.
 - (2) Rationale: N/A.
 - (3) Repairability: N/A.

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

Table 3

Statistical no-effect concentration (NOEC) (lb ai/A), EC₂₅ and EC₅₀ values (lb ai/A) for parameters measured during a nontarget plant study with Prometon Technical at 7 days after treatment.

Crop	Seed Germination			Radicle Lengths		
	NOEC ^y	EC ₂₅	EC ₅₀	NOEC	EC ₂₅	EC ₅₀
Soybean	30.0	ND ^z	ND	30.0	ND	ND
Lettuce	30.0	ND	ND	15.0	ND	ND
Carrot	30.0	ND	ND	30.0	ND	ND
Tomato	30.0	ND	ND	7.5	16.764	ND
Cucumber	30.0	ND	ND	0.938	8.643	ND
Cabbage	30.0	ND	ND	7.5	17.134	ND
Oat	30.0	ND	ND	30.0	ND	ND
Ryegrass	30.0	ND	ND	7.5	ND	ND
Corn	30.0	ND	ND	3.75	12.238	ND
Onion	30.0	ND	ND	15.0	24.146	ND

^y Highest treatment concentration which was not statistically different from the control (p < 0.05).

^z ND = Not determined. If a dose response was not evident or the highest treatment concentration tested did not result in a significant effect, EC₂₅ and EC₅₀ values could not be determined.

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cucumber radicle length

Summary Statistics and ANOVA

Transformation = None

Group	n	Mean	s.d.	cv%
1 = control	40	74.3500	12.2046	16.4
2* 0.49	40	64.6750	12.8470	19.9
3 0.938	40	71.1250	10.9525	15.4
4* 1.88	40	61.8250	9.2067	14.9
5* 3.75	40	59.5750	9.9946	16.8
6* 7.5	40	58.8000	8.9276	15.2
7* 15.0	40	53.8750	7.6634	14.2
8* 30.0	40	43.9750	8.5109	19.4

NOEC = 0.938 16 ai/A.

*) the mean for this group is significantly less than the control mean at alpha = 0.05 (1-sided) by Dunnett's test

Minimum detectable difference for Dunnett's test = -5.327920
This difference corresponds to -7.17 percent of control

Between groups sum of squares = 25696.250000 with 7 degrees of freedom.

Error mean square = 103.684455 with ** degrees of freedom.

Bartlett's test p-value for equality of variances = .014

cucumber radicle length

Estimated EC Values and Confidence Limits

Point	Conc.	95% Confidence Limits	
		Lower	Upper
EC 1.00	0.0669	0.0080	0.2093
EC 5.00	0.5130	0.1482	1.0239
EC10.00	1.5197	0.6817	2.4524
EC15.00	3.1626	1.8368	4.5966
EC50.00	70.0445	37.6261	210.7750
EC85.00	1551.3491	420.5777	17711.7305
EC90.00	3228.4966	739.6693	50862.7540
EC95.00	9563.0049	1704.6738	243154.2660
EC99.00	73301.0780	8136.4985	4587844.0000

$$y = 3.58 + 0.77(x)$$

$$y = \% \text{ probit inhibition}$$

$$x = \log(\text{rate})$$

$$EC_{25} = 9.42 \text{ } \mu\text{g ai/A.}$$

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