Relationship between Ethyl Methanesulphonate Concentration and Immersion Interval, and Germination and Seedling Vigor in Oats¹

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INTRODUCTION

The prime problem encountered when a mutagen is used on a plant species for the first time is to determine the appropriate dosage. This problem is further complicated because so many other factors, such as the moisture content of the treated seed $(1,3,5,12)^3$, characteristics of the atmosphere (3,5,12), seed covering (6), seed size (6), and genotype (1,12)influence the degree of mutagen effect upon the seed germination, M_1^4 seedling survival, uniformity and sterility, and frequency of chromosomal aberrations and mutations.

Some of the criteria used to assay the effects of the mutagen treatment upon seed are germination percentage and M_1 seedling height, relative to the untreated check. This paper reports data from experiments conducted to determine the effects of varying doses of ethyl methanesulphonate, a strong mutagen (10,11), upon the M_1 generation of hexaploid oats.

MATERIALS AND METHODS

Two experiments were conducted in the greenhouse at Ames, Iowa, in which primary oat seed of the variety Clintland 60 were treated with ethyl methanesulphonate⁵ at varying concentrations and immersion intervals. In experiment 1, 400 seed lots were immersed in water and in EMS solutions of 0.04-, 0.08-, and 0.12-mol concentrations for 2- and 24-hour intervals at room temperature. In experiment 2, two concentrations, 0.04 and 0.12 mol, and two immersion intervals, 2 and 4 hours, were used. After treatment the seed were rinsed with tapwater and immediately planted $\frac{1}{2}$ -inch deep in flats containing a soil mixture of loam, sand, and peat in the ratio of 2:1:1.

The experimental design was a randomized block with four replications

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³ Italic numbers in parentheses refer to Literature Cited, pp. 310-1.

 $^{4}M_{1}$ refers to the plants grown from seed treated with a mutagen.

⁶ Ethyl methanesulphonate will be abbreviated herein to EMS.

for experiment 1 and two replications for experiment 2. A plot consisted of 100 seed planted in a row across a flat (22 inches). The plots were spaced 2 inches apart so that each flat contained eight plots. The experiments were conducted in the greenhouse. Germination percentages and mean seedling heights were recorded on a plot basis 14 days after planting.

EXPERIMENTAL RESULTS

The mean germination percentages and M_1 seedling heights for oatseed lots in experiment 1 are presented in table 1. Soaking the seed for 2 hours in 0.04-, 0.08-, and 0.12-mol EMS solutions did not reduce germination significantly, but there was a tendency toward lower germination with an increased concentration of EMS. The 24-hour immersion interval killed

TABLE 1.—Mean germination percentages and M_1 seedling heights from seed lots of Clintland 60 oat variety after treatment with 3 concentrations of EMS, using 2 immersion intervals in experiment 1

Treatment	Germination ¹	Seedling height
	Percent	Mm.
Check	95ª	27ª
0.04 mol for 2 hours	96ª	25 ^b
.08 mol for 2 hours	93ª	24 ^b
.12 mol for 2 hours	86ª	22°
.04 mol for 24 hours	33 ^b	17 ^d

¹ Means with the same letter superscript belong to the same significance group according to Duncan's multiple-range test (14).

all seed soaked in 0.08- and 0.12-mol solutions of EMS, and seed germination was reduced to 33 percent by 0.04-mol level.

Each of the EMS solutions caused a significant reduction in M_1 seedling height. The 2-hour immersion of oat seed in the 0.04- and 0.08-mol solutions caused a 10-percent reduction in seedling height. A similar treatment in the 0.12-mol solution caused a 20-percent reduction, and a 24-hour immersion in the 0.04-mol solution caused a 40-percent reduction in M_1 seedling height.

In experiment 2 (table 2) each of the treatment combinations, with the exception of the 2-hour immersion in the 0.04-mol EMS solution reduced the germination percentage significantly, and all of the treatments reduced seedling height by 25 and 30 percent, respectively.

The reductions in germination percentage and seedling height from the treatment of oat seed with 0.12-mol EMS solution for 4 hours were similar to those resulting from the thermal-neutron treatment applied in some mutation breeding studies $(1.4-2.0 \times 10^{13} \text{ neutrons per square centimeter}).^{6}$

⁶ Unpublished data from Iowa Agriculture and Home Economics Experiment Station.

SUMMARY

Seed of Clintland 60 variety of oats were immersed for 2- and 24-hour intervals in water and in EMS solutions of 0.04-, 0.08-, and 0.12-mol strength, and for 2- and 4-hour intervals in 0.04- and 0.12-mol solutions. None of the EMS concentrations reduced germination percentages or seedling heights markedly when the immersion interval was 2 hours. However, the 24-hour immersion was lethal, with the 0.08- and 0.12-mol solutions, and reduced germination percentage to 33 with the 0.04-mol solution. Reduction in germination percentage and seedling height after treatment with 0.12-mol EMS solution for 4 hours closely approximated the M₁ damage for treating oat seed with $1.4-2.0 \times 10^{13}$ neutrons per square centimeter.

TABLE 2.—Mean germination percentages and seedling heights from seed lots of Clintland 60 oat variety after treatment with 2 concentrations of EMS, using 2 immersion intervals in experiment 2

Treatment	Germination ¹	Seedling height
	Percent	Mm.
Check	99ª	20ª
0.04 mol for 2 hours	91ª	18 ^b
.04 mol for 4 hours	86 ^b	18 ^b
.12 mol for 2 hours	85 ^b	17 ^b
.12 mol for 4 hours	75°	14°

¹ Means with the same letter superscript belong to the same significance group according to Duncan's multiple-range test (14).

RESUMEN

Semillas de la variedad de avena Clintland 60 se sumergieron por un período de 2 y 24 horas en soluciones de EMS a concentraciones de 0.04, 0.08 y 0.12 mol y también durante un período de 2 y 4 horas en soluciones de 0.04 y 0.12 mol. Ninguna de las concentraciones de EMS redujo marcadamente los porcentajes de germinación, ni el tamaño de las plántulas cuando el período de inmersión de la semilla fue de 2 horas. Sin embargo, una inmersión por 24 horas fue letal a las semillas en las concentraciones de 0.08 y 0.12 mol y el porcentaje de germinación se redujo un 33 con la solución 0.04 mol. La reducción en el porcentaje de germinación y altura de las plántulas, después de tratarse con una solución de EMS a una concentración de 0.12-mol por 4 horas, se aproxima bastante al daño causado en la M_1 de semillas de avena sometidas a una dosis de 1.4–2.0 × 10¹³ neutrones por centímetro cuadrado.

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