GENETIC VARIABILITY FOR QUANTITATIVE CHARACTERS
AMONG MUTAGEN-DERIVED BARLEY POPULATIONS*

A. GHAFOOR ARAIN** AND K. W. SHEPHERD

Department of Agronomy, Waite Agricultural Research Institute, Glen Osmond, South Australia.

Abstract

Seeds of five barley cultivars (Clipper, C.I. 3576, Proctor, Ketch and Prior) having constrasting yields and adaptation characteristics were treated with 0.04M ethyl methanesulphonate (EMS) for 8 hours and populations of M₂-derived treated and control lines were assessed in replicated field trials over four different sites in South Australia in the M₄ generation.

The treated populations of each of five cultivars gave significantly reduced mean yields over all sites and exhibited significant increases in genotypic variance, compared with the respective control populations. Similarly, the genotypic variances for heading date, plant height and seed weight were significantly increased in the treated populations. The mean heading date of treated lines of each cultivar was shifted towards lateness whereas the mean values for height and seed weight in the treated lines were not significantly altered except for seed weight of Prior which was significantly reduced.

Introduction

During the last five decades adequate research has been conducted to prove that ionizing radiations and chemicals, when applied to plant and animal tissues, can induce mutations at loci that govern that expression of both qualitatively and quantitatively inherited characters (Muller, 1927; Stadler, 1928a, b; Auerbach, 1943; Gustafsson, 1947, 1951). The first extensive study involving induction of mutations, affecting quantitative characters was conducted by Gregory (1955) in peanuts (Arachis hypogaea). Since then, many other workers (Oka et al., 1958; Rawlings et al., 1958; Brock & Latter, 1961; Krull & Frey, 1961; Abrams & Frey, 1964; Brock, 1965; Miah & Yamaguchi, 1965; Gaul et al. 1966; Ghafoor et al. 1968; Gaul et al. 1969) have used different mutagens on a number of crop plants to induce genetic variability in quantitative characters. These studies showed that mutagens were effective in generating variability in quantitative characters and progress could be made from selecting among the variants.

The genetic variability induced among the mutagen-derived populations has been measured only in one environment in most of these studies. However, Gaul et al. (1969) have tested the M₂-derived families at two locations in successive generations and their study revealed that genotypic variance for yield was inconsistent between generations and between locations within generations.

*Part of the graduate research carried out at the University of Adelaide, South Australia.
**Present address: Plant Genetics Division, Atomic Energy Agricultural Research Centre, Tandojam, Pakistan.
The present paper reports the magnitude of genotypic variance among different quantitative characters in EMS-treated material of diverse barley cultivars when grown over different sites in M₄ generation.

**Materials and Methods**

Five barley cultivars viz. Clipper, C. I. 3576, Proctor, Ketch and Prior with diverse yield and adaptation patterns (Ara in, 1974) were selected for treatment with chemical mutagen. The mutagenic treatment consisted of soaking 200 seeds of each cultivar in 200 ml of freshly prepared unbuffered aqueous solution of EMS (0.04 M) for 8 hr. at 23°C.

The main spikes were taken from 65 randomly chosen treated M₁ plants and 65 control M₁ plants from each cultivar and the seeds from each spike were sown in 25.5 cm diameter pots in the glass house in January 1969 to raise M₂ plants. In M₂ generation, plants having drastic changes were removed and the remainder were thinned at random to give 5 plants per pot. One normal-appearing plant per pot was selected and harvested at maturity as M₃ seed. In July, 1969, the M₃ seeds from each of the 65 randomly selected M₂ plants were sown in the field in single rows (3.05 m) with 36 cm space in between rows in order to obtain sufficient M₄ seed. Thirty five treated and 25 control lines from each of the four cultivars (Clipper, C. I. 3576, Proctor and Ketch) and 25 treated and 15 control lines of Prior with at least 200 gm of seed were selected and grown in a two-replicate randomized layout at each of the four contrasting sites (Bundaleer, Roseworthy, Minlaton & Waite) in the cereal growing areas of South Australia during 1970.

The plots were sown with a magazine-loaded cone seeder using 20 gm per plot (67 Kg/ha). The individual plots consisted of four rows each 4m in length and 15 cm apart with 30 cm space between adjacent plots and 1 m wide pathways between bays of plots.

The data for yield and other quantitative characters were recorded for each plot at each site as under:

*Heading date:* The heading date was scored on a whole plot basis when approximately 50% of tillers in a plot showed at least 2.5 cm awn emergence from the boot. Heading was recorded as the number of days from the date of planting at each site.

*Plant height (cm):* The mean height of plants in each plot was recorded as the distance from the ground surface to the tips of spikes excluding awns, averaged over 3 plants selected at random within the plot.

*Yield:* When the plants were mature, 30 cm at each end of the plot was removed and the remainder of the plot (3.40 m) was harvested with a “Waite Gravelly Harvester”. The weight of clean grain was recorded in gm/plot.

*Seed weight (gm/100 seed):* A 15 gm sample of grain was taken from each plot and the number of seeds was counted with an electronic seed counter to give 1000 grain weights.

The data for each character were analyzed by analysis of variance combined over all sites for treated and control lines of each cultivar separately. The estimates of genotypic variances due to lines (σ²p) were calculated for both the populations of
GENETIC VARIABILITY IN BARLY

...each cultivar from the mean squares in the analysis of variance similar to Johnson et al. (1955) as under:

\[ MSP - MSPS = \frac{\sigma^2_p}{rs} \]

Where,

- MSP = mean square expectations due to lines
- MSPS = mean square expectations due to lines x sites interaction

and s, r represent the number of sites and replicates per site respectively.

The significant differences between treated and control population means were obtained by using 't' test as under:

\[ t = \frac{\bar{x}_1 - \bar{x}_2}{\sqrt{s^2\left(\frac{1}{n_1} + \frac{1}{n_2}\right)}} \]

Where

- \( \bar{x}_1 \) = mean value of control population
- \( \bar{x}_2 \) = mean value of treated population

and, \( s^2 = \frac{TSS_1 + TSS_2}{TDF_1 + TDF_2} \)

Where

- TSS_1 = total sum of squares of control population
- TSS_2 = total sum of squares of treated population

and, TDF_1 and TDF_2 are total degrees of freedom of control and treated populations respectively.

**Results**

The frequency distributions for yield, heading date, plant height, and seed weight combined over all sites of M_2 derived treated and control lines of each cultivar shown in Figures 1, 2, 3, and 4 respectively, whereas the mean values and genotypic variances (\( \sigma^2_p \)) for these characters are presented in Table 1.

(a) *Yield (gm/plot):*

A characteristic of the frequency distribution curves is a pronounced shift of the EMS-treated lines away from their respective controls towards lower yield in
the M₄ generation (Fig. 1). The mean yields of the treated lines of each cultivar were significantly less than the means of corresponding control populations in all cases (Table 1). The greater reduction in the mean yield was observed in the treated material of Proctor and Prior. Even though the overall mean yield was reduced, some of the treated lines in Clipper performed identical to that of the highest yielding control lines and with C.I. 3576 three of the treated lines out-yielded the highest yielding control line (Fig. 1).

The magnitude of αp induced by EMS-treatment among the treated material of each cultivar was several-fold greater than corresponding controls except with Ketch, where it showed only a two-fold increase (Table 1). The largest αp was obtained with the treated lines of C.I. 3576 followed in order by Prior, Proctor, Clipper and Ketch treated lines.

(b) Heading date (days):

The control lines of each cultivar exhibited uniform heading over a 2-4 day interval except one control line of Clipper (No 23C) and Proctor (No. 10 C) were 2
### TABLE 1. Mean values and genotypic variances ($\sigma^2p$) for yield, heading date, height and seed weight of $M_2$—derived treated and control lines of the five barley cultivars grown over four sites in $M_4$ generation.

<table>
<thead>
<tr>
<th>Cultivar</th>
<th>Lines</th>
<th>Yield (gm/plot)</th>
<th>Mean value</th>
<th>$\sigma^2p$</th>
<th>Head date (days)</th>
<th>Mean value</th>
<th>$\sigma^2p$</th>
<th>Plant height (cm)</th>
<th>Mean value</th>
<th>$\sigma^2p$</th>
<th>Seed weight (gm/1000 seeds)</th>
<th>Mean value</th>
<th>$\sigma^2p$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clipper</td>
<td>Control</td>
<td>611.3</td>
<td>115.1</td>
<td>1.03</td>
<td>84.2</td>
<td>7.22</td>
<td>0.21</td>
<td>40.2</td>
<td>0.72</td>
<td>39.7NS</td>
<td>40.2</td>
<td>0.59</td>
<td>40.2</td>
</tr>
<tr>
<td></td>
<td>Treated</td>
<td>553.8**</td>
<td>116.2**</td>
<td>2.35</td>
<td>85.6NS</td>
<td>5.23</td>
<td>39.7NS</td>
<td>40.2</td>
<td>0.21</td>
<td>39.7NS</td>
<td>40.2</td>
<td>0.72</td>
<td>40.2</td>
</tr>
<tr>
<td>C.I. 3576</td>
<td>Control</td>
<td>677.5</td>
<td>108.3</td>
<td>0.02</td>
<td>84.6</td>
<td>0.34</td>
<td>0.21</td>
<td>44.9</td>
<td>3.21</td>
<td>44.5NS</td>
<td>44.9</td>
<td>0.21</td>
<td>44.9</td>
</tr>
<tr>
<td></td>
<td>Treated</td>
<td>607.8**</td>
<td>109.6**</td>
<td>5.27</td>
<td>83.7NS</td>
<td>11.35</td>
<td>44.5NS</td>
<td>44.9</td>
<td>3.21</td>
<td>44.5NS</td>
<td>44.9</td>
<td>0.21</td>
<td>44.9</td>
</tr>
<tr>
<td>Proctor</td>
<td>Control</td>
<td>603.3</td>
<td>126.4</td>
<td>0.76</td>
<td>80.7</td>
<td>0.0</td>
<td>35.1</td>
<td>0.16</td>
<td>1.16</td>
<td>35.5NS</td>
<td>35.1</td>
<td>0.16</td>
<td>35.1</td>
</tr>
<tr>
<td></td>
<td>Treated</td>
<td>515.3**</td>
<td>127.4**</td>
<td>2.47</td>
<td>80.6NS</td>
<td>2.94</td>
<td>35.5NS</td>
<td>35.1</td>
<td>0.16</td>
<td>35.5NS</td>
<td>35.1</td>
<td>0.16</td>
<td>35.1</td>
</tr>
<tr>
<td>Ketch</td>
<td>Control</td>
<td>536.0</td>
<td>97.6</td>
<td>0.14</td>
<td>87.8</td>
<td>0.06</td>
<td>41.4</td>
<td>0.07</td>
<td>0.54</td>
<td>41.2NS</td>
<td>41.4</td>
<td>0.07</td>
<td>41.4</td>
</tr>
<tr>
<td></td>
<td>Treated</td>
<td>499.3*</td>
<td>98.4*</td>
<td>1.58</td>
<td>87.5</td>
<td>1.48</td>
<td>41.2NS</td>
<td>41.4</td>
<td>0.54</td>
<td>41.2NS</td>
<td>41.4</td>
<td>0.07</td>
<td>41.4</td>
</tr>
<tr>
<td>Prior</td>
<td>Control</td>
<td>495.0</td>
<td>113.4</td>
<td>0.08</td>
<td>102.1</td>
<td>0.70</td>
<td>41.9</td>
<td>0.0</td>
<td>0.0</td>
<td>40.7**</td>
<td>41.9</td>
<td>0.0</td>
<td>40.7**</td>
</tr>
<tr>
<td></td>
<td>Treated</td>
<td>435.3**</td>
<td>114.2*</td>
<td>4.50</td>
<td>99.5NS</td>
<td>5.21</td>
<td>40.7**</td>
<td>41.9</td>
<td>0.0</td>
<td>40.7**</td>
<td>41.9</td>
<td>0.0</td>
<td>40.7**</td>
</tr>
</tbody>
</table>

*Mean value significantly different at 5% probability.

**Mean value significantly different at 1% probability.

NS = Non-significant.
Fig. 2. Frequency distribution for heading date combined over sites of M2-derived treated and control lines of five barley cultivars grown in M4 generation.
days later than the rest of their respective control lines (Fig. 2). There was an overall shift towards lateness among the treated lines of all cultivars as shown by the frequency distributions (Fig. 2) and significant changes in mean (Table 1). Nevertheless at least one of the treated lines was earlier than the earliest control line in all cultivars except Clipper.

The genotypic variance for heading date was negligible among the control lines of all cultivars except with Clipper and Proctor where the estimates were increased by the presence of one late line in each cultivar. However, the EMS-treatment resulted in a considerable increase of genotypic variance ($\sigma^2P$) among all five cultivars as compared with respective controls with the largest increase occurring with C.I. 3576 and Prior (Table 1).

(c) Plant height (cm):

The mean heights of the control lines were reasonably homogeneous except one line of Clipper (No. 12C) which was clearly much taller than the rest of the controls (Fig. 3). As with heading date the distribution of height among the treated lines were spread over a wide range but there was no overall trend to either increased or decreased height except with Prior, where a general decrease in height of treated lines was observed, but this was not significant (Table 1).

The only substantial genotypic variance ($\sigma^2P$) for height among the control groups was observed with Clipper and apparently this was due mainly to line No.
12C which was 12 cm taller than the mean of the control group. The genotypic variance of the treated lines of the other cultivars greatly exceeded that of respective controls and the most pronounced difference occurred with the C.I. 3576 lines (Table 1).

(d) **Seed Weight (gm/1000 seeds):**

The frequency distributions for seed weight of all cultivars (Fig. 4) show an increase in spread of this character for the treated lines over their respective controls but no consistent trend was observed in all the cultivars. There was an overall trend towards decrease in seed weight among the treated Clipper and Prior lines, whereas spread of seed weight occurred in both directions with C.I. 3576, Proctor and Ketch cultivars. The mean value of seed weight was not significantly altered by EMS-treatment in any of the cultivars except Prior (Table 1).

The genotypic variances for seed weight were considerably increased in the treated lines of all cultivars compared with the corresponding control, indicating a substantially greater variability for this character among the treated material (Table 1).
Discussion

In common with other workers it was found that EMS-treatment of barley cultivars caused a reduction in mean yield and a large increase in genotypic variance for yield and other quantitative characters. The maximum reduction in mean yield in M4 generation occurred among the treated lines of Proctor, followed in order by those of Prior, C.I. 3576 Clipper and Ketch (Table 1). These results are closely paralleled by the response with respect to M2 chlorophyll mutation frequency and M4 seed sterility except in these cases Clipper was less responsive than Ketch (Arain, 1974). On the other hand, the magnitude of induced genotypic variance in M4 did not follow the same trend except that the least amount occurred in Ketch (Table 1). Gaul (1965) has observed that M3 survival, chlorophyll mutation frequency, reduction of yield mean and increased genotypic variance are closely correlated phenomena and he has used these joint responses to gauge the effectiveness of the mutagen treatment. Therefore, it can be concluded that in the present study the EMS treatment was most effective with Proctor and Prior and least effective with Ketch and Clipper. The reasons for these differences are not known but it is possible that the cultivars differ in their seed coat permeability or in the physiological state of the embryo.

The other characters studied, namely heading date, height and seed weight all showed an increase in genotypic variances among the treated populations in M4 generation except for height of Clipper, where genotypic variance of the control lines was much inflated by the presence of one tall line (No. 12C).

The mean heading date of treated lines in all cultivars was shifted towards lateness. This delay in heading has often been observed in cereals after irradiation and EMS-treatments (Abrams & Frey, 1964; Krull & Frey, 1961; Gaul et al., 1966). In the present study, the shift of mean heading date towards lateness does not fit Brock’s (1965) hypothesis since these shifts occurred in EMS treated lines of all 5 cultivars including early, medium-late and late types. On the other hand, it provides support for the hypothesis of Gaul & Aastveit (1966) which states that the change in mean values of quantitative characters occurs in the direction associated with reduced vitality independent of the genotype used. In particular, many of the treated lines of Proctor (which is very late in heading), were 2 days later than the mean of controls and each of these lines had low yield, indicative of reduced vitality.

The mean values for plant height and seed weight in the treated populations were not significantly altered from that of controls except with Prior, where seed weight was significantly reduced. The non-significant shifts in mean values of these characters is in agreement with the findings of Oka et al. (1958) in rice and Rawalings et al. (1958) in soybeans. In the present study, mutations for plant height occurred in both plus and minus directions resulting in no significant shift in the mean value, with all cultivars except Prior. With Prior (the tallest cultivar studied) the treated lines were shifted only towards shortness but the reduction in the mean height was not significant (Table 1). The pattern of seed weight changes was similar to that observed for height for all cultivars except Clipper and Prior. The changes in these two cultivars were towards reduced seed weight, particularly with Prior where the shift in mean was significant and many treated lines had much reduced seed weight. No obvious reason could be found for this different behaviour of Prior.

The present studies have provided an evidence on the induction of genetic variability connected with yield and other quantitative characters when tested over a range of sites among diverse barley cultivars after EMS-treatment. The genetic variability thus induced can effectively be utilized for evolving new strains possessing desirable characters.
References


